Official Title: A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector‑Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII levels ≤1 IU/dL and Pre-existing Antibodies Against AAV5

NCT Number: NCT03520712

Applicant/MAH: BioMarin Pharmaceutical Inc.

Version Date: 07 July 2022

16 APPENDICES

16.1 Study Information

16.1.1 Protocol and Protocol Amendments

This appendix includes the original 270-203 study protocol and protocol amendments 1-4, France-specific protocol amendments 2-3, and Tawain-specific protocol amendment 4 (no participants were enrolled in France or Taiwan):

Original Protocol (dated 29 September 2017)

Protocol Amendment 1 (dated 5 October 2018)

Protocol Amendment 2 (dated 4 October 2019)

Protocol Amendment 2 France-Specific (dated 01 November 2019)

Protocol Amendment 3 France-Specific (dated 21 August 2020)

Protocol Amendment 3 (dated 24 August 2020)

Protocol Amendment 4 (dated 04 August 2021)

Protocol Amendment 4 Taiwan-Specific (dated 07 July 2022)

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CLINICAL STUDY PROTOCOL

Study Title:	A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5
Protocol Number:	270-203
Active Investigational Product:	AAV5-hFVIII-SQ
European Union Drug Regulating Authorities Clinical Trials (EudraCT) Number:	2017-000662-29
Indication:	Hemophilia A
Sponsor:	BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949
Development Phase:	Phase 1/2
Sponsor's Responsible Medical Monitor:	PI MD 105 Digital Drive Novato, CA 94949 U.S.A.
Study Design:	Single-arm, open-label
Duration of Subject Participation:	Up to 264 weeks
Dose:	6E13 vg/kg as single infusion
Study Population:	Males ≥ 18 years of age with severe hemophilia A and detectable pre-existing antibodies against AAV5 vector capsid
Date of Original Protocol:	29 September 2017

2 SYNOPSIS

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	**SUMMARY TABLE** Referring to Part of the Dossier: Volume: Page: Reference:	**FOR NATIONAL AUTHORITY USE ONLY:**

TITLE OF STUDY:

A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5

PROTOCOL NUMBER:

270-203

STUDY SITES:

Approximately 2-3 sites in the United Kingdom

PHASE OF DEVELOPMENT:

Phase 1/2

STUDY RATIONALE:

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. The clinical phenotype of HA patients generally correlates tightly with the level of residual expression. Severe HA is classified as FVIII activity less than 1% of wild-type (< 1 IU/dL), moderate disease comprises 1-5% of wild-type activity, and the mild form is 5-40% activity. The clinical manifestations of severe HA are frequent spontaneous bleeding episodes, predominantly in joints and soft tissues, with a substantially increased risk of death from hemorrhage when the brain is involved.

Treatment of severe HA presently consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study (Berntorp, 2017) and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with on-demand-only therapy in the same studies, respectively. Continued repeated bleeding events compound

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death.

Chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) and bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half-life to maximum of 18 hours, leaving critical periods when FVIII activity levels are below the therapeutic range and leaving patients vulnerable to bleeding and concomitant sequelae. As such, despite currently available FVIII replacement therapies, a high unmet need remains for maintenance of a sustained, consistent FVIII level compatible with a normal and hemorrhage-free life.

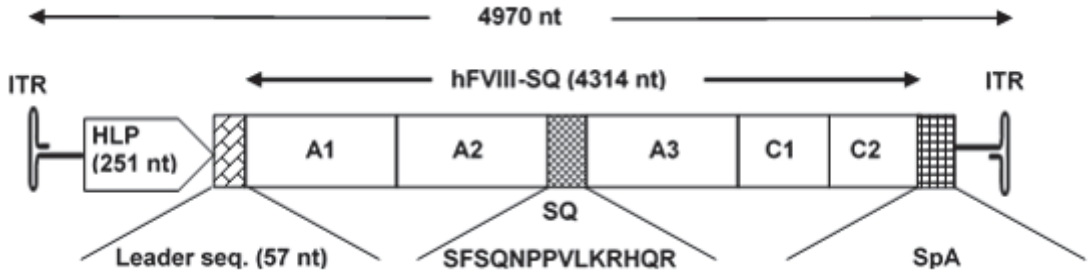
Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of active FVIII following a single intravenous infusion of a vector encoding the appropriate gene sequence. Hemophilia A is well-suited for a gene replacement approach because clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in minute amounts (200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and even modest increases in the level of FVIII (any increase of the plasma level by 2 ng/ml induces an increase in activity of 1%) can ameliorate the severe form of hemophilia A. Thus, relatively small changes in endogenous FVIII activity can result in clinically relevant improvements in disease phenotype. Moreover, the circulating FVIII response to gene transduction can be assessed using validated quantitative endpoints that are easily assayed using established laboratory techniques.

Several different gene transfer strategies for FVIII replacement have been evaluated, with adeno-associated viral (AAV) vectors used in most hemophilia clinical trials. AAV vectors have an excellent and well-defined safety profile, and can direct long-term transgene expression with tropism and promoter specificity for specific tissues, such as the liver (eg serotypes 2, 5, and 8). Indeed, an ongoing gene therapy clinical trial for a related disorder, hemophilia B, has established over 7 years of stable human factor IX (hFIX) expression at levels sufficient for conversion of their bleeding phenotype from severe to moderate or mild, following a single peripheral vein infusion of AAV8-hFIX vector (Nathwani, 2014). Several participants in this trial have been able to discontinue factor prophylaxis without suffering spontaneous hemorrhages, even when they undertook activities that previously resulted in bleeding. Thus, gene therapy treatment has resulted in a substantial improvement in their quality of life.

BMN 270 is an AAV5-based gene therapy vector that expresses the SQ form of hFVIII under the control of a hybrid human liver-specific promoter (Figure 1).

NAME OF COMPANY	SUMMARY TABLE	FOR NATIONAL AUTHORITY USE ONLY:
BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	Referring to Part of the Dossier: Volume: Page: Reference:	

Figure 1. hFVIII-SQ Vector Genome Schematic



Legend –Note that schematic is not to scale; nt = nucleotides

BMN 270 is delivered by a single IV dose and is designed to achieve stable, potentially life-long expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.

BMN 270 is currently being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. Preliminary results from 270-201 have demonstrated that following gene transfer, sustained FVIII activity above 5% (5 IU/dL) up to a year's observation is achievable at doses of 4-6E13 vg/kg with an acceptable safety profile (Pasi, 2016). The majority of subjects achieved FVIII levels within the normal range for FVIII, with few to no bleeding episodes and discontinuation of FVIII prophylaxis.

Subjects enrolled and infused in 270-201 were screened and negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	**SUMMARY TABLE** Referring to Part of the Dossier: Volume: Page: Reference:	**FOR NATIONAL AUTHORITY USE ONLY:**

Current data describing the extent of neutralization or inhibited transduction has been inconsistent across studies, each employing one of several varying methodologies to evaluate both vector-specific antibodies and transduction inhibition (TI). Early studies using AAV2 for the treatment of hemophilia B described inhibited transduction in patients with neutralizing antibody titers of 2 or 3 (Manno, 2006), but more recent studies have described successful transduction in patients who, in retrospect, were determined to have pre-existing antibody to the vector capsid (Majowicz, 2017). Importantly, no drug-related adverse events were described in these patients. As the sensitivity of assays used to detect pre-existing antibody has increased greatly in recent years, the biological relevance of any given titer determination, and comparisons across studies using assays with differing titer sensitivities, requires additional study.

Data from a non-clinical study conducted by BioMarin in cynomolgus monkeys further demonstrated that transduction efficiency can vary greatly in the presence of pre-existing anti-AAV5 antibody. In non-human primates, the ability to attain FVIII levels in the face of measurable AAV5 antibody titers without significant safety risks supports the possibility of achieving a similar response in patients with AAV5 antibody positivity. The totality of available data shows the effect of pre-existing immunogenicity to AVV delivery vectors is not completely understood, and indicates that some patients may benefit from AAV5 gene therapy who are currently excluded.

In this study, patients with severe HA and pre-existing antibodies against AAV5 will be enrolled to evaluate the clinical response to BMN 270 gene therapy. The aim is to evaluate the safety, tolerability, and efficacy of BMN 270 in patients with severe HA and pre-existing antibodies against AAV5 vector capsid at various levels of AAV5 antibody titers.

OBJECTIVES:

The primary objective of the study is to:

- Assess the safety of a single intravenous administration of BMN 270 in severe HA subjects with pre-existing antibody to AAV5 vector capsid, including development of FVIII neutralizing antibody

Secondary objectives of the study are to:

- Assess the efficacy of BMN 270 defined as FVIII activity at or above 5 IU/dL at Week 26
- Assess the impact of BMN 270 on usage of exogenous FVIII replacement therapy

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	**SUMMARY TABLE** Referring to Part of the Dossier: Volume: Page: Reference:	**FOR NATIONAL AUTHORITY USE ONLY:**

- Assess the impact of BMN 270 on the number of bleeding episodes requiring exogenous FVIII therapy
- Evaluate the pharmacodynamics of FVIII expression following IV infusion of BMN 270
- Assess the impact of BMN 270 on patient-reported outcomes (PROs)

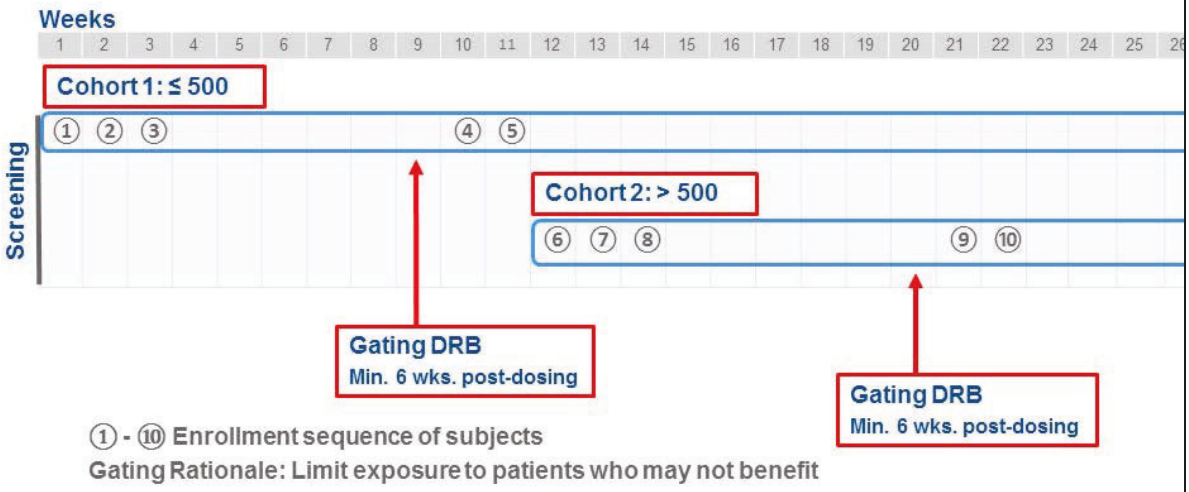
NAME OF COMPANY	SUMMARY TABLE	FOR NATIONAL AUTHORITY USE ONLY:
BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	Referring to Part of the Dossier: Volume: Page: Reference:	

STUDY DESIGN AND PLAN:

This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to the AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects will be enrolled at 2-3 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb titer ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb titer > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date.

The Data Review Board (DRB) will consist of the 2 Principal Investigators and Sponsor's Medical Monitor, as well as hemophilia expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201. The DRB will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 2 represents one dosing schedule scenario):

Figure 2: 270-203 Dosing Schedule (One Possible Scenario)



NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	**SUMMARY TABLE** Referring to Part of the Dossier: Volume: Page: Reference:	**FOR NATIONAL AUTHORITY USE ONLY:**

Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from a minimum of 6 weeks post-infusion) reviewed. FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response, based on data from 270-201. Up to 6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness).

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DRB evaluation of safety and efficacy in treated subjects, with the following triggers that may potentially pause further enrollment:

- any related SAE;
- any related AE with a severity > CTCAE Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

Following a temporary halt of enrolment, the DRB may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed and additional blood samples will be collected (tryptase, C3, C3a, C4, C5, and C5a within 1 hour of hypersensitivity reaction, and IgE between 8-24 hours after the reaction). Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	**SUMMARY TABLE** Referring to Part of the Dossier: Volume: Page: Reference:	**FOR NATIONAL AUTHORITY USE ONLY:**

Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DRB based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL. Throughout the study, subjects with FVIII activity below 5 IU/dL may be monitored more frequently at the discretion of the Investigator and the Medical Monitor. In subjects who show an initial response to BMN 270 but who later have FVIII activity decline to < 5 IU/dL, the investigator and medical monitor will review the subject's FVIII activity levels and discuss whether to resume prior FVIII prophylaxis. In addition, the investigator will notify the subject of his FVIII activity levels and will discuss with the subject the risk of bleeding and when (and if) prior FVIII prophylaxis will be resumed.

The study analysis will be performed after all subjects have been followed for 26 weeks post-BMN 270 infusion, with presentation of safety, efficacy, and FVIII PD assessments. After the safety, efficacy, and FVIII PD analyses at 26 weeks post-BMN 270 infusion, long-term safety and efficacy will be assessed in all subjects for up to a total of 5 years post-infusion. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the DRB.

NUMBER OF SUBJECTS PLANNED:

Approximately 10 subjects

DIAGNOSIS AND ALL CRITERIA FOR INCLUSION AND EXCLUSION:

Individuals eligible to participate in this study must meet all of the following criteria:

1. Males ≥ 18 years of age with hemophilia A and residual FVIII levels ≤ 1 IU/dL as evidenced by medical history, at the time of signing the informed consent.
2. Detectable pre-existing antibodies against the AAV5 vector capsid as measured by AAV5 total antibody ELISA
3. Treated/exposed to FVIII concentrates or cryoprecipitate for a minimum of 150 exposure days (EDs)
4. Subject must have been on prophylactic FVIII replacement therapy for at least 12 months prior to study entry.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures.
6. No history of FVIII inhibitor, and results from a Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) on 2 consecutive occasions (the most recent one of which should be tested at the central laboratory) at least one week apart within the past 12 months
7. HIV positive patients may be enrolled, only if the patient has a CD4 count > 200/mm^3 and an undetectable viral load.
8. Sexually active participants must agree to use an acceptable method of double barrier contraception for at least 6 months post-infusion, which may include hormonal contraception for a female partner. After 6 months, subjects may stop contraception use only if they have had 3 consecutive semen samples with no detectable viral vector DNA.
9. Willing to abstain from consumption of alcohol for at least the first 26 weeks following BMN 270 infusion.

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Any evidence of active infection or any immunosuppressive disorder, except for HIV infection as described in the inclusion criterion above.
2. Significant liver dysfunction with any of the following abnormal laboratory results:
    - ALT (alanine transaminase) or AST >2X ULN
    - Total bilirubin >2X ULN
    - Alkaline phosphatase >2X ULN or
    - INR (international normalized ratio) ≥ 1.4

    Subjects whose liver laboratory assessments fall outside of these ranges may undergo repeat testing and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor. In addition, subjects with abnormal laboratory results related to confirmed benign liver conditions (eg, Gilbert's syndrome) are considered eligible for the study notwithstanding their abnormal laboratory results and may be enrolled after discussion with the Medical Monitor.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

3. Prior liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used
4. Evidence of any bleeding disorder not related to hemophilia A
5. Platelet count of $< 100 \times 10^9$/L
6. Creatinine ≥ 1.5 mg/dL
7. Liver cirrhosis of any etiology as assessed by liver ultrasound
8. Chronic or active hepatitis B as evidenced by positive serology testing and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results in the Laboratory Manual.
9. Active Hepatitis C as evidenced by detectable HCV RNA, or currently on antiviral therapy
10. Active malignancy, except non-melanoma skin cancer
11. History of hepatic malignancy
12. History of arterial or venous thromboembolic events (eg, deep vein thrombosis, non-hemorrhagic stroke, pulmonary embolism, myocardial infarction, arterial embolus), with the exception of catheter-associated thrombosis for which anti-thrombotic treatment is not currently ongoing.
13. Known inherited or acquired thrombophilia, including conditions associated with increased thromboembolic risk, such as atrial fibrillation.
14. A history of known inflammatory, connective tissue, or autoimmune disorders (eg, vasculitis).
15. Treatment with any Investigational Product within 30 days prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study
16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including possible corticosteroid treatment outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.
17. Prior treatment with any vector or gene transfer agent

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	**SUMMARY TABLE** Referring to Part of the Dossier: Volume: Page: Reference:	**FOR NATIONAL AUTHORITY USE ONLY:**

18. Major surgery planned in the 26-week period following the infusion with BMN 270
19. Use of systemic immunosuppressive agents, not including corticosteroids, or live vaccines within 30 days before the BMN 270 infusion
20. Concurrent enrollment in another clinical study, unless it is an observational (non-interventional) clinical study that does not interfere with the requirements of the current protocol or have the potential to impact the evaluation of safety and efficacy of BMN 270 and with prior consultation with the Medical Monitor
21. Known allergy or hypersensitivity to investigational product formulation
22. Unwilling to receive blood or blood products for treatment of an adverse event and/or a bleed

INVESTIGATIONAL PRODUCT(S), DOSE, ROUTE AND REGIMEN:
Each subject will receive a single IV infusion of BMN 270 at 6E13 vg/kg. The volume of infusion will depend on the subject's weight.

REFERENCE THERAPY(IES), DOSE, ROUTE AND REGIMEN:
No reference therapy will be evaluated in this study.

DURATION OF TREATMENT:
BMN 270 is given as a single dose by IV infusion.

CRITERIA FOR EVALUATION:

Safety:

The following safety outcome measurements will be assessed:

- Incidence of adverse events (AEs), including serious AEs (SAEs)
- Change in clinical laboratory tests (serum chemistry and hematology)
- Change in vital signs
- Change in physical examination
- Vector shedding (blood, urine, semen, feces, saliva)
- Liver function tests (LFTs, including ALT, AST, GGT, LDH, bilirubin, alkaline phosphatase)
- Immune response to FVIII transgene product and AAV5 vector capsid

No major toxicity is expected based on 270-201 data and non-clinical studies. Each subject will have comprehensive surveillance monitoring of LFTs (once per week for Weeks 1-26). During the

NAME OF COMPANY	SUMMARY TABLE	FOR NATIONAL AUTHORITY USE ONLY:
BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	Referring to Part of the Dossier: Volume: Page: Reference:	

long-term safety evaluation, LFTs will be monitored every three months for up to 5 years post-infusion; the frequency and duration of LFT testing may be changed based on discussion between the Medical Monitor and the Investigator and review of subject data.

There will be a detailed assessment of cellular and humoral responses to AAV5 vector capsid and FVIII.

Efficacy:

The efficacy measure will be to assess plasma FVIII activity. The efficacy goal is to achieve FVIII activity ≥ 5 IU/dL at 26 weeks post-BMN 270 administration. Other efficacy measures include assessing the impact of BMN 270 on the use of FVIII replacement therapy and on the number of bleeding episodes during the study. Subjects will be asked to keep a subject diary, provided by the sponsor, to record the relevant details.

Other efficacy endpoints:

- Change from baseline in the total score of HAEMO-QoL-A at Week 26 of the study post-BMN 270 infusion
- Change from baseline in the EQ-5D-5L score at Week 26 of the study post-BMN 270 infusion.
- Change from baseline in the Haemophilia Activities List (HAL) score at Week 26 of the study post-BMN 270 infusion.
- Change from baseline in the Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) score at Week 26 of the study post-BMN 270 infusion.

Pharmacodynamics:

The FVIII antigen and activity level, as measured by a validated immunoassay and a validated FVIII activity assay, respectively, will be used for plasma profiles; FVIII antigen and activity will be used to determine PD parameters.

STATISTICAL METHODS:

Approximately 10 subjects may be dosed in the study. The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using longitudinal methods, such as a mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at ≥ 5 IU/dL at 26 weeks following study treatment. FVIII

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:
activity will be assessed weekly during the study period. Assessment of the true steady state of FVIII will require that FVIII activity is measured after a minimum of 72 hours (or 5x the known half-life of the FVIII replacement therapy administered) has elapsed since the last infusion of FVIII concentrates. Analysis of neutralizing antibody response, other immunological parameters, and vector shedding will be primarily descriptive and involve both inter-subject and intra-subject comparisons across cohorts.		

3 TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

4 LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviations

AAV	adeno-associated virus
ABR	annualized bleeding rate
ADL	activities of daily living
ADR	adverse drug reaction
AE	adverse event
ALT	alanine aminotransferase
APTT	activated partial thromboplastin time
ART	anti-retroviral therapy
BPV	BioMarin Pharmacovigilance
BU	Bethesda Unit
CFR	Code of Federal Regulations
CRA	clinical research associate
CRF	case report form
CSR	clinical study report
CTCAE	Common Terminology Criteria for Adverse Events
DRB	Data Review Board
EC	ethics committee
eCRF	electronic case report form
ED	exposure days
EOSI	events of special interest
ETV	early termination visit
EudraCT	European Union Drug Regulating Authorities Clinical Trials
FAS	Full Analysis Set
FDA	Food and Drug Administration
FIH	first-in-human
FVIII	coagulation factor VIII
GCP	Good Clinical Practice
HA	Hemophilia A
HAL	Haemophilia Activities List
hFIX	human coagulation factor IX
hFVIII	human coagulation factor VIII
HIPAA	Health Insurance Portability and Accountability Act

IB	investigator brochure
ICF	informed consent form
ICH	International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
ICH E6 [R2]	ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6
IND	Investigational New Drug (application)
INR	international normalized ratio
IP	investigational product
IRB	institutional review board
IV	intravenous
LFT	liver function test
MedDRA	Medical Dictionary for Regulatory Activities
PBMC	peripheral blood mononuclear cells
PCR	polymerase chain reaction
PD	pharmacodynamics
PEG	polyethylene glycol
PK	Pharmacokinetics
PRO	patient-reported outcome
rhFVIII	recombinant human FVIII protein
SAE	serious adverse event
SAP	statistical analysis plan
SDV	source data verification
SoA	schedule(s) of activities
TGA	thrombin generation assay
ULN	upper limit of normal
vg	vector genomes
VWF:Ag	von Willebrand factor Antigen
WPAI+CIQ:HS	Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific

Definition of Terms:

Investigational Product (IP):

"A pharmaceutical form of an active ingredient or placebo being tested or used as a reference in a clinical trial, including a product with a marketing authorization when used or assembled (formulated or packaged) in a way different from the approved form, or when used for an unapproved indication, or when used to gain further information about an approved use" (from International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6 [ICH E6] (R2)]).

The terms "IP" and "study drug" may be used interchangeably in the protocol.

5 ETHICS

BioMarin Pharmaceutical Inc. (hereafter referred to as BioMarin or the Sponsor) conducts its studies according to the highest ethical and scientific standards. The following Sections articulate standards to which Investigators will be held accountable, as well as matters of compliance to document adherence to such standards.

5.1 Institutional Review Board or Independent Ethics Committee

Investigators are expected to interact with Ethics Committees (ECs) promptly, as required, during the course of the study. This includes, but is not limited to, providing appropriate documentation to support study initiation and maintaining appropriate flow of safety and other information during the course of the study and for study close-out activities. BioMarin (or designee) will assist Investigators with access to timely and accurate information and with assurance of prompt resolution of any queries.

Prior to initiating the study, the Investigator will obtain written confirmation that the institutional review board (IRB) or independent ethics committee (EC) is properly constituted and compliant with International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) and Good Clinical Practice (GCP) requirements, applicable laws, and local regulations. A copy of the confirmation from the IRB/EC will be provided to BioMarin Pharmaceutical Inc. (BioMarin) or its designee. The Investigator will provide the IRB/EC with all appropriate material, including the protocol, Investigator's Brochure (IB), the Informed Consent Form (ICF) including compensation procedures, and any other written information provided to the subjects, including all ICFs translated for patients who do not speak the local language at the clinical site. The study will not be initiated and Investigational Product (IP) supplies will not be shipped to the site until appropriate documents from the IRB/EC confirming unconditional approval of the protocol, the ICF and all subject recruitment materials are obtained in writing by the Investigator and copies are received at BioMarin or its designee. The approval document should refer to the study by protocol title and BioMarin protocol number (if possible), identify the documents reviewed, and include the date of the review and approval. BioMarin will ensure that the appropriate reports on the progress of the study are made to the IRB/EC and BioMarin by the Investigator in accordance with applicable guidance documents and governmental regulations.

5.2 Ethical Conduct of Study

It is expected that Investigators understand and comply with the protocol. This includes, but is not limited to: establishing and meeting enrollment commitments, including providing eligible subjects for study enrollment; adhering to adverse event reporting, diagnostic, or other procedures as specified in the protocol; and assuring appropriate compliance with study treatment administration and accountability.

This study will be conducted in accordance with the following:

- European Clinical Trial Directive 2001/20/EC and Good Clinical Practice Directive 2005/28/EC, for studies conducted within any European country
- US Code of Federal Regulations (CFR) Sections that address clinical research studies, and/or other national and local regulations, as applicable
- ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6(R2) (ICH E6R2)
- The ethical principles established by the Declaration of Helsinki

Specifically, this study is based on adequately performed laboratory and animal experimentation. The study will be conducted under a protocol reviewed and approved by an IRB/EC and will be conducted by scientifically and medically qualified persons. The benefits of the study are in proportion to the risks. The rights and welfare of the subjects will be respected and the investigators conducting the study do not find the hazards to outweigh the potential benefits. Each subject will provide written, informed consent before any study-related tests or evaluations are performed.

5.3 Subject Information and Informed Consent

A properly written and executed informed consent form (ICF), in compliance with ICH E6R2 (Section 4.8), United States Code of Federal Regulations (CFR) 21 CFR §50, European Clinical Trial Directive 2001/20/EC and Good Clinical Practice Directive 2005/28/EC, and other applicable local regulations, will be obtained for each subject prior to entering the subject into the study. The Investigator will prepare the ICF and provide the documents to BioMarin for approval prior to submission to the IRB/EC approval. BioMarin and the IRB/EC must approve the documents before they are implemented. A copy of the approved ICF and if applicable, a copy of the approved subject information sheet and all ICFs translated to a language other than the native language of the clinical site must also be received by BioMarin or designee prior to any study-specific procedures being performed.

6 INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

During administration of informed consent, expectations regarding participation in the study should be made clear to subjects. Subjects who are not willing and/or are not able to comply with all aspects of the study should not be encouraged to participate.

Prior to beginning the study, the Investigator at each site must provide to BioMarin or designee a fully executed and signed Statement of Investigator (SOI) form. A US Food and Drug Administration (FDA) Form FDA 1572 serves as an acceptable SOI form. If Form FDA 1572 may not be used in a particular region, the Investigator must provide a fully executed SOI on the form provided by the Sponsor. All Investigators and Sub-Investigators must be listed on Form FDA 1572 or its equivalent SOI. Financial Disclosure Forms must also be completed for all Investigators and Sub-Investigators listed on the Form FDA 1572 or SOI who will be directly involved in the treatment or evaluation of subjects in this study.

The study will be administered by and monitored by employees or representatives of BioMarin. Clinical Research Associates (CRAs) or trained designees will monitor each site on a periodic basis and perform verification of source documentation for each subject as well as other required review processes. BioMarin's Regulatory Affairs Department (or designee) will be responsible for the timely reporting of serious adverse events (SAEs) to appropriate regulatory authorities as required.

In multicenter studies, a Coordinating Investigator will be identified who will be responsible for study overview. The Coordinating Investigator will read the clinical study report (CSR) and confirm that it accurately describes the conduct and results of the study, to the best of his or her knowledge. The Coordinating Investigator will be chosen on the basis of active participation in the study, ability to interpret data, and willingness to review and sign the report in a specified timeframe. The identity of the Coordinating Investigator and a list of all Investigators participating in the study will be provided in the CSR.

Clinical Laboratory assessments will be performed at a nominated central laboratory. Bioanalytical samples will be sent to the appropriate specialty laboratories for testing. Refer to laboratory manual for more details.

7 INTRODUCTION

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males (Nathwani, 1992). It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. Clinical manifestations of severe FVIII deficiency are frequent unprovoked bleeding episodes in joints and soft tissues causing permanent disability and occasionally death mostly after brain hemorrhage. Treatment in Western countries (Berntorp, 2012) consists of intravenous injection of plasma-derived or recombinant FVIII protein concentrates at the time of a bleed to control it or prophylactically to prevent bleeding episodes. The short half-life for FVIII (12-18 hours) necessitates frequent infusions and makes this treatment prohibitively expensive for the majority of the world's hemophilia A patients. These individuals develop debilitating arthropathy and have a substantially increased risk of death from hemorrhage in life (Stonebraker, 2010). Chemical modification or bioengineering of FVIII may improve half-life to 18-19 hours (Kaufman, 2013). However, these extended half-life FVIII variants do not eliminate the need for lifelong FVIII protein administration (Hay, 2012).

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of human FVIII (hFVIII) following a single administration of vector. Hemophilia A is well-suited for this approach because its clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in low amounts (100-200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and a modest increase in the level of FVIII (a plasma level of 2 ng/ml protein leads to a 1% expression) can ameliorate the severe phenotype (Srivastava, 2013); thus, the therapeutic goal for gene therapy is a modest increase in hFVIII. Finally, the consequences of gene transfer can be assessed using validated quantitative endpoints that can be easily assayed in most clinical laboratories.

BMN 270 contains the cDNA for the B-domain-deleted SQ FVIII with a liver-specific HLP transcription promoter. The expression cassette is inserted between AAV2 ITRs, and this genome is packaged in the AAV5 capsid. A comprehensive review of BMN 270 is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.1 Nonclinical Studies

The nonclinical program supports a single IV infusion of BMN 270, the planned clinical route of administration, for the treatment of hemophilia A in male patients. This nonclinical program took into account the guidelines and reflection papers for gene therapy medicinal products under EMA Advanced Therapies as well as FDA guidance. The primary pharmacodynamics (PD), pharmacokinetics (PK), and toxicity of IV BMN 270 were characterized in a series of single dose studies in species that were vector permissive and responsive to the transgene including normal CD-1 mice, a B- and T-cell deficient mouse model of hemophilia A (B6;129S-*F8*^{tm1Kaz}/J x B6.129S6-*Rag2*^{tm1Fwa} N12; FVIII KO x Rag2), and normal cynomolgus and rhesus monkeys. Some PD studies evaluated additional PK, immunogenicity and toxicity endpoints.

The comparative pharmacodynamics of BMN 270 in cynomolgus monkeys with varying pre-existing AAV5 transduction inhibition (TI) titer and AAV5 TAb status was evaluated in study BMN270-16-021. BMN 270 was administered to 4 groups of monkeys, a control group (Group 1, n=3) that tested negative for both TI and AAV5 TAb, Group 2 (n=4) that was AAV5 TAb negative, and low TI titer (2-5) positive. Group 3 (n=4) was also AAV5 TAb negative, but had higher TI titers (5-10). Group 4 (n=5) tested positive for both AAV5 TAb and TI (TI titers were >5). Administration of BMN 270 by a single intravenous bolus injection was well-tolerated in cynomolgus monkeys regardless of baseline TI titer or TAb status. After dosing, all monkeys showed FVIII-SQ levels above the LLOQ, with the exception of two monkeys in the group that presented with both positive TI and TAb titers at baseline. Though these TAb+ monkeys, regardless of TI titers, showed a significant mean reduction in FVIII expression (68% less) compared to TAb negative monkeys, three of five monkeys showed detectable levels of FVIII-SQ, with one having levels similar to that observed in the TI and TAb negative control group. Monkeys that were TI+ but TAb-at baseline had FVIII expression levels that were similar to those of the TI and TAb negative control group.

Results of the nonclinical program to date are detailed in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.2 Previous Clinical Studies

Study 270-201 is an ongoing Phase 1/2, dose-escalation study to assess the safety, tolerability, and efficacy of BMN 270 in patients with severe hemophilia A (FVIII $\leq$1 IU/dL). Subjects received a single BMN 270 infusion and are to be followed for safety and efficacy for up to 5 years. A total of 15 subjects have been enrolled and dosed

with a single IV infusion of BMN 270 at one of 4 dose levels (6E12, 2E13, 4E13, and 6E13 vg/kg).

A comprehensive review of safety, efficacy, and immunogenicity results from 270-201 as of the latest data cut is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.3 Study Rationale

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. The clinical phenotype of HA patients generally correlates tightly with the level of residual expression. Severe HA is classified as FVIII activity less than 1% of wild-type (< 1 IU/dL), moderate disease comprises 1-5% of wild-type activity, and the mild form is 5-40% activity. The clinical manifestations of severe HA are frequent spontaneous bleeding episodes, predominantly in joints and soft tissues, with a substantially increased risk of death from hemorrhage when the brain is involved.

Treatment of severe HA presently consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study (Berntorp, 2016) and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with on-demand-only therapy in the same studies, respectively. Continued repeated bleeding events compound debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death.

Chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) and bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half-life to maximum of 18 hours, leaving critical periods when FVIII activity levels are below the therapeutic range and leaving patients vulnerable to bleeding and concomitant sequelae. As such, despite

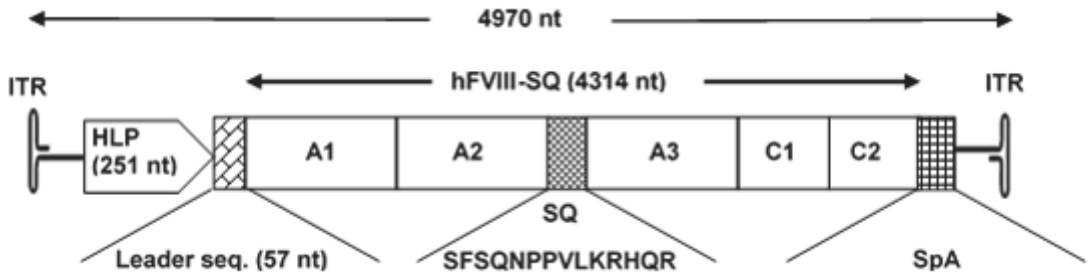
currently available FVIII replacement therapies, a high unmet need remains for maintenance of a sustained, consistent FVIII level compatible with a normal and hemorrhage-free life.

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of active FVIII following a single intravenous infusion of a vector encoding the appropriate gene sequence. Hemophilia A is well-suited for a gene replacement approach because clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in minute amounts (200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and even modest increases in the level of FVIII (any increase of the plasma level by 2 ng/ml induces an increase in activity of 1%) can ameliorate the severe form of hemophilia A. Thus, relatively small changes in endogenous FVIII activity can result in clinically relevant improvements in disease phenotype. Moreover, the circulating FVIII response to gene transduction can be assessed using validated quantitative endpoints that are easily assayed using established laboratory techniques.

Several different gene transfer strategies for FVIII replacement have been evaluated, with adeno-associated viral (AAV) vectors used in most hemophilia clinical trials. AAV vectors have an excellent and well-defined safety profile, and can direct long-term transgene expression with tropism and promoter specificity for specific tissues, such as the liver (eg serotypes 2, 5, and 8). Indeed, an ongoing gene therapy clinical trial for a related disorder, hemophilia B, has established over 7 years of stable human factor IX (hFIX) expression at levels sufficient for conversion of their bleeding phenotype from severe to moderate or mild, following a single peripheral vein infusion of AAV8-hFIX vector (Nathwani, 2014). Several participants in this trial have been able to discontinue factor prophylaxis without suffering spontaneous hemorrhages, even when they undertook activities that previously resulted in bleeding. Thus, gene therapy treatment has resulted in a substantial improvement in their quality of life.

BMN 270 is an AAV5-based gene therapy vector that expresses the SQ form of hFVIII under the control of a hybrid human liver-specific promoter (Figure 7.3.1).

Figure 7.3.1: hFVIII-SQ Vector Genome Schematic



Legend –Note that schematic is not to scale; nt = nucleotides

BMN 270 is delivered by a single IV dose and is designed to achieve stable, potentially life-long expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.

BMN 270 is currently being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. Preliminary results from 270-201 have demonstrated that following gene transfer, sustained FVIII activity above 5% (5 IU/dL) up to a year's observation is achievable at doses of 4-6E13 vg/kg with an acceptable safety profile (Pasi, 2016). The majority of subjects achieved FVIII levels within the normal range for FVIII, with few to no bleeding episodes and discontinuation of FVIII prophylaxis.

Subjects enrolled and infused in 270-201 were screened and negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes.

Current data describing the extent of neutralization or inhibited transduction has been inconsistent across studies, each employing one of several varying methodologies to evaluate both vector-specific antibodies and transduction inhibition (TI). Early studies using AAV2 for the treatment of hemophilia B described inhibited transduction in patients with neutralizing antibody titers of 2 or 3 (Manno, 2006), but more recent studies have described successful transduction in patients who, in retrospect, were determined to have pre-existing antibody to the vector capsid (Majowicz, 2017). Importantly, no drug-related adverse events were described in these patients. As the sensitivity of assays used to detect pre-existing antibody has increased greatly in recent years, the biological relevance of any given titer determination, and comparisons across studies using assays with differing titer sensitivities, requires additional study.

Data from a non-clinical study conducted by BioMarin in cynomolgus monkeys further demonstrated that transduction efficiency can vary greatly in the presence of pre-existing anti-AAV5 antibody. In non-human primates, the ability to attain FVIII levels in the face of measurable AAV5 antibody titers without significant safety risks supports the possibility of achieving a similar response in patients with AAV5 antibody positivity. The totality of available data shows the effect of pre-existing immunogenicity to AVV delivery vectors is not completely understood, and indicates that some patients may benefit from AAV5 gene therapy who are currently excluded.

In this study, patients with severe HA and pre-existing antibodies against AAV5 will be enrolled to evaluate the clinical response to BMN 270 gene therapy. The aim is to evaluate the safety, tolerability, and efficacy of BMN 270 in patients with severe HA and pre-existing antibodies against AAV5 vector capsid at various levels of AAV5 antibody titers.

7.4 Summary of Overall Risks and Benefits

The majority of subjects in the ongoing 270-201 clinical study who have received 4E13 or 6E13 vg/kg doses of BMN 270 have had Grade 1 asymptomatic elevations in ALT. For most subjects, the elevations have reached only slightly above the upper limit of normal (ULN). Based on the effectiveness of transient oral corticosteroid use to suppress a presumed Class 1 (cytotoxic T-cell) response to hepatocytes transduced in prior studies with hepatic transduction with AAV vectors (Mingozzi, 2013), subjects were treated with 7-32 weeks of oral corticosteroids prophylactically or in response to the elevations in ALT to ensure preservation of the transduced hepatocytes. Using this approach, no sustained loss of FVIII activity has been observed in subjects with ALT elevations, consistent with maintaining a high level of hepatocyte function. Moreover, the rise in ALT levels were not accompanied by

significant or lasting aberrations in other liver function tests such as AST, bilirubin or albumin, indicating that extent of toxicity is limited. Overall, the literature suggests that transient elevations in liver enzymes are expected following AAV-based gene therapy for the treatment for hemophilia B without any long-term concerns of hepatic injury (Manno, 2006; Nathwani, 2011; George, 2016; Miesbach, 2016).

At the highest dose evaluated in 270-201 (6E13 vg/kg), the majority of subjects achieved FVIII levels above 50 IU/dL at 52 weeks post-infusion. Subjects in that cohort also reported markedly decreased bleeding episodes compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. Subjects at all dose levels continue to be followed. Subjects in 270-203, who have pre-existing immunity to the AAV5 vector capsid, may get no benefit from the study (in terms of increased FVIII activity) while possibly creating cross-reactive antibodies that may potentially preclude dosing with other serotypes.

The current data available for BMN 270 does not yet permit comprehensive assessment of the benefit:risk profile of this investigational drug. Given the monitoring measures in place in the clinical protocol(s) to minimize the risk to subjects participating in the existing studies, the identified risks are justified by the anticipated benefits that may be afforded to subjects. Each subject in 270-203 will have a comprehensive surveillance plan that monitors LFTs during the study, and elevations in LFT will be addressed according to the guidelines set forth in the protocol. Safety will be assessed by adverse event reporting and clinical laboratory assessments.

For additional information on findings in 270-201, refer to the current version of the IB.

8 STUDY OBJECTIVES

The primary objective of the study is to:

- Assess the safety of a single intravenous administration of BMN 270 in severe HA subjects with pre-existing antibody to AAV5 vector capsid, including development of FVIII neutralizing antibody

The secondary objectives of the study are to:

- Assess the efficacy of BMN 270 defined as FVIII activity at or above 5 IU/dL at Week 26
- Assess the impact of BMN 270 on usage of exogenous FVIII replacement therapy
- Assess the impact of BMN 270 on the number of bleeding episodes requiring exogenous FVIII therapy
- Evaluate the pharmacodynamics of FVIII expression following IV infusion of BMN 270
- Assess the impact of BMN 270 on patient-reported outcomes (PROs)

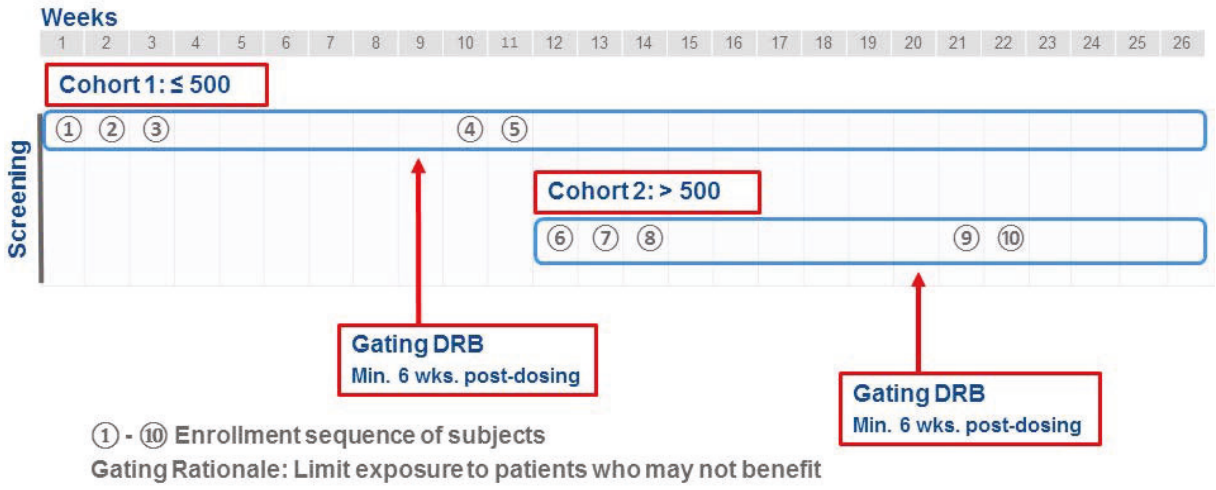
9 INVESTIGATIONAL PLAN

9.1 Overall Study Design and Plan

This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects will be enrolled at 2-3 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date.

The Data Review Board (DRB) will consist of the 2 Principal Investigators and Sponsor's Medical Monitor, as well as hemophilia expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201. The DRB will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 9.1.1 represents one dosing schedule scenario):

Figure 9.1.1: 270-203 Dosing Schedule (One Scenario)



Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from a minimum of 6 weeks post-infusion) reviewed. Based on data from 270-201, FVIII

activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response. Up to 6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness).

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DRB evaluation of safety and efficacy in treated subjects with the following triggers that may potentially pause further enrollment:

- any related SAE;
- any related AE with a severity > CTCAE Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects at 6 weeks post-BMN 270 infusion.

Following a temporary halt of enrolment, the DRB may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed and additional blood samples will be collected (tryptase, C3, C3a, C4, C5, and C5a within 1 hour of hypersensitivity reaction, and IgE between 8-24 hours after the reaction). Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.

Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DRB based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL. Throughout the study, subjects with FVIII activity below 5 IU/dL may be monitored more frequently at the discretion of the Investigator and the Medical Monitor. In subjects who show an initial response to BMN 270 but who later have FVIII activity decline to < 5 IU/dL, the investigator and medical monitor will review the subject's FVIII activity

levels and discuss whether to resume prior FVIII prophylaxis. In addition, the investigator will notify the subject of his FVIII activity levels and will discuss with the subject the risk of bleeding and when (and if) prior FVIII prophylaxis will be resumed.

The study analysis will be performed after all subjects have been followed for 26 weeks post-BMN 270 infusion, with presentation of safety, efficacy, and FVIII PD assessments. After the safety, efficacy, and FVIII PD analyses at 26 weeks post-BMN 270 infusion, long-term safety and efficacy will be assessed in all subjects for up to a total of 5 years post-infusion. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the DRB.

A summary of all assessments is provided in the Schedule of Activities (SoA) in Table 9.1.1, Table 9.1.2, and Table 9.1.3.

Table 9.1.1: Schedule of Activities

Assessment	Prior to BMN 270 Infusion			BMN 270 Infusion Visit (Day 1)[k]
	Screening (Day -28 to Day -1)	Smart Rescreening[i] (Day -28 to Day -1)	Baseline (Day -7 to Day -1)[h]	
Informed consent	X			
Demographics (age, sex, race, ethnicity)	X			
Medical History (including hemophilia A history, Hepatitis B, Hepatitis C, and HIV)	X			
Physical Examination[a]	X		X	X
Height and Weight[a]	X			
Vital Signs	X	X	X	X
Assessment of Adverse Events and Concomitant Medications	X	X	X	X
Documentation of bleeding episodes and FVIII usage for previous 12 months (by either subject or clinical information)	X	X	X	
Distribution of subject diaries and training in their use[l]	X			
Electrocardiogram	X			
Liver Ultrasound	X			
hFVIII Assays[b]	X	X[j]	X	
AAV5 TAb Assays[c]	X	X	X	X
AAV5 TI Assay[c]			X	X
Screen for Hepatitis B, Hepatitis C, HIV[d]	X			
Blood chemistry, hematology, and coagulation tests[e]	X	X	X	
Blood fasting lipid panel				X
Urine Tests[e]	X	X	X	

Assessment	Prior to BMN 270 Infusion			BMN 270 Infusion Visit (Day 1)[k]
	Screening (Day -28 to Day -1)	Smart Rescreening[i] (Day -28 to Day -1)	Baseline (Day -7 to Day -1)[h]	
Liver Function Tests[e]	X	X	X	
PBMC collection (for baseline determination of AAV5 and FVIII specific cellular immunity)			X	
Von Willebrand Factor Antigen (VWF:Ag)	X			
Direct Thrombin Activity Test			X	
Thrombin Generation Assay			X	
PCR of vector DNA in blood, saliva, urine, semen, and stools			X	
Biomarker testing[f]	X			
Exploratory biomarker assessments[g]			X	
Cytokine bead array assay			X	
Hypersensitivity blood assessments[m]			X	
Haemo-QOL-A assessment			X	
EQ-5D-5L assessment			X	
HAL assessment			X	
WPAI+CIQ:HS assessment			X	
BMN 270 Infusion				X

[a] Complete physical examination should be done at Screening. Brief physical examination may be done at Baseline and at the BMN 270 Infusion Visit.

[b] Includes baseline FVIII activity (chromogenic FVIII assay and one-stage clot FVIII assay), coagulation exploratory assay, hFVIII inhibitor level (Bethesda assay with Nijmegen modification), hFVIII total antibody titer, and hFVIII antigen assay. Baseline activity should be assessed at trough (at least >72 hours after last dose of replacement FVIII therapy, or 5x the known half-life of the FVIII replacement therapy administered).

[c] Sample collection on the day of the infusion visit must be performed before the BMN 270 infusion is given. Screening, Smart Re-screening, and Infusion Day samples will be tested in an AAV5 Tab pre-screening assay specifically developed for enrolment purposes. Baseline and all post-dose samples will be tested in a different AAV5 TAb post-dose immunogenicity monitoring assay

[d] Subjects with documented negative results within the last 30 days do not need to be retested.

[e] Detailed in the Section of laboratory assessments and liver function tests. Urine tests will be performed locally; all other laboratory assessments will be performed at the central laboratory.

[f] Includes HLA genotyping and FVIII genotyping; may include TNFα and IL10a single nucleotide polymorphisms.

[g] Blood samples will be collected from subjects to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A disease. The exploratory genetic/genomic testing on these samples is optional.

[h] Should the screening visit occur within 30 days of the drug infusion, physical examination, blood chemistry, LFTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline.

[i] Smart rescreening should only be performed if a subject has been determined to be eligible for the study and is unable to complete the Baseline assessments and Infusion prior to the closing of the original Screening window (28 + 14 days). Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once. Any screening assessment has not been done at the Screening, it must be done during Smart Rescreening.

[j] Only the hFVIII inhibitor level (Bethesda assay with Nijmegen modification) assay must be done at smart rescreening.

[k] Assessments on the day of infusion must be performed prior to the infusion. On the day of the BMN 270 Infusion, vital signs will be monitored prior to the infusion, during the infusion every 15 minutes (± 5 minutes), and following the infusion hourly (± 5 minutes) for a total time of 24 hours during the subject's stay in the clinic.

[l] Diaries should be distributed to subjects who have consented to participate in the study and who have been determined to meet all study eligibility criteria.

[m] In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination, vital signs will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, and C5a) and one sample for IgE between 8-24 hours after the reaction, if possible. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.

Table 9.1.2: Schedule of Activities – Post-Infusion Follow-Up (Week 1-16)

Assessment	Follow-Up After BMN 270 Infusion – Weeks*																	
	Week 1																	
	D2	D4	D8	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Study Day*	2	4	8	15	22	29	36	43	50	57	64	71	78	85	92	99	106	113
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight						X				X				X				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X		X		X		X						X		
Urine Tests[b]														X				
Liver Function Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII antibody titer				X		X		X		X		X		X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]	X	X	X	X	X	X		X		X				X				X
Exploratory biomarker assessments[e]				X				X				X				X		
Haemo-QOL-A assessment														X				
EQ-5D-5L assessment														X				
HAL assessment														X				
WPAI+CIQ:HS assessment														X				
AAV5 TAb Assay	X			X		X		X		X		X		X		X		X

Assessment	Follow-Up After BMN 270 Infusion – Weeks*																	
	Week 1																	
	D2	D4	D8	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Study Day*	2	4	8	15	22	29	36	43	50	57	64	71	78	85	92	99	106	113
AAV5 TI Assay	X			X		X		X		X		X		X		X		X
Testing for reactivation of hepatitis B and hepatitis C																		X[f]
PBMC collection (for determination of AAV5 and FVIII specific immunity)	X			X		X		X		X		X		X		X		X
VWF:Ag						X				X				X				X
Direct Thrombin Activity test															X			
Cytokine bead array assay	X		X	X				X				X				X		

* Visit windows are ± 48 hours starting with the Week 2 visit.

[a] Brief physical examination should be done at all weekly visits.

[b] Detailed in the Section of laboratory assessments and liver function tests. Urine tests will be performed locally; all other laboratory assessments will be performed at the central laboratory. LFTs may be monitored more or less frequently (and in particular when ALT values are >1.5x ULN) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LFTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT > 1.5x ULN during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LFTs during any study week may be performed if: (1) the ALT has increased to above ULN; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and liver enzymes may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic FVIII assay and one-stage clot FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen assay. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Collection to occur until at least 3 consecutive negative results are obtained. Collection and testing of semen samples must continue through Week 12, even if 3 consecutive negative results in that compartment have already been recorded.

[e] Blood samples will be collected from subjects to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A disease. The exploratory genetic/genomic testing on these samples is optional.

[f] Testing for reactivation of hepatitis B and hepatitis C at Week 16 should be performed only in subjects with evidence of prior exposure and who have not received therapeutic oral corticosteroids prior to Week 16; subjects who have received therapeutic oral corticosteroids will have hepatitis B and hepatitis C testing at the time points indicated in Table 9.1.4.

Table 9.1.3: Schedule of Activities – Post-Infusion Follow-Up (Week 17-260)

Assessment	Follow-Up After BMN 270 Infusion – Weeks*										6 mon – 5 years	
	17	18	19	20	21	22	23	24	25	26	Q3M	EOT
Study Day*	120	127	134	141	148	155	162	169	176	183		
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X
Weight[a]				X						X	X	X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X						X	X	X
Urine Tests[b]										X	X	X
Liver Function Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]	X	X	X	X	X	X	X	X	X	X	X	X
FVIII antibody titer				X						X	X	X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]				X						X	X[d]	X
Exploratory biomarker assessments[e]		X				X				X	X	X
Haemo-QOL-A assessment										X	X[f]	X
EQ-5D-5L assessment										X	X[f]	X
HAL assessment										X	X[f]	X
WPAI+CIQ:HS assessment										X	X[f]	X
AAV5 TAb Assay		X		X		X		X		X	X	X
AAV5 TI Assay		X		X		X		X		X	X	X

Assessment	Follow-Up After BMN 270 Infusion – Weeks*										6 mon – 5 years	
	17	18	19	20	21	22	23	24	25	26	Q3M	EOT
Study Day*	120	127	134	141	148	155	162	169	176	183		
PBMC collection (for determination of AAV5 and FVIII specific cellular immunity)		X		X		X		X		X	X	X
VWF:Ag				X						X	X	X
Direct Thrombin Activity Test										X	X	X
Thrombin Generation Assay				X				X		X	X	X
Cytokine bead array assay		X				X				X		

* Visit windows are ± 48 hours through Week 26, and then ± 2 weeks through the end of Year 5.

[a] Brief physical examination should be done at all weekly visits except Week 26, where a complete physical examination should be performed. Weight should be recorded at every 4 weeks and Week 26. After Week 26, complete physical examination should be done at Week 52 and then yearly, brief physical examination at other visits.

[b] Detailed in the Section of laboratory assessments and liver function tests. Urine tests will be performed locally; all other laboratory assessments will be performed at the central laboratory. LFTs may be monitored more or less frequently (and in particular when ALT values are >1.5x ULN) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LFTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT > 1.5x ULN during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LFTs during any study week may be performed if: (1) the ALT has increased to above ULN; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and liver enzymes may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic FVIII assay and one-stage clot FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Collection to occur until at least 3 consecutive negative results are obtained. Sample testing during Weeks 27-260 is not required if at least 3 consecutive samples are clear during the Post-Infusion Follow Up period in Weeks 1-26. Subjects who have not had 3 consecutive negative semen samples by Week 26 should continue to have PCR testing of semen every 4 weeks until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).

[e] Blood samples will be collected from subjects to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A disease. The exploratory genetic/genomic testing on these samples is optional.

[f] Patient-reported outcome (PRO) assessments will be done once each year after Week 26.

Table 9.1.4: Therapeutic Corticosteroids for LFT Elevations or Decreased FVIII Activity

	Steroid Treatment Period[b]								Post-Steroid Period[c]				
	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8[b]	Week 1	Week 2	Week 3	Week 4	Week 13
Therapeutic corticosteroids (dose in mg/day)[a]	60 mg	60 mg	40 mg	40 mg	40 mg	30 mg	20 mg	10 mg					
FVIII activity testing									X	X	X	X	
Liver function testing									X	X	X	X	
Hepatitis B testing[d]						X			X				X
HCV Viral Load[d]						X			X				X

[a] Therapeutic oral corticosteroids may be initiated according to the parameters set out in Section 9.4.8.2.

[b] Following initiation or completion of steroid regimen, if a recurrence of ALT values ≥ 1.5x ULN is reported, steroid management decisions will based on discussions between the Investigator and Medical Monitor. Modification of the steroid regimen may take into consideration possible confounders for the ALT elevation, relationship between increases in ALT and FVIII activity, ALT/FVIII levels post steroid initiation, and adverse events related to steroid dosing. Guidance for tapering oral corticosteroid dosing can be found in Section 9.4.8.2.

[c] After discontinuation of oral corticosteroids, weekly labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values. If these assessments are already being done as part of normal study follow-up, they do not need to be duplicated.

[d] Should only be performed in subjects with a history of hepatitis B or hepatitis C.

9.2 Discussion of Study Design, Including Choice of Control Group

Study 270-203 is designed to be a Phase 1/2, single-arm, open-label study in hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid. Hemophilia A patients who provide written informed consent and meet the entry criteria will be eligible to enroll in the study.

Approximately 10 subjects will be enrolled at the 6E13 vg/kg BMN 270 dose. Subjects will be followed for 26 weeks post-BMN 270 infusion during which safety and efficacy assessments will be taken. After the final analysis at 26 weeks post-infusion, safety and efficacy will then continue to be assessed long-term for approximately a total of 5 years.

9.3 Selection of Study Population

Approximately 10 hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid may enroll into the study.

Additional criteria for participation in the study are provided in Section 9.3.1 and Section 9.3.2.

9.3.1 Inclusion Criteria

Individuals eligible to participate in this study must meet all of the following criteria:

1. Males ≥ 18 years of age with hemophilia A and residual FVIII levels ≤ 1 IU/dL as evidenced by medical history, at the time of signing the informed consent.
2. Detectable pre-existing antibodies against the AAV5 vector capsid as measured by AAV5 total antibody ELISA
3. Treated/exposed to FVIII concentrates or cryoprecipitate for a minimum of 150 exposure days (EDs)
4. Subject must have been on prophylactic FVIII replacement therapy for at least 12 months prior to study entry.
5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures.
6. No history of FVIII inhibitor, and results from a Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) on 2 consecutive occasions (the most recent one of which should be tested at the central laboratory) at least one week apart within the past 12 months
7. HIV positive patients may be enrolled, only if the patient has a CD4 count > 200/mm^3 and an undetectable viral load.

8. Sexually active participants must agree to use an acceptable method of double barrier contraception for at least 6 months post-infusion, which may include hormonal contraception for a female partner. After 6 months, subjects may stop contraception use only if they have had 3 consecutive semen samples with no detectable viral vector DNA.
9. Willing to abstain from consumption of alcohol for at least the first 26 weeks following BMN 270 infusion.

9.3.2 Exclusion Criteria

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Any evidence of active infection or any immunosuppressive disorder, except for HIV infection as described in the inclusion criterion above.
2. Significant liver dysfunction with any of the following abnormal laboratory results:
    - ALT (alanine transaminase) or AST >2X ULN
    - Total bilirubin >2X ULN
    - Alkaline phosphatase >2X ULN or
    - INR (international normalized ratio) ≥ 1.4

    Subjects whose laboratory assessments fall outside of these ranges may undergo repeat testing and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor. In addition, subjects with abnormal laboratory results related to confirmed benign conditions (eg, Gilbert's syndrome) are considered eligible for the study notwithstanding their abnormal laboratory results and may be enrolled after discussion with the Medical Monitor.
3. Prior liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used
4. Evidence of any bleeding disorder not related to hemophilia A
5. Platelet count of < 100 x 10^9/L
6. Creatinine ≥ 1.5 mg/dL
7. Liver cirrhosis of any etiology as assessed by liver ultrasound
8. Chronic or active hepatitis B as evidenced by positive serology testing and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results in the Laboratory Manual.

9. Active Hepatitis C as evidenced by detectable HCV RNA, or currently on antiviral therapy
10. Active malignancy, except non-melanoma skin cancer
11. History of hepatic malignancy
12. History of arterial or venous thromboembolic events (eg, deep vein thrombosis, non-hemorrhagic stroke, pulmonary embolism, myocardial infarction, arterial embolus), with the exception of catheter-associated thrombosis for which anti-thrombotic treatment is not currently ongoing.
13. Known inherited or acquired thrombophilia, including conditions associated with increased thromboembolic risk, such as atrial fibrillation.
14. A history of known inflammatory, connective tissue, or autoimmune disorders (eg, vasculitis).
15. Treatment with any Investigational Product within 30 days prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study
16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including possible corticosteroid treatment outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.
17. Prior treatment with any vector or gene transfer agent
18. Major surgery planned in the 26-week period following the infusion with BMN 270
19. Use of systemic immunosuppressive agents, not including corticosteroids, or live vaccines within 30 days before the BMN 270 infusion
20. Concurrent enrollment in another clinical study, unless it is an observational (non-interventional) clinical study that does not interfere with the requirements of the current protocol or have the potential to impact the evaluation of safety and efficacy of BMN 270 and with prior consultation with the Medical Monitor
21. Known allergy or hypersensitivity to investigational product formulation
22. Unwilling to receive blood or blood products for treatment of an adverse event and/or a bleed

9.3.3 Removal of Subjects from Treatment or Assessment

Subjects may withdraw their consent to participate in the study at any time without prejudice. The Investigator must withdraw from the study any subject who requests to be withdrawn. Such subjects will always be asked about the reason(s) for withdrawal. The Investigator will

discuss with the subject appropriate procedures for withdrawal from the study. The Investigator should ask the subject's consent to perform the procedures listed under the early termination visit. Should a subject withdraw from the study, all efforts will be made to complete and report the observations as thoroughly as possible up to the date of the withdrawal.

A subject's participation in the study may be discontinued at any time at the discretion of BioMarin or of the Investigator and in accordance with his/her clinical judgment. When possible, the tests and evaluations listed for the termination visit should be carried out and every effort will be made to gather follow-up safety data if possible.

BioMarin must be notified of all subject withdrawals as soon as possible. BioMarin also reserves the right to discontinue the study at any time for either clinical or administrative reasons and to discontinue participation by an individual Investigator or site for poor enrollment or noncompliance.

Reasons for which the Investigator or BioMarin may withdraw a subject from the study include, but are not limited to, the following:

- Subject requires medication or medical procedure prohibited by the protocol
- Subject was erroneously enrolled into the study or does not meet entry criteria and not yet been dosed with BMN 270; subjects who do not meet entry criteria but who erroneously receive BMN 270 should remain in the study for safety monitoring
- Subject is lost to follow-up

If a subject fails to return for scheduled visits, a documented effort must be made to determine the reason. If the subject cannot be reached by telephone, a certified letter should be sent to the subject requesting contact with the Investigator. This information should be recorded in the study records.

The Investigator or designee must explain to each subject, before enrollment into the study, that for evaluation of study results, the subject's protected health information obtained during the study may be shared with the study Sponsor, regulatory agencies, and IRB/EC It is the Investigator's (or designee's) responsibility to obtain written permission to use protected health information per country-specific regulations, such as the Health Insurance Portability and Accountability Act (HIPAA) in the US, from each subject. If permission to use protected health information is withdrawn, it is the Investigator's responsibility to obtain a written request, to ensure that no further data will be collected from the subject and the subject will be removed from the study.

9.3.3.1 Study Safety Evaluation Criteria

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DRB evaluation of safety and efficacy in treated subjects with the following triggers for pausing further enrollment:

- any related SAE;
- an related AE with a severity > Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

Additionally, the DRB should consider whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction. Following a temporary halt of enrolment, the DRB may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

If a Grade 4 or Grade 5 adverse event assessed as related to study drug occurs in a study subject, further enrollment into the trial will be halted until a full safety review of the data by the DRB has taken place. Relevant reporting and discussion with the Sponsor and the DRB will take place before resumption of dosing.

If any of the following events occur in a subject in the study who has received BMN 270 infusion, further enrollment into the trial will be temporarily put on hold pending prompt evaluation by the DRB.

1. Liver dysfunction (criteria do not apply to ALT elevations with an extra-hepatic etiology):
    - ALT >5x ULN, for more than 2 weeks
    - ALT >3x ULN and (total bilirubin >2x ULN **or** INR >1.5)
    - ALT >3x ULN with signs and symptoms of liver dysfunction
2. The occurrence of an AE of hepatic failure.
3. The detection of high titer neutralizing antibodies (>5 BU) to hFVIII following BMN 270 infusion in two subjects.
4. The occurrence of any cancer (except non-melanoma skin cancer) at any point after BMN 270 infusion.
5. The occurrence of a thromboembolic event with FVIII activity > 150 IU/dL in one subject.

If any of the following events occurs in a subject in the study who has received BMN 270 infusion, a prompt evaluation by the DRB will be required. Further enrollment into the trial will continue while DRB evaluation is ongoing, unless deemed otherwise by the DRB.

1. Acute hypersensitivity assessed as related to BMN 270
2. The detection of high titer neutralizing antibodies (>5 BU) to hFVIII following BMN 270 infusion in one subject
3. Occurrence of a thromboembolic event

9.3.4 Subject Identification and Replacement of Subjects

Each subject will be assigned a unique subject identifier. This unique identifier will be on all eCRF pages. In legal jurisdictions where use of initials is not permitted, a substitute identifier will be used.

Subjects who withdraw from the study after receiving BMN 270 will not be replaced.

9.3.5 Duration of Subject Participation

The duration of participation for each subject will be approximately 264 weeks. This includes 4 weeks of screening, 1 day of BMN 270 infusion, 26 weeks of Post-Infusion Follow-Up, and 234 weeks of Long-Term Follow-Up.

9.4 Treatments

9.4.1 Treatments Administered

BioMarin and/or its designee will provide the study site with a supply of IP sufficient for the completion of the study. BioMarin is responsible for shipping study drug to clinical sites.

9.4.2 Identity of Investigational Product

9.4.2.1 Product Characteristics and Labeling

BMN 270 is a sterile, clear, colorless-to-pale yellow solution for IV infusion and is supplied in a 2 mL polypropylene cryovial. Each vial contains 1.1 mL of AAV5-hFVIII-SQ at a concentration of 2E13 vector genomes per mL in a pH 7.4 phosphate buffer.

The study drug label includes the following information: lot number, required storage conditions, a precautionary statement, expiry date, study number, and BioMarin name and address. Labelling will be done according to country specific requirements.

9.4.3 Storage

At the study site, all IP must be stored under the conditions specified in the Pharmacy Manual in a secure area accessible only to the designated pharmacists and clinical site personnel. All IP must be stored and inventoried and the inventories must be carefully and accurately documented according to applicable state, federal and local regulations, ICH GCP, and study procedures.

9.4.4 Directions for Administration

After a physical examination performed by the Investigator or designee, subjects will be admitted on the day of BMN 270 infusion. If the subject is found to have an active acute illness at the time of planned infusion, then the infusion should be deferred until the illness has resolved; screening procedures may require repetition if outside the specified window. An IV catheter will be inserted into a suitable peripheral vein (eg, the median cubital vein) and flushed with saline. FVIII replacement therapy will not be given since venipuncture is a minimally invasive procedure in these individuals under ordinary conditions.

BMN 270 will be prepared and infused as a pure solution over a dose-dependent time. Prepared drug will be kept at room temperature prior to administration. An electric syringe pump will be used to infuse through an in-line, low protein binding 0.22 micron filter. BMN 270 will be infused through the catheter using an appropriate infusion pump at a constant rate of 4 mL/min while monitoring the vital signs (pulse, blood pressure, respiration rate and temperature) at 15 minute (±5 minutes) intervals. The anticipated maximum interval from initiation of thawing of BMN 270 to completion of the infusion is 4 hours, although the IP has been shown to be stable at room temperature for 6 hours.

Following completion of the infusion, vital signs will be monitored hourly (± 5 minutes). If the vital signs are stable the catheter will be removed 8 hours after the infusion. Hemostasis at the puncture site will be established by applying pressure according to standard protocol for infusing FVIII concentrates. Subjects will remain in the clinic for at least 24 hours to observe for any immediate toxicity of the procedure; in-patient observation can be extended beyond 24 hours if needed per Investigator discretion. After 24 hours, subjects will be discharged from the clinic unless toxicity has been observed in which case the stay in the clinic may be extended or the subject may transfer to a separate facility based on the evaluation and judgment of the Principal Investigator after consultation with the Medical Monitor.

9.4.5 Method of Assigning Subjects to Treatment Groups

Subjects who meet all eligibility criteria (refer to Section 9.3.1 and Section 9.3.2) may be enrolled into the study. Approval by the Medical Monitor will be required prior to enrollment of each study subject. Upon their enrollment into the study, subjects will be assigned a unique subject number by the Sponsor.

9.4.6 Selection of Dose Used in the Study

Data from an ongoing first in human study (270-201) indicates that following single escalated doses of BMN 270 (6E12, 2E13, 4E13, 6E13 vg/kg), dose-related increases in FVIII activity were observed, with concurrent improvements in bleeding episodes and exogenous FVIII utilization, particularly at the 4E13 and 6E13 vg/kg dose levels. At all dose levels, BMN 270 is considered to be well-tolerated with mild increases in ALT as the most common adverse event. Please refer to the IB for detailed efficacy and safety data. The 6E13 vg/kg dose has been selected for this study to maximize the likelihood of transduction in the face of pre-existing AAV5 antibodies.

9.4.6.1 Selection of Timing of Dose for Each Subject

9.4.7 Blinding

This is an open-label study.

9.4.8 Prior and Concomitant Medications

All prescription and over-the-counter medications (including dietary and herbal supplements) taken by a subject for 30 days before Screening will be recorded on the designated eCRF. The Investigator may prescribe additional medications, deemed necessary to provide adequate prophylactic or supportive care, during the study, as long as the prescribed medication is not prohibited by the protocol. In the event of an emergency, any needed medications may be prescribed without prior approval, but the Medical Monitor must be notified of the use of any contraindicated medications immediately thereafter. Any concomitant medications added or discontinued during the study should be recorded on the eCRF. Medications should, whenever possible, not be recorded in the electronic database with a frequency of PRN.

The following medications are prohibited starting 30 days before Screening and through the end of the study, and the Sponsor must be notified if a subject receives any of these during the study:

- Any investigational therapy
- Systemic immunosuppressive agents, except for corticosteroids

The following medications should be avoided, starting 30 days prior to and for at least 26 weeks after BMN 270 infusion and minimized throughout the remaining duration of the study.

- Alcohol
- Herbal and natural remedies and dietary supplements
- Medications which may be hepatotoxic

Vaccines should also be avoided during this period, but in particular during the first 26 weeks unless clinically indicated.

The following medications should be avoided during oral corticosteroid therapy:

- Vaccines
- NSAIDs

9.4.8.1 Concomitant Hemophilia Treatments

Subjects on prophylactic FVIII therapy will discontinue their regular treatment regimen starting 4 weeks following the day of infusion or after FVIII activity has reached at least 5 IU/dL (whichever is earlier) and switch to an “on-demand” schedule. FVIII replacement therapy can always be taken as needed by the subject for treatment of an acute bleeding episode; the subject must carefully record his treatment and bleeding episodes in his diary. Prophylactic FVIII can be used on a case-by-case basis and in consultation with the Medical Monitor to prevent bleeding in extenuating circumstances (eg, peri-operative).

In addition, information on FVIII usage and bleeding episodes by medical history will be collected from subjects for the 12-month period immediately preceding study enrollment.

9.4.8.2 Therapeutic Glucocorticoid Treatment for Elevated Hepatic Transaminases

Therapeutic oral corticosteroids (prednisone or converted equivalent) should be initiated when either of the following occurs post-BMN 270 infusion in any subject and after

consultation with the Medical Monitor (or their designee if consultation is required outside of the Medical Monitor's waking hours):

- ALT ≥ 1.5x ULN in 2 consecutive assessments within 72 hours and alternative etiologies have been ruled out, or ALT ≥ 3x ULN in 2 consecutive assessments within 48 hours (refer to Table 9.7.6.3.2)
    - Whenever possible, a confirmatory lab draw for ALT should be performed, along with FVIII activity, prior to initiating oral corticosteroids.
    - Corticosteroids may be delayed if elevations in ALT are clearly not related to BMN 270 (eg, elevated in ALT with concurrent increase in CPK due to intensive exercise)

In addition, if FVIII activity drops > 50% at any time post-BMN 270 infusion, a course of therapeutic oral corticosteroids should be considered upon consultation between the Investigator and the Medical Monitor.

The prescribed regimen for therapeutic oral corticosteroids is detailed in Table 9.1.4. Changes to the corticosteroid regimen should be made as follows:

Table 9.4.8.2.1: Adjustments to Corticosteroid Regimen

Tapering Corticosteroid Dose	Subject has been receiving oral corticosteroids <3 weeks	Corticosteroids may be discontinued **if**: • ALT <1.5 ULN; and • FVIII levels > 20 IU/dL and within 10% of the pre-decline FVIII levels; and • There is no concern for adrenal insufficiency post-withdrawal
	Subject has been receiving oral corticosteroids ≥3 weeks	Corticosteroids may be tapered by 10 mg weekly **if**: • ALT <1.5 ULN; and • FVIII levels > 20 IU/dL and within 10% of the pre-decline FVIII levels; and • There is no concern for adrenal insufficiency post-withdrawal
Increasing Corticosteroid Dose	If ALT level is increasing or FVIII level is decreasing while on oral corticosteroids, any increases in oral corticosteroid dosing should be made only upon consultation with the Medical Monitor	

After discontinuation of oral corticosteroids, labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values.

Following initiation or completion of therapeutic oral corticosteroids, if ALT elevation ≥1.5x ULN is reported, corticosteroid management decisions will based on discussions

between the Investigator and Medical Monitor. Modification of the corticosteroid regimen may take into consideration possible confounders for the ALT elevation and impact on FVIII expression.

Management and monitoring of reactions to corticosteroids should be determined by the Investigator's clinical judgment in consultation with the Sponsor's Medical Monitor. This includes the contraindicated use of NSAIDs during corticosteroid treatment and specific monitoring not already covered by the SoA. The use of COX-2 inhibitors, while not contraindicated during corticosteroid treatment, should be limited, if possible. Practical management to prevent complications related to oral corticosteroid therapy may be undertaken at the discretion of the Investigator (eg, evaluation of glucose intolerance, hyperlipidemia etc.). Hepatitis B status and HCV viral load will be rechecked 6 weeks after the start of oral corticosteroid treatment and then 1 week and 13 weeks after the completion of oral corticosteroid treatment. All adverse events (including any adverse events suspected to be caused by or related to corticosteroid use) should be reported as outlined in Section 10 of the protocol.

9.4.8.3 Monitoring of HIV-Positive Subjects

HIV-positive subjects may be enrolled in 270-203 if the subject has a CD4 count > 200/mm^3 and an undetectable viral load.

Subjects should continue anti-retroviral therapy (ART) as prescribed and follow routine monitoring of CD4 count and viral load (Angus, 2016). No alterations in the monitoring are indicated for enrolled immunocompetent HIV-positive subjects who receive corticosteroids as part of their enrollment in 270-203.

9.4.9 Treatment Compliance

Study drug will be administered to subjects at the dosing facility by a qualified health care professional. The quantity dispensed, returned, used, lost, etc. must be recorded on the dispensing log provided for the study. Sites will be instructed to return or destroy all used and unused study drug containers.

9.5 Investigational Product Accountability

The Investigator or designee is responsible for maintaining accurate records (including dates and quantities) of IP(s) received and IP lost or accidentally or deliberately destroyed. The Investigator or designee must retain all unused or expired study supplies until the study monitor (on-site CRA) has confirmed the accountability data.

9.5.1 Return and Disposition of Clinical Supplies

Unused study drug must be kept in a secure location for accountability and reconciliation by the study monitor. The Investigator or designee must provide an explanation for any destroyed or missing study drug or study materials.

Unused study drug may be destroyed on site, per the site's standard operating procedures, but only after BioMarin has granted approval for drug destruction. The monitor must account for all study drug in a formal reconciliation process prior to study drug destruction. All study drug destroyed on site must be documented. Documentation must be provided to BioMarin or designee and retained in the Investigator study files. If a site is unable to destroy study drug appropriately, the site can return unused study drug to BioMarin upon request. The return of study drug or study drug materials must be accounted for on a Study Drug Return Form provided by BioMarin.

All study drug and related materials should be stored, inventoried, reconciled, and destroyed or returned according to applicable state and federal regulations and study procedures. For additional information, please refer to the Study Pharmacy Manual.

9.6 Dietary or Other Protocol Restrictions

There are no dietary or other protocol restrictions for this study. Alcohol should be avoided for the first 26 weeks of the study, and particularly within 48 hours prior to lab work.

9.7 Efficacy and Safety Variables

9.7.1 Efficacy and Safety Measurements Assessed

The SoA (Table 9.1.1, Table 9.1.2, Table 9.1.3) describe the timing of required evaluations.

9.7.2 Efficacy Variables

9.7.2.1 FVIII Activity

Efficacy (response to treatment) will be defined as FVIII activity ≥ 5 IU/dL at Week 26 following BMN 270 infusion.

Values for FVIII activity will be excluded from analysis if obtained within 72 hours since the last infusion of exogenous FVIII protein concentrates (or within 5x the known half-life of the FVIII concentrates administered).

In the event of an FVIII activity level decline during the study:

- If FVIII activity has declined at least 20% from the peak but less than 35% and has declined for at least 2 consecutive assessments, FVIII activity and LFTs should be repeated every 7 days until FVIII activity is stable or increasing
- If FVIII activity has declined >35% from the peak and has declined for at least 2 consecutive assessments, FVIII activity and LFTs should be repeated every 72 hours until FVIII activity is stable or increasing.

In subjects who show an initial response to BMN 270 but who later have FVIII activity decline to < 5 IU/dL, the investigator and medical monitor will review the subject's FVIII activity levels and discuss whether to resume prior FVIII prophylaxis. In addition, the investigator will notify the subject of his FVIII activity levels and will discuss with the subject the risk of bleeding and when (and if) prior FVIII prophylaxis will be resumed.

Note that fluctuations in FVIII activity are common, and if no clear trend indicating a decline in FVIII activity is observed, then this additional testing may be deferred (upon consultation between the Investigator and the Medical Monitor) until either a more clear trend of decline has been demonstrated or until the FVIII activity levels stabilize or increase.

Details on collecting FVIII activity samples are included in the Laboratory Manual.

9.7.2.2 Factor VIII Replacement Therapy/Bleeding Episodes

Additional efficacy variables are:

- Change of annualized utilization (IU/kg) of exogenous FVIII replacement therapy during Week 5 to Week 26 post BMN 270 infusion from the baseline utilization of exogenous FVIII replacement therapy calculated using subjects' historical medical records during the year prior to the enrollment.
- Change of ABRs requiring exogenous FVIII replacement treatment during Week 5 to Week 26 of the study post BMN 270 infusion from the baseline ABR calculated using subjects' historical medical records during the year prior to the enrollment.

During the study, subjects will be asked at each study visit to report the use of factor replacement therapy and the number of bleeding episodes since the previous visit. This information will be captured on the subject's diary or other subject records.

Subjects are strongly encouraged to immediately consult Investigator for guidance regarding exogenous FVIII administration for suspected bleeds or bleeding episodes within the first 6 weeks post BMN 270 infusion.

9.7.2.3 Patient-Reported Outcomes (PRO)

The Haemo-QoL-A questionnaire is a validated hemophilia-specific health-related quality of life questionnaire for adults (Rentz, 2008). It consists of 41 questions covering six domains (Physical Functioning, Role Functioning, Worry, Consequences of Bleeding, Emotional Impact and Treatment Concerns). Items are answered on a 6-point Likert-type scale, ranging from 0 (None of the time) to 5 (All of the time). Higher scores mean better health-related quality of life or less impairment for a particular subscale (Haemo-QoL Study Group, 2017). Details regarding the Haemo-QoL-A assessment will be included in the Study Reference Manual.

The EQ-5D-5L instrument is a self-reported questionnaire designed to measure general health status (The EuroQol Group, 1990, Health Policy) (Brooks, 1996, Health Policy). The EQ-5D-5L is composed of 2-parts: a descriptive system that assesses 5 levels of perceived problems (mobility, self-care, usual activities, pain/discomfort, and anxiety/depression) in 5 dimensions and the EQ visual analogue scale (EQ VAS) assessment for overall health. A sample copy of the EQ-5D-5L and additional information are provided in the Study Reference Manual.

The Haemophilia Activities List (HAL) measures the impact of hemophilia on self-perceived functional abilities in adults (van Genderen, 2006). The instrument consists of multiple domains including lying/sitting/kneeling/standing, leg and arm function, use of transportation, self-care, household tasks, and leisure activities where subjects are asked to rate their level of difficulty with activities of daily living on a 6-point Likert-type scale from 1 (Impossible) to 6 (Never). For some items, subjects are given the choice to answer 'Not applicable'. A sample copy of the HAL and additional information are provided in the Study Reference Manual.

The Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) instrument is designed to measure the effect of disease symptom severity on work productivity and classroom productivity (if applicable) (Recht, 2014). The WPAI+CIQ:HS questionnaire yields scores related to work/classroom absenteeism, reduced on-the-job effectiveness, overall work/classroom impairment, and activity impairment. WPAI+CIQ:HS outcomes are expressed as impairment percentages, with higher numbers indicating greater impairment and less productivity (Reilly, 2002). A sample copy of the WPAI+CIQ:HS and additional information are provided in the Study Reference Manual.

9.7.3 Immunogenicity

Immunogenicity assays will be performed on plasma and PBMCs. The assays will include detection of anti-AAV5 vector capsid and anti-FVIII total antibodies, as well as determination of neutralizing antibodies against FVIII (FVIII inhibitors) and against the AAV5 vector capsid (Transduction Inhibitors, TI). FVIII Inhibitors will be assessed using the Bethesda assay with Nijmegen modification. Any abnormality of the liver parameters will lead to a retrospective immunogenicity assessment to evaluate FVIII- and vector capsid-specific cellular immunogenicity. FVIII- and vector capsid-specific cellular immunity will be assessed by stimulated cytokine secretion using an ELISpot assay performed on collected PBMCs.

9.7.4 Pharmacodynamics

The FVIII protein concentration and activity level as measured by a validated immunoassay and by a validated FVIII activity assay, respectively, will be used for plasma profiles; FVIII protein and activity will be used to determine PD parameters.

9.7.5 Exploratory Assessments

A cytokine bead array assay assessment will be performed at Baseline and then weekly through Week 26.

In addition, blood samples will be collected from subjects at the time points indicated in Table 9.1.1, Table 9.1.2, and Table 9.1.3 to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, and to develop assays used for these evaluations. The exploratory genetic/genomic research to study or try to discover genes that are not yet known to be associated with hemophilia A is optional.

All biomarker samples collected in this study may be used for exploratory biomarker research, including evaluation of additional biomarkers not specifically listed in the protocol. In addition, samples collected for other purposes in this study may be used for exploratory research once testing for the primary purpose has been completed.

9.7.6 Safety Variables

Safety in this study will be determined from evaluation of AEs, clinical laboratory assessments with a particular attention to the liver function, vital signs assessments, physical examinations, and immunogenicity.

9.7.6.1 Adverse Events

The determination, evaluation and reporting of AEs will be performed as outlined in Section 10.

9.7.6.2 Clinical Laboratory Assessments

The scheduled clinical laboratory tests are listed in Table 9.7.6.2.1. Refer to the Study Reference Manual for instructions on obtaining and shipping samples. Urine tests will be performed locally; all other laboratory assessments will be performed at the central laboratory.

Any abnormal test results determined to be clinically significant by the Investigator should be repeated (at the Investigator's discretion) until: (1) the cause of the abnormality is determined; (2) the value returns to baseline or to within normal limits; or (3) the Investigator determines that the abnormal value is no longer clinically significant.

All abnormal clinical laboratory results should be initialed and dated by an Investigator, along with a comment regarding whether or not the result is clinically significant. Each clinically significant laboratory result should be recorded as an adverse event.

The diagnosis, if known, associated with abnormalities in clinical laboratory tests that are considered clinically significant by the Investigator will be recorded on the AE eCRF.

Table 9.7.6.2.1: Clinical Laboratory Tests

Blood Chemistry	Hematology	Urine Tests	Coagulation Screen including:
Albumin	Hemoglobin	Appearance	APTT
BUN	Hematocrit	Color	PT/INR
Calcium	WBC count	pH	TT
Chloride	RBC count	Specific gravity	
Total cholesterol	Platelet count	Ketones	
CPK	Differential cell count	Protein	
Creatinine	RBC indices (MCV and MCH)	Glucose	
CRP		Bilirubin	
Glucose		Nitrite	
Phosphorus		Urobilinogen	
Potassium		Hemoglobin	
Total protein			
Sodium			
Uric Acid			

BUN, blood urea nitrogen; CPK, creatinine phosphokinase; CRP, C-reactive protein; PT, prothrombin time; APTT, activated partial thromboplastin time; RBC, red blood cell; WBC, white blood cell; TT, thrombin time; INR, international normalized ratio; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin.

In addition to scheduled clinical laboratory assessments, a fasting blood lipid panel (including triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) will be assessed at the BMN 270 infusion visit.

9.7.6.3 Liver Function and Hepatitis Testing

Subjects will be screened for evidence of previous or active hepatitis B or hepatitis C infection at Screening. Subjects with documented results showing an absence of active hepatitis B or hepatitis C infection (as measured by positive surface antigen for hepatitis B or positive RNA testing for hepatitis C) 30 days prior to providing signed informed consent do not need to repeat those tests during the screening period.

Evidence of ongoing hepatitis B or hepatitis C infection is exclusionary. Subjects who have cleared a Hepatitis B infection or are seronegative do not need to receive the hepatitis B vaccination. Subjects with evidence of prior exposure will be tested for hepatitis B and hepatitis C reactivation at Week 16. Subjects who receive therapeutic oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.4.

A liver ultrasound and liver function testing at Screening will identify any significant hepatic dysfunction.

Liver function tests will be monitored on a regular basis; at each time point specified in the SoA, the following LFTs should be assessed:

Table 9.7.6.3.1: Liver Function Tests

Liver Function Tests			
Alkaline Phosphatase	AST (SGOT)	Total Bilirubin	LDH
ALT (SGPT)	Direct Bilirubin	GGT	

ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma-glutamyltransferase; LDH, lactate dehydrogenase; SGOT, serum glutamic-oxaloacetic transaminase; SGPT, serum glutamic-pyruvic transaminase

Elevated ALT levels (above the upper limit of normal range) should be evaluated according to the following plan:

Table 9.7.6.3.2: Evaluation of ALT Elevations

ALT Level	Work-Up
Above ULN and <1.5x ULN	• Continue to monitor LFTs and FVIII per protocol (repeat within 7 days if next protocol scheduled visit is >7 days from the time of the reported ALT elevation) • Consider evaluation to rule out alternative etiology (eg, concomitant medications, viral or autoimmune hepatitis, alcohol use, recreational drug use, special diets, strenuous exercise, prior and/or concurrent illnesses, exposure to environmental and/or industrial chemicals, etc.) (refer to Table 9.7.6.3.3)
1.5 - <3x ULN	• Repeat LFTs and FVIII within 72 hours • Continue to monitor LFTs weekly until ALT is stable or improving • Evaluate and rule out alternative etiologies (as above) • Consult with Medical Monitor • If ALT is ≥ 1.5x ULN in 2 consecutive assessments within 72 hours and alternative etiologies have been ruled out, start oral corticosteroids (refer to Section 9.4.8.2)
≥3x ULN	• Consult with Medical Monitor • Evaluate and rule out alternative etiologies (as above) • Repeat LFTs and FVIII within 48 hours, and continue with monitoring of LFTs at least twice weekly for as long as the subject's ALT remains ≥ 3x ULN • If ≥3x ULN in 2 consecutive assessments within 48 hours, start oral corticosteroids (refer to Section 9.4.8.2) • Obtain other possibly relevant laboratory evaluations (albumin, PT/INR, CRP, etc.) • Obtain complete blood count with differential to assess for eosinophilia • Obtain PBMC to evaluate potential immune response (prior to starting oral corticosteroids) • If no improvement in 14 days, consider gastroenterology and/or hepatology consult, abdominal workup, imaging (including MRI or ultrasound), and/or liver biopsy as appropriate

When ruling out alternative viral or autoimmune hepatitis as part of the elevated ALT workup, the following tests should be performed:

Table 9.7.6.3.3: Viral and Autoimmune Hepatitis Testing

Viral Hepatitis Workup Testing	Autoimmune Hepatitis Workup Testing
Hepatitis A	Smooth muscle antibody
Hepatitis B	Mitochondrial antibody
Hepatitis C	Liver/kidney microsomal antibodies
Cytomegalovirus (CMV)	Antinuclear antibody (ANA) HEP-2
Epstein-Barr virus (EBV)	
Herpes simplex virus (HSV) 1 & 2	

9.7.6.4 HIV Testing

HIV testing will be performed at Screening. Subjects with documented negative results within the last 30 days prior to screening do not need to be retested.

9.7.6.5 Vital Signs, Physical Examinations and Other Observations Related to Safety

Vital signs will include seated systolic and diastolic blood pressure, heart rate, respiration rate, and temperature. Any clinically significant change in vital signs will be recorded as an AE.

Systolic blood pressure, diastolic blood pressure, heart rate, respiration rate, and temperature will be assessed at Screening, Baseline, and at the beginning of each visit during the Post-Infusion Follow-Up and Long-Term Follow-Up periods. On the day of the BMN 270 Infusion, vital signs will be monitored prior to infusion, during the infusion every 15 minutes (± 5 minutes), following the infusion hourly (± 5 minutes) for at least 8 hours during the subject's stay in the clinic. Any abnormal vital sign assessments should be repeated, and both values should be recorded in the eCRF.

A complete physical examination should be performed at Screening, Week 26, Week 52, and then yearly until the end of the study. At other visits, brief physical examinations may be performed at the discretion of the Investigator based on the subject's clinical condition. Particular attention should be given to signs of bleeding, as well as assessing possible hemarthroses.

A complete physical examination will include general appearance (head, eyes, ears, nose, and throat), cardiovascular, dermatologic, lymphatic, respiratory, gastrointestinal, genitourinary, musculoskeletal, and neurologic systems.

A brief physical examination will include general appearance, cardiovascular, dermatologic, respiratory, gastrointestinal, musculoskeletal, and neurologic assessments.

Height will be recorded at Screening only. Weight will be recorded at Screening and then every 4 weeks thereafter through Week 20, at Week 26, and then every 3 months thereafter until the end of the study.

9.7.6.6 Vector Shedding

During the Post-Infusion Follow-Up period, subjects will undergo testing of various bodily samples to look for evidence of vector shedding for possible viral transmission. Bodily fluids will be tested by PCR. Fluids tested will include:

- Blood
- Saliva
- Semen
- Urine
- Stool

Vector shedding will also be extensively studied in the present clinical trial, at the time points indicated in Table 9.1.1, Table 9.1.2, and Table 9.1.3. Testing will continue until at least 3 negative results are obtained. Testing of semen will continue at least through Week 12, even if 3 consecutive negative results have been recorded in that compartment prior to that time point. Subjects who have not had 3 consecutive negative semen samples by Week 26 should continue to have PCR testing in semen every 4 weeks until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).

Samples may be fractionated prior to shedding analysis in order to better characterize the presence, structure, and location of vector DNA and/or vector capsid within each matrix. If needed, the fractionation will be performed with samples collected specifically for shedding analysis (saliva, blood, semen, urine, stool). Alternatively, the vector DNA characterization during shedding analysis may utilize already fractionated exploratory samples obtained from the above biofluids, such as exploratory plasma samples, exploratory PBMC samples, and red blood cells recovered during PBMC/plasma isolations.

Fractionation of semen to collect purified sperm separately from non-sperm cells may be performed in parallel at any visit where semen samples are collected. The shedding analysis of a fractionated semen sample will only be performed if vector DNA was detected in the whole semen sample for the same visit. Fractionation of semen during shedding analysis

may be stopped if purified sperm tested positive for vector DNA on at least three visits, or if purified sperm tested negative for vector DNA on at least three consecutive visits.

Contraception use may need to be extended beyond 26 weeks in individual subjects based on observed vector shedding in semen. After 26 weeks, subjects may stop contraception use only if they have had 3 consecutive negative semen samples.

Details for sample collection and storage are provided in the Laboratory Manual.

10 REPORTING ADVERSE EVENTS

10.1 Safety Parameters and Definitions

Safety assessments will consist of monitoring and recording protocol-defined AEs and SAEs; measurement of protocol-specified hematology, clinical chemistry, and urinalysis variables; measurement of protocol-specified vital signs; and other protocol-defined events of special interest that are deemed critical to the safety evaluation of the study drug.

10.1.1 Adverse Events

For this protocol, an adverse event (AE) is any untoward medical occurrence in a subject, temporally associated with the use of study intervention, whether or not considered related to the study intervention.

An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of study intervention.

Events meeting the AE definition include:

- Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition.
- New conditions detected or diagnosed after study intervention administration even though it may have been present before the start of the study.
- Signs, symptoms, or the clinical sequelae of a suspected drug-drug interaction.

Events not meeting the AE definition include:

- Any clinically significant abnormal laboratory findings or other abnormal safety assessments which are associated with the underlying disease, unless judged by the investigator to be more severe than expected for the subject's condition.
- The disease/disorder being studied or expected progression, signs, or symptoms of the disease/disorder being studied, unless more severe than expected for the subject's condition.
- Medical or surgical procedure (eg, endoscopy, appendectomy): the condition that leads to the procedure is the AE.
- Situations in which an untoward medical occurrence did not occur (social and/or convenience admission to a hospital).
- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

10.1.1.1 Bleeding and Suspected Bleeding Events

All bleeding events and suspected bleeding events, regardless of the need for exogenous FVIII therapy as treatment, should be captured in subject diaries and recorded on the designated bleeding eCRF. Bleeding events and suspected bleeding events should not be reported as adverse events, with the following exception:

- All bleeding events and suspected bleeding events which meet one or more of the criteria for being serious (refer to Section 10.1) should be reported as serious adverse events (whether or not they are bleeding events that are normal sequelae of hemophilia, and whether or not they required exogenous FVIII as treatment).

10.2 Serious Adverse Events

A serious adverse event (SAE) is any untoward medical occurrence that, at any dose:

- Results in death
- Is life-threatening

  Note: Life-threatening refers to an event in which the subject was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.
- Requires inpatient hospitalization or prolongation of existing hospitalization

  Note: In general, hospitalization signifies that the subject has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization, the event is serious.

  Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE.
- Results in persistent or significant disability or incapacity

- Is a congenital anomaly or birth defect in the child or fetus of a subject exposed to IP prior to conception or during pregnancy
- Is an important medical event or reaction – that, based on medical judgment, may jeopardize the subject or require medical/surgical intervention to prevent one of the other outcomes listed above (eg, anaphylaxis)

10.2.1 Events of Special Interest (EOSI)

The following EOSI need to be reported to the Sponsor within 24 hours of site awareness, irrespective of seriousness, severity or causality:

- Elevation of ALT > 1.5x ULN, regardless of whether that elevation triggers an initiation or modification of oral corticosteroid treatment
- Thromboembolic event
- Drug-related anaphylactoid or hypersensitivity reactions

10.3 Methods and Timing for Capturing and Assessing Safety Parameters

10.3.1 Adverse Event Reporting Period

The study AE reporting period is as follows: After informed consent but prior to initiation of study drug, only SAEs associated with any protocol-imposed interventions will be collected. After informed consent is obtained and following infusion of study drug, the reporting period for all non-serious AEs and SAEs begins and continues for approximately 5 years or until study discontinuation/termination, whichever is longer. The criteria for determining, and the reporting of SAEs is provided in Section 10.1.

10.3.2 Eliciting Adverse Events

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and non-leading verbal questioning of the subject is the preferred method to inquire about AE occurrences. The Investigator will record all relevant AE/SAE/EOSI information in the subject's medical record and AE Case Report Form (eCRF).

10.3.3 Assessment of Seriousness, Severity, and Causality

The Investigator responsible for the care of the subject or medically qualified designee will assess AEs for severity, relationship to study drug, and seriousness (refer to Section 10.2 for SAE definitions). These assessments must be made by a study clinician with the training and authority to make a diagnosis (eg, MD/DO, physician's assistant, nurse practitioner, or DDS).

10.3.3.1 Seriousness

The Investigator will assess if an AE should be classified as "serious" based on the seriousness criteria enumerated in Section 10.2. Seriousness serves as a guide for defining regulatory reporting obligations.

10.3.3.2 Severity

Severity (as in mild, moderate, or severe headache) is not equivalent to seriousness, which is based on subject/event outcome or action criteria usually associated with events that pose a threat to a subject's life or functioning. The Investigator will determine the severity of each AE, SAE and EOSI using the NCI CTCAE v4.03. Adverse events that do not have a corresponding CTCAE term will be assessed according to the general guidelines for grading used in the CTCAE v4.03 as stated in Table 10.3.3.2.1.

Table 10.3.3.2.1: Adverse Event Grading (Severity) Scale

Grade	Description	
1	Mild: asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated	
2	Moderate: minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living (ADL)[a]	
3	Severe or medically significant but not immediately life-threatening: hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL[b]	
4	Life threatening consequences; urgent intervention indicated	Grade 4 and 5 AEs should always be reported as SAEs
5	Death related to AE	

[a] Instrumental ADL refer to the following examples: preparing meals, shopping for groceries or clothes, using the telephone, managing money.

[b] Self-care ADL refer to the following examples: bathing, dressing and undressing, feeding oneself, using the toilet, taking medications, not bedridden.

10.3.3.3 Causality

The Investigator will determine the relationship of an AE to the study drug and will record it on the source documents and AE eCRF. To ensure consistency of causality assessments, Investigators should apply the guidance in Table 10.3.3.3.1.

Table 10.3.3.3.1: Causality Attribution Guidance

Relationship	Description
Not Related	• Exposure to the IP has not occurred **OR** • The administration of the IP and the occurrence of the AE are not reasonably related in time **OR** • The AE is considered likely to be related to an etiology other than the use of the IP; that is, there are no facts, evidence, or arguments to suggest a causal relationship to the IP.
Related	• The administration of the IP and the occurrence of the AE are reasonably related in time **AND** • The AE could possibly be explained by factors or causes other than exposure to the IP **OR** • The administration of IP and the occurrence of the AE are reasonably related in time **AND** • The AE is more likely explained by exposure to the IP than by other factors or causes.

Factors suggestive of a causal relationship could include (but are not limited to):

- Plausible temporal relationship
- Absence of alternative explanations
- Rarity of event in a given patient or disease state
- Absence of event prior to study drug exposure
- Consistency with study product pharmacology
- Known relationship to underlying mechanism of study drug action
- Similarity to adverse reactions seen with related drug products
- Abatement of AE with discontinuation of study drug, and/or recurrence of AE with reintroduction of study drug

The Investigator's assessment of causality for individual AE reports is part of the study documentation process. Regardless of the Investigator's assessment of causality for individual AE reports, the Sponsor will promptly evaluate all reported SAEs against cumulative product experience to identify and expeditiously communicate possible new safety findings to Investigators and applicable regulatory authorities.

10.4 Procedures for Recording Adverse Events

10.4.1 Recording Adverse Events on a eCRF

Investigators should use precise medical terminology when recording AEs or SAEs on the AE eCRF. Avoid colloquialisms and abbreviations.

Record only one diagnosis, sign, or symptom per event field on the AE eCRF (eg, nausea and vomiting should not be recorded in the same entry, but as 2 separate entries).

In order to classify AEs and diseases, preferred terms will be assigned by the Sponsor to the original terms entered on the AE eCRF, using MedDRA (Medical Dictionary for Regulatory Activities) terminology.

10.4.1.1 Diagnosis versus Signs and Symptoms

The Investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE. Using accepted medical terminology, enter the diagnosis (if known). If not known, enter sign(s) and/or symptom(s). If a diagnosis subsequently becomes available, then this diagnosis should be entered on the AE (or SAE, as appropriate) eCRF, replacing the original entries where appropriate.

10.4.1.2 Adverse Events Occurring Secondary to Other Events

In general, AEs occurring secondary to other events (eg, cascade events) should be identified by their primary cause. For example, if severe diarrhea is known to have resulted in dehydration, it is sufficient to record only diarrhea as an AE or SAE on the AE eCRF. However, medically important events that may be linked and/or separated in time should be recorded as independent events on the AE eCRF. For example, if severe hemorrhage leads to renal failure, both events should be recorded separately on the AE eCRF.

10.4.1.3 Persistent or Recurrent Adverse Events

A persistent AE (duration of adverse event > 7 days) is one that extends continuously, without resolution, between subject evaluation time points. Events that change in severity

necessitate the recording of an additional AE. AEs that do not have a change in severity should be recorded only once on the eCRF.

A recurrent AE is one that occurs and resolves between subject evaluation time points, but then subsequently recurs. All recurrences of the AE should be recorded on the AE eCRF. For example, if a subject has an adverse event of ALT increased that subsequently resolves, but the subject's ALT increases again, that should be reported as two adverse events – the initial ALT increase, and the second ALT increase.

10.4.1.4 Abnormal Laboratory Values

Laboratory test results (including any local FVIII activity or liver function test results) will be recorded on the laboratory results pages of the eCRF, or appear on electronically produced laboratory reports submitted directly from the central laboratory, if applicable.

Any laboratory result abnormality fulfilling the criteria for a SAE or EOSI should be reported as such, and recorded in the AE eCRF.

Any laboratory result abnormality of CTCAE Grade 4 or 5 should be recorded as an SAE in the AE eCRF.

A clinical laboratory abnormality is considered clinically significant and should be documented as an AE if not refuted by a repeat test to confirm the abnormality and **any** one or more of the following conditions is met:

- Accompanied by clinical symptoms
- Requiring a change in concomitant therapy (eg, addition of, interruption of, discontinuation of, or any other change in a concomitant medication, therapy or treatment).
- The abnormality suggests a disease and/or organ toxicity
- The abnormality is of a degree that requires active management (eg, change of dose, discontinuation of study drug, more frequent follow-up assessments, further diagnostic investigation, etc.)

This applies to any protocol and non-protocol specified safety and efficacy laboratory result from tests performed after the first dose of study medication that falls outside the laboratory reference range and meets the clinical significance criteria.

This does not apply to any abnormal laboratory result that falls outside the laboratory reference range but that does not meet the clinical significance criteria (these will be analyzed and reported as laboratory abnormalities), those that are considered AEs of the type

explicitly exempted by the protocol, or those which are a result of an AE that has already been reported.

For purposes of this study, laboratory tests showing a decreased level of FVIII activity should not be reported as adverse events unless there is an impact to clinical outcomes (eg, increased rate of bleeding, worsening of joint disease).

10.4.1.5 Pre-existing Conditions

A pre-existing condition is one that is present prior to administration of BMN 270. Such conditions should be recorded as medical history on the appropriate eCRF.

A pre-existing condition should be recorded as an AE or SAE during the study **only** if the frequency, intensity, or character of the condition worsens during the study period. It is important to convey the concept that a pre-existing condition has changed by including applicable language in the verbatim description of the event (eg, *more frequent* headaches).

10.4.1.6 General Physical Examination Findings

At screening, any clinically significant abnormality should be recorded as a pre-existing condition (refer to Section 10.4.1.5). During the study, any new clinically significant findings and/or abnormalities discovered on physical examination that meet the definition of an AE (or an SAE) must be recorded and documented as an AE or SAE on the AE eCRF.

10.4.1.7 Hospitalization, Prolonged Hospitalization, or Surgery

Any AE that results in hospitalization or prolonged hospitalization should be documented and reported as an SAE unless specifically instructed otherwise in this protocol (refer to Section 10.2).

There are some hospitalization scenarios that do not require reporting as an SAE when there is no occurrence of an AE. These scenarios include planned hospitalizations or prolonged hospitalizations to:

- Perform a protocol-mandated efficacy measurement
- Undergo a diagnostic or elective surgical procedure for a pre-existing medical condition that has not worsened
- Insert an in-dwelling IV catheter (such as a Port-a-Cath or other brand, if applicable) for administration of study drug or FVIII replacement therapy
- Receive scheduled therapy (study drug or otherwise) for the study indication

10.4.1.8 Deaths

All deaths that occur during the AE reporting period (refer to Section 10.3.1), regardless of attribution, will be recorded on the AE eCRF and expeditiously reported to the Sponsor as an SAE.

When recording a death, the event or condition that caused or contributed to the fatal outcome should be recorded as the single medical concept on the eCRF. If the cause of death is unknown and cannot be ascertained at the time of reporting, record "Unexplained Death" or "Death of Unknown Cause" on the AE eCRF.

10.4.1.9 Pregnancy

Although not an AE per se, pregnancy in the partner of a subject taking trial medication should be reported expeditiously to the Sponsor to facilitate outcome monitoring by the Sponsor. Pregnancy in partner should be reported during the period up to 5 years after viral infusion.

Pregnancy in a partner should be reported within 24 hours of the site becoming aware of the pregnancy by entering the information on the Pregnancy eCRF and submitting to BPV within 24 hours of the site becoming aware of the event. The Investigator must make every effort to follow the subject's partner (with that partner's consent) through resolution of the pregnancy (delivery or termination) and to report the resolution on the Pregnancy Follow-up eCRF. In the event of pregnancy in the partner of a study subject, the Investigator should make every reasonable attempt to obtain the woman's consent for release of protected health information.

Abortion, whether therapeutic or spontaneous, should always be classified as an SAE (as the Sponsor considers these to be medically significant), recorded on the AE eCRF, and expeditiously reported to the Sponsor as an SAE.

10.5 Reporting Requirements

10.5.1 Expedited Reporting Requirements

All SAEs and EOSI that occur during the course of the AE Reporting Period (refer to Section 10.3.1), whether or not considered related to study drug, must be reported by entering the information in the AE eCRF and submitting to BPV within 24 hours of the site becoming aware of the event. Investigators should not wait to collect information that fully documents the event before notifying BPV of an SAE. BioMarin may be required to report certain SAEs to regulatory authorities within 7 calendar days of being notified about the event; therefore, it is important that Investigators submit any information requested by BioMarin as

soon as it becomes available. IND safety reports will be submitted within 7 calendar days for unexpected fatal or life-threatening unexpected suspected adverse reactions (SUSARs) and within 15 calendar days for other non-life-threatening SUSARs.

The Sponsor is responsible for identifying, preparing and reporting all SUSARs to the relevant competent authorities, ethics committees and Investigators in accordance with the requirements identified in the Clinical Trials Regulations.

If the EDC is unavailable, all SAEs should be reported to BPV by completing the SAE Report Form and faxing or emailing the completed form to BPV within 24 hours of the site becoming aware of the event. Once the EDC is available, the information should be entered in the AE eCRF.

10.5.2 Institutional Review Board or Independent Ethics Committee Reporting Requirements

Reporting of SAEs to the Independent Ethics Committee (EC) or Institutional Review Board (IRB) will be done in compliance with the standard operating procedures and policies of the IRB/EC and with applicable regulatory requirements. Adequate documentation must be obtained by BioMarin showing that the IRB/EC was properly and promptly notified as required.

10.6 Follow-up of Subjects after Adverse Events

After the initial AE/SAE/EOSI report, the Investigator is required to proactively follow each subject at subsequent visits/contacts. All AEs/SAEs/EOSI will be followed until resolution, stabilization, the event is otherwise explained, or the subject is lost to follow-up. Resolution of AEs/SAEs/EOSI (with dates) should be documented on the AE eCRF and submitted to BioMarin Pharmacovigilance and in the subject's medical record to facilitate source data verification.

For some SAEs and EOSI, the Sponsor may follow up by telephone, fax, electronic mail, and/or a monitoring visit to obtain additional case details (eg, hospital discharge summary, consultant report, or autopsy report) deemed necessary to appropriately evaluate the SAE or EOSI report.

10.7 Post-Study Adverse Events

At the last scheduled visit, the Investigator should instruct each subject to report, to the Investigator and/or to BPV directly, any subsequent SAEs that the subject's personal physician(s) believes might be related to prior study drug.

The Investigator should notify the study Sponsor of any death or SAE occurring at any time after a subject has discontinued or terminated study participation, if the Investigator believes that the death or SAE may have been related to prior study drug. The Sponsor should also be notified if the Investigator should become aware of the development of cancer or of a congenital anomaly in a subsequently conceived offspring of a subject that participated in this study.

10.8 Urgent Safety Measures

The regulations governing clinical trials state that the Sponsor and Investigator are required to take appropriate urgent safety measures to protect subjects against any immediate hazards that may affect the safety of subjects, and that the appropriate regulatory bodies should be notified according to their respective regulations. According to the European Union (EU) Clinical Trial Directive 2001/20/EC, "…in the light of the circumstances, notably the occurrence of any new event relating to the conduct of the trial or the development of the investigational medicinal product where that new event is likely to affect the safety of the patients, the Sponsor and the Investigator shall take appropriate urgent safety measures to protect the patients against any immediate hazard. The Sponsor shall forthwith inform the competent authorities of those new events and the measures taken and shall ensure that the IRB/EC is notified at the same time."

The reporting period for these events which may require the implementation of urgent safety measures is the period from the time of signing of the ICF through the completion of the last study visit or at the Early Termination Visit (ETV). Investigators are required to report any events which may require the implementation of urgent safety measures to BioMarin within 24 hours.

Examples of situations that may require urgent safety measures include discovery of the following:

- Lack of study scientific value, or detrimental study conduct or management
- Discovery that the quality or safety of the IP does not meet established safety requirements

10.9 BioMarin Pharmacovigilance Contact Information

Contact information for BioMarin Pharmacovigilance is as follows:

BioMarin Pharmaceutical Inc.
Address 105 Digital Drive
Novato, CA 94949
Phone: PI
Fax: PI
E-mail: drugsafety@bmrn.com

The Investigator is encouraged to discuss with the medical monitor any AEs for which the issue of seriousness is unclear or questioned. Contact information for the medical monitor is as follows:

Name: PI, MD
Address 105 Digital Drive
Novato, CA 94949
Phone: PI
E-mail: PI

11 APPROPRIATENESS OF MEASUREMENTS

The measures of efficacy to be used in this study are standard, ie, widely used and generally recognized as reliable, accurate, and relevant (able to discriminate between effective and ineffective agents). The measures of safety used in this study are routine clinical and laboratory procedures.

The chromogenic FVIII assay and the one-stage clot FVIII assay are both validated and utilize CE marked reagents. The exploratory FVIII activity assay will be used for exploratory purposes only.

12 STUDY PROCEDURES

12.1 Prestudy

An ICF must be signed and dated by the subject, the Investigator or designee and witness (if required) before any study-related procedures are performed.

12.2 Screening Visit(s)

Screening assessments should be performed within 28 days of BMN 270 infusion (and must be performed within 42 days prior to BMN 270 infusion), while baseline assessments will take place within 7 days prior to BMN 270 infusion (Day 1). Should the screening visit occur within 30 days of the drug infusion, physical examination, vital signs, blood chemistry, LFTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline.

The following procedures will be performed during the Screening Period:

- Demographics (age, sex, race, ethnicity)
- Full medical history, including hemophilia A history, Hepatitis B, Hepatitis C, and HIV
- Complete Physical Examination
- Height and weight
- Vital Signs (systolic and diastolic blood pressure, heart rate, respiration rate, and temperature)
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information) for the previous 12 months
- Distribution of subject diaries and training in diary completion
- Electrocardiogram
- Liver Ultrasound
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic FVIII assay
    - Baseline FVIII activity level – one-stage clot FVIII assay
    - hFVIII coagulation activity exploratory assay (collected but not tested prior to enrollment)
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)

  - hFVIII total antibody assay (collected but not tested prior to enrollment)
  - hFVIII antigen assay (collected but not tested prior to enrollment)
- Blood sample for AAV5 total antibody (TAb) assay
  - Screening sample will be tested in an AAV5 Tab pre-screening assay specifically developed for enrolment purposes.
- Screen for Hepatitis B, Hepatitis C, and HIV if required (subjects with documented negative results 30 days prior to informed consent being obtained do not need to be retested)
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Function Tests (refer to Table 9.7.6.3.1)
- Von Willebrand Factor Antigen (VWF:Ag)
- Blood samples for Biomarker testing (may include HLA genotyping, FVIII genotyping status, TNFα and IL10a single nucleotide polymorphisms)

12.2.1 "Smart Rescreening" Visit

Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once.

If a patient has to be screened again because the original assessments have fallen out of the 28 + 14 day period allowed for Screening (refer to Section 12.2), then only the following assessments need to be performed (rather than the full list indicated in Section 12.2) for the patient to be successfully re-screened for the study:

- Vital signs
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- hFVIII Assays (only the hFVIII inhibitor level (Bethesda assay with Nijmegen modification))
- Blood sample for AAV5 Total Antibody assay
  - Smart Re-screening sample will be tested in an AAV5 Tab pre-screening assay specifically developed for enrolment purposes.

- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Function Tests (refer to Table 9.7.6.3.1)

12.3 Baseline Visit

Baseline values will be recorded from 1 to 7 days prior to the infusion visit. The following procedures will be performed during the Baseline Period:

- Brief physical examination
- Vital signs
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic FVIII assay
    - Baseline FVIII activity level – one-stage clot FVIII assay
    - hFVIII coagulation activity exploratory assay (collected but not tested prior to enrollment)
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
    - hFVIII total antibody assay (collected but not tested prior to enrollment)
    - hFVIII antigen assay (collected but not tested prior to enrollment)
- Blood sample for AAV5 Total Antibody assay
    - Baseline sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Function Tests (refer to Table 9.7.6.3.1)
- PBMC collection for CTL baseline
- Direct Thrombin test
- Blood sample for AAV5 TI assay
- Thrombin Generation Assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools

- Exploratory biomarker assessments
- Cytokine bead array assay
- Hypersensitivity blood assessments
- Haemo-QoL-A QoL assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.4 Infusion Visit/BMN 270 Infusion Visit (Day 1)

There will be one infusion visit for each subject. Subjects will remain in the clinic for at least 24 hours for the BMN 270 Infusion Visit. The following procedures will be performed during the BMN 270 Infusion Visit:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Blood sample for AAV5 TAb Assay (sample collected pre-infusion for analysis)
    - Infusion Day sample will be tested in an AAV5 Tab pre-screening assay specifically developed for enrolment purposes.
- Blood sample for AAV5 TI assay (sample collected pre-infusion for analysis)
- Fasting lipid panel (blood triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol)
- BMN 270 Infusion
- Vital Signs
    - Vital signs will be recorded prior to BMN 270 infusion and then every 15 minutes (± 5 minutes) during BMN 270 infusion. Following infusion, vital signs will be monitored every 1 hour (± 5 minutes) for at least 8 hours during the subject's stay in the clinic.

12.5 BMN 270 Infusion Follow-Up Visits

12.5.1 Week 1

During Week 1, the subject will be assessed on Study Day 2, Study Day 4, and Study Day 8.

12.5.1.1 Week 1, Study Day 2

On Study Day 2, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Function Tests (refer to Table 9.7.6.3.1)
- PCR of vector DNA in blood, saliva, urine, semen, and stools
- Blood sample for AAV5 TAb Assay (sample collected pre-infusion for analysis)
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection
- Cytokine bead array assay

12.5.1.2 Week 1, Study Day 4

On Study Day 4, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Function Tests (refer to Table 9.7.6.3.1)
- PCR of vector DNA in blood, saliva, urine, semen, and stools

12.5.1.3 Week 1, Study Day 8

On Study Day 8, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Function Tests (refer to Table 9.7.6.3.1)
- Samples for FVIII Assays
    - FVIII activity level (chromogenic FVIII assay)

- FVIII activity level (one-stage clot FVIII assay)
- FVIII coagulation activity exploratory assay
- Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level
- hFVIII antigen assay

- PCR of vector DNA in blood, saliva, urine, semen, and stools
- Cytokine bead array assay

12.5.2 Weeks 2-26

After Week 1 (Day 8), subjects will return to the study site once a week (± 48 hours) during Weeks 2-26.

12.5.2.1 Once per week (Weeks 2 through 26)

The following procedures will be performed once per week from Weeks 1 through 26:

- Brief physical examination (complete physical examination at Week 26)
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
- Vital Signs
- Liver Function Tests (refer to Table 9.7.6.3.1)
  - LFTs may be monitored more or less frequently (and in particular when ALT values are >1.5x ULN) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LFTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN..
- Samples for FVIII Assays
  - FVIII activity level (chromogenic FVIII assay)
  - FVIII activity level (one-stage clot FVIII assay)
  - FVIII coagulation activity exploratory assay
  - Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level
  - hFVIII antigen assay

12.5.2.2 Every Other Week (Weeks 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, and 26)

The following procedures will be performed every other week (at Weeks 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, and 26):

- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection

12.5.2.3 Weeks 2, 3, 4, 6, 8, 12, 16, 20, and 26

The following procedures will be performed at Weeks 2, 3, 4, 6, 8, 12, 16, 20, and 26:

- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Collection to occur until at least 3 consecutive negative results are obtained. Semen samples should continue to be collected and tested through Week 12, even if 3 consecutive negative results in that compartment have been recorded prior to that timepoint.

12.5.2.4 Weeks 2, 4, 6, 8, 10, 12, 16, 20, and 16

The following procedures will be performed at Weeks 2, 4, 6, 8, 10, 12, 16, 20, and 26:

- hFVIII total antibody assay

12.5.2.5 Weeks 2, 4, 6, 8, 14, 20, and 16

The following procedures will be performed at Weeks 2, 4, 6, 8, 14, 20, and 26:

- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)

12.5.2.6 Weeks 2, 6, 10, 14, 18, 22, and 26

The following procedures will be performed at Weeks 2, 6, 10, 14, 18, 22, and 26:

- Exploratory biomarker assessments
- Cytokine bead array assay

12.5.2.7 Weeks 4, 8, 12, 16, 20, and 26

The following procedures will be performed at Weeks 4, 8, 12, 16, 20, and 26:

- Weight
- VWF:Ag

12.5.2.8 Weeks 12 and 26

The following procedures will be performed at Weeks 12 and 26:

- Urine tests (to be performed locally)
- Haemo-QoL-A QoL assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.5.2.9 Weeks 13 and 26

The following procedure will be performed at Weeks 13 and 26:

- Direct thrombin activity test

12.5.2.10 Week 16

The following procedure will be performed at Week 16:

- Testing for reactivation of hepatitis B and hepatitis C
    - Subjects with evidence of prior exposure will be tested for hepatitis B and hepatitis C reactivation at Week 16. Subjects who receive therapeutic oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.4.

12.5.2.11 Weeks 20, 24, and 26

The following procedure will be performed at Weeks 20, 24, and 26:

- Thrombin Generation Assay

12.5.3 Post-Infusion Follow-Up – Weeks 27-260

Subjects will continue with assessments at Weeks 39, 52, 65, 78, 91, 104, 117, 130, 143, 156, 169, 182, 195, 208, 221, 234, 247, and 260. Visit windows during this period are ± 2 weeks.

12.5.3.1 Every 3 months (Weeks 39, 52, 65, 78, 91, 104, 117, 130, 143, 156, 169, 182, 195, 208, 221, 234, 247, and 260)

The following procedures will be performed every 3 months (Weeks 39, 52, 65, 78, 91, 104, 117, 130, 143, 156, 169, 182, 195, 208, 221, 234, 247, and 260) (± 2 weeks):

- Brief physical examination (complete physical examination should be done at Week 52, 104, 156, 208, and 260)
- Weight
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Vital signs
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Function Tests (refer to Table 9.7.6.3.1)
    - LFTs may be monitored more or less frequently (and in particular when ALT values are >1.5x ULN) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LFTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN..
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic FVIII assay
    - Baseline FVIII activity level – one-stage clot FVIII assay
    - hFVIII coagulation activity exploratory assay (collected but not tested prior to enrollment)
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
    - hFVIII total antibody assay (collected but not tested prior to enrollment)
    - hFVIII antigen assay (collected but not tested prior to enrollment)
- hFVIII total antibody assay
- Exploratory biomarker assessments
- Blood sample for AAV5 Total Antibody assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection for CTL baseline
- VWF:Ag

- Direct Thrombin test
- Thrombin Generation Assay

12.5.3.2 Every 12 months (Weeks 52, 104, 156, 208, and 260)

The following procedures will be performed every 12 months (Weeks 52, 104, 156, 208, and 260) (± 2 weeks):

- Haemo-QoL-A QoL assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.5.3.3 Every 4 Weeks (As Needed)

The following assessment should be performed every 4 weeks during Years 2-5, as needed:

- PCR of vector DNA in blood, saliva, urine, semen, and stool

Sample testing during Weeks 27-260 is not required if at least 3 consecutive samples are clear during the Post-Infusion Follow-Up period in Weeks 1-26. Subjects who have not had 3 consecutive negative semen samples by Week 26 should continue to have PCR testing of semen every 4 weeks until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).

12.6 Early Termination Visit

The Early Termination visit will occur on the date the subject withdraws from the study, even if the date does not correspond to a protocol-specific visit.

If a subject leaves the study prior to the Week 260 visit, the subject will be asked to return to the study site and complete an Early Termination visit. At the Early Termination visit, as many of the following assessments as possible should be done:

- Complete physical examination
- Weight
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Vital signs

- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Function Tests (refer to Table 9.7.6.3.1)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic FVIII assay
    - Baseline FVIII activity level – one-stage clot FVIII assay
    - hFVIII coagulation activity exploratory assay
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
    - hFVIII antigen assay
- hFVIII total antibody assay
- Exploratory biomarker assessments
- Blood sample for AAV5 Total Antibody assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection for CTL baseline
- VWF:Ag
- Direct Thrombin test
- Thrombin Generation Assay
- Haemo-QoL-A QoL assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire
- PCR of vector DNA in blood, saliva, urine, semen, and stool
    - Sample testing at the Early Termination Visit is not required if at least 3 consecutive samples are clear during the period of the subject's participation in the study.

12.7 End of Study

The study will end after the last subject yet to complete the last Long-Term Follow-Up visit (Week 260) does so, has transferred to another BMN 270 study, is withdrawn from the study, or discontinues from the study. BioMarin reserves the right to discontinue the study any time for clinical or administrative reasons and to discontinue participation of an individual Investigator or site for clinical or administrative reasons, including, but not limited to, poor enrollment or noncompliance with procedures of the protocol or GCP. In addition, the study may be terminated if, in the opinion of BioMarin, the safety of the study subjects may be compromised.

13 DATA QUALITY ASSURANCE

BioMarin personnel or designees will visit the study site prior to initiation of the study to review with the site personnel information about the IP, protocol and other regulatory document requirements, any applicable randomization procedures, source document requirements, CRFs, monitoring requirements, and procedures for reporting AEs, including SAEs.

At visits during and after the study, a CRA will monitor the site for compliance with regulatory documentation, with a focus on accurate and complete recording of data on CRFs from source documents, adherence to protocol, randomization (if applicable), SAE reporting and drug accountability records.

Sites will enter study data into eCRFs into the study EDC system. Data Quality Control will be performed by BioMarin Clinical Data Management or designee through implementation of quality control checks specified in the study data management plan and edit check specifications.

14 STATISTICAL METHODS AND DETERMINATION OF SAMPLE SIZE

14.1 Statistical and Analytical Plans

The statistical analysis plan (SAP) will provide additional details on the planned statistical analysis. Unless otherwise stated, all analyses will be performed using SAS.

14.1.1 Interim Analyses

No interim analysis is planned.

14.1.2 Procedures for Accounting for Missing, Unused and Spurious Data

Because the completeness of the data affects the integrity and accuracy of the final study analysis, every effort should be made to ensure complete, accurate, and timely data collection and, therefore, avoid missing data.

Sensitivity analyses will be conducted to assess the impact of missing data on the primary efficacy endpoint analysis. Additional details regarding the handling of missing data will be provided in the SAP.

14.2 Efficacy Analysis

The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using longitudinal methods, such as mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at $\geq$ 5 IU/dL at 26 weeks following study treatment. FVIII activity will be assessed weekly during the study period. We can only assess the true steady state of FVIII produced from BMN 270 after a minimum of 72 hours (or 5x the known half-life of the FVIII replacement therapy administered) has elapsed since the last infusion of FVIII concentrates.

14.3 Safety Analysis

The Medical Dictionary for Regulatory Activities terminology (MedDRA) will be used by the Sponsor to assign system organ class and preferred term classification to events and diseases, based on the original terms entered on the eCRF.

All AEs will be coded using the current version of MedDRA. The incidence of AEs will be summarized by system organ class, preferred term, relationship to study drug, and severity. A by-subject listing will be provided for those subjects who experience a serious AE (SAE),

including death, or experience an AE associated with early withdrawal from the study or study drug.

Clinical laboratory data will be summarized by the type of laboratory test. For each clinical laboratory test, descriptive statistics will be provided on Baseline as well as all subsequent visits. Descriptive statistics for physical examination results and vital signs will also be provided.

Analysis of neutralizing antibody response and other immunological parameters as well as vector shedding will be primarily descriptive and involve both inter-subject and intra-subject comparisons across doses.

Detailed statistical methods will be provided in the SAP.

14.4 Determination of Sample Size

The sample size is based upon clinical considerations and is sufficient to detect a strong clinical efficacy signal. Approximately 10 subjects may be dosed in the study.

14.5 Analysis Populations

The efficacy analysis set will be comprised of all subjects who have received the BMN 270 infusion.

The safety population will consist of all subjects who receive BMN 270 infusion during the study.

14.6 Changes in the Conduct of the Study or Planned Analyses

Only BioMarin may modify the protocol. Any change in study conduct considered necessary by the Investigator will be made only after consultation with BioMarin, who will then issue a formal protocol amendment to implement the change. The only exception is when an Investigator considers that a subject's safety is compromised without immediate action. In these circumstances, immediate approval of the chairman of the IRB/EC must be sought, and the Investigator should inform BioMarin and the full IRB/EC within 2 working days after the emergency occurs.

With the exception of minor administrative or typographical changes, the IRB/EC must review and approve all protocol amendments. Protocol amendments that have an impact on subject risk or the study objectives, or require revision of the ICF, must receive approval from the IRB/EC prior to their implementation.

When a protocol amendment substantially alters the study design or the potential risks or burden to subjects, the ICF will be amended and approved by BioMarin and the IRB/EC, and all active subjects must again provide informed consent.

15 DATA REVIEW BOARD

The Data Review Board (DRB) will consist of the 2 Principal Investigators and Sponsor's Medical Monitor, as well as hemophilia expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201. The DRB will review available safety and efficacy data during the study on an ongoing basis to determine, based on emerging data and the benefit/risk profile, whether further enrollment should continue. The DRB will meet weekly until all subjects have completed Week 26, and then monthly thereafter.

Duties of the DRB include:

- Conducting an ongoing review of individual subject safety and efficacy data during the study;
- Recommending whether to proceed with enrollment of subjects at a different gating schedules based on emerging data from 270-203 and the overall risk/benefit analysis of BMN 270;
- If applicable, considering whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction.
- Making other recommendations on the conduct and reporting of the trial based on their evaluation of clinical data including institution of any pause or stopping stages.

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DRB determination of safety and efficacy in treated subjects with the following triggers for pausing further enrollment:

- any related SAE;
- any related AE with a severity > Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

16 COSTS, COMPENSATION, AND SUBJECT INJURY

There will be no charge to study subjects to be in this study. BioMarin will pay all costs of tests, procedures, and treatments that are part of this study. In addition, after IRB/EC approval, BioMarin may reimburse the reasonable cost of travel for study-related visits in accordance with BioMarin's travel and reimbursement policy. BioMarin will not pay for any hospitalizations, tests, or treatments for medical problems of any sort related solely to the study subject's disease. Costs associated with such hospitalizations, tests, and treatments should be billed and collected in the way that such costs are usually billed and collected outside the study.

The Investigator should contact BioMarin immediately upon notification that a study subject has been injured by the study drug or by procedures performed as part of the study. Any subject who experiences a study-related injury should be instructed by the Investigator to seek immediate medical treatment at a pre-specified medical institution if possible, or at the closest medical treatment facility if necessary. The subject should be given the name of a person to contact to seek further information about, and assistance with, treatment for study-related injuries. The treating physician should bill the subject's health insurance company or other third party payer for the cost of this medical treatment. If the cost of the medical treatment is not covered by health insurance or another third party that usually pays these costs, then either BioMarin or the institution may pay for reasonable and necessary medical services to treat the injuries caused by the study drug or study procedures. In some jurisdictions, BioMarin is obligated by law to pay for study-related injuries without prior recourse to third party payer billing and/or regardless of fault. If this is the case, BioMarin will comply with the law.

17 CASE REPORT FORMS AND SOURCE DOCUMENTS

Electronic case report forms will be provided for each subject. The Investigator must review and electronically sign the completed eCRF casebook to verify its accuracy.

eCRFs must be completed using a web–based application developed and validated. Study site personnel will be trained on the application and will enter the clinical data from source documentation. Unless explicitly allowed in the eCRF instructions, blank data fields are not acceptable.

In the event of an entry error, or if new information becomes available, the value will be corrected by deselecting the erroneous response and then selecting or entering the factual response. In compliance with ICH GCP Guidelines and 21 CFR Part 11, the system will require the personnel making the correction to enter a reason for changing the value. The documented audit trail will include the reason for the change, the original value, the new value, the time of the correction and the identity of the operator.

BioMarin's policy is that study data on the eCRFs must be verifiable to the source data, which necessitates access to all original recordings, laboratory reports, and subject records. In addition, all source data should be attributable (signed and dated). The Investigator must therefore agree to allow direct access to all source data. Subjects must also allow access to their medical records, and subjects will be informed of this and will confirm their agreement when giving informed consent. If direct source document verification of study data by the site monitor is prohibited by institutional policy or local law, then the Investigator must make available facilities and/or personnel to allow GCP-compliant source verification to occur. Examples of such methods include certified copies of records which have study data visible but sensitive information redacted, or other GCP-compliant means agreed between the Investigator and the Sponsor.

A site monitor designated by BioMarin will compare the eCRFs with the original source documents at the study site and evaluate them for completeness and accuracy before designating them as "Source Data Verified" (SDV). If an error is discovered at any time or a clarification is needed, the site monitor, or designee, will create an electronic query on the associated field. Site personnel will then answer the query by either correcting the data or responding to the query. The site monitor will then review the response and determine either to close the query or re–query the site if the response does not fully address the question. This process is repeated until all open queries have been answered and closed.

Before a subject's eCRF casebook can be locked, data fields must be source data verified and all queries closed. Refer to the Study Monitoring Plan for details on which fields must be source data verified. The Investigator will then electronically sign the casebook, specifying that the information on the eCRFs is accurate and complete. The Data Manager, or designee, will then set the status of the forms, visits, and the entire casebook to Locked. Upon completion of the CSR, an electronic copy of each site's casebooks will be copied to a compact disk (CD) and sent to each site for retention with other study documents.

18 STUDY MONITORING AND AUDITING

Qualified individuals designated by BioMarin will monitor all aspects of the study according to GCP and standard operating procedures for compliance with applicable government regulations. The Investigator agrees to allow these monitors direct access to the clinical supplies, dispensing, and storage areas and to the clinical files, including original medical records, of the study subjects, and, if requested, agrees to assist the monitors. The Investigator and staff are responsible for being present or available for consultation during routinely scheduled site visits conducted by BioMarin or its designees.

Members of BioMarin's GCP Compliance Department or designees may conduct an audit of a clinical site at any time before, during, or after completion of the study. The Investigator will be informed if an audit is to take place and advised as to the scope of the audit. Representatives of the FDA or other Regulatory Agencies may also conduct an audit of the study. If informed of such an inspection, the Investigator should notify BioMarin immediately. The Investigator will ensure that the auditors have access to the clinical supplies, study site facilities, original source documentation, and all study files.

19 RETENTION OF RECORDS

The Investigator must retain all study records required by BioMarin and by the applicable regulations in a secure and safe facility. The Investigator must consult a BioMarin representative before disposal of any study records, and must notify BioMarin of any change in the location, disposition or custody of the study files. The Investigator /institution must take measures to prevent accidental or premature destruction of essential documents, that is, documents that individually and collectively permit evaluation of the conduct of a study and the quality of the data produced, including paper copies of study records (eg, subject charts) as well as any original source documents that are electronic as required by applicable regulatory requirements.

All study records must be retained for at least 2 years after the last approval of a marketing application in the US or an ICH region and until (1) there are no pending or contemplated marketing applications in the U.S. or an ICH region or (2) at least 2 years have elapsed since the formal discontinuation of clinical development of the IP. The Investigator /institution should retain subject identifiers for at least 15 years after the completion or discontinuation of the study. Subject files and other source data must be kept for the maximum period of time permitted by the hospital, institution or private practice, but not less than 15 years. These documents should be retained for a longer period, however, if required by the applicable regulatory requirements or by a BioMarin agreement. BioMarin must be notified and will assist with retention should Investigator /institution be unable to continue maintenance of subject files for the full 15 years. It is the responsibility of BioMarin to inform the Investigator /institution as to when these documents no longer need to be retained.

20 USE OF INFORMATION AND PUBLICATION

BioMarin recognizes the importance of communicating medical study data and therefore encourages the publication of these data in reputable, peer-reviewed scientific journals and at seminars or conferences. The details of the processes of producing and reviewing reports, manuscripts, and presentations based on the data from this study will be described in the Clinical Trial Agreement between BioMarin and the Investigator/Institution. Consideration for authorship of all publications will be based on compliance with the Uniform Requirements for Manuscripts Submitted to Biomedical Journals ("Uniform Requirements") of the International Committee of Medical Journal Editors (ICMJE) (http://www.icmje.org/ethical_1author.html) and good publication practices (GPP).

21 REFERENCES

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22 INVESTIGATOR RESPONSIBILITIES

22.1 Conduct of Study and Protection of Human Subjects

In accordance with FDA Form 1572 and/or principles of ICH E6[R2] GCP, the Investigator will ensure that:

- He or she will conduct the study in accordance with the relevant, current protocol and will only make changes in a protocol after notifying the sponsor, except when necessary to protect the safety, rights, or welfare of subjects.
- He or she will personally conduct or supervise the study.
- He or she will inform any potential subjects, or any persons used as controls, that the drugs are being used for investigational purposes, and he or she will ensure that the requirements relating to obtaining informed consent in 21 CFR Part 50 and/or ICH Sections 2.9 and 4.8 are met. As well, he or she will ensure that IRB/EC review and approval in 21 CFR Part 56 and/or ICH E6[R2] Section 2.6 are met.
- He or she will report to the sponsor adverse experiences that occur in the course of the investigation in accordance with 21 CFR 312.64 and/or ICH E6[R2] Section 4.11.
- He or she has read and understands the information in the Investigator's Brochure, including potential risks and side effects of the drug.
- His or her staff and all persons who assist in the conduct of the study are informed about their obligations in meeting the above commitments
- Adequate and accurate records in accordance with 21 CFR 312.62 and/or ICH E6[R2] Section 4.9 are kept, and those records are available for inspection in accordance with 21 CFR 312.68 and/or ICH E6[R2] Section 4.9.7.
- The IRB/EC complies with the requirements of 21 CFR Part 56, ICH E6[R2] Section 3.0, and other applicable regulations, and conducts initial and ongoing reviews and approvals of the study. He or she will also ensure that any change in research activity and all problems involving risks to human subjects or others are reported to the IRB/EC. Additionally, he or she will not make any changes in the research without IRB/EC approval, except where necessary to eliminate apparent immediate hazards to human subjects.
- He or she agrees to comply with all other requirements regarding the obligations of clinical Investigators and all other pertinent requirements in 21 CFR Part 312 and/or ICH E6[R2].

23 SIGNATURE PAGE

Protocol Title: A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5

Protocol Number: 270-203

I have read the foregoing protocol and agree to conduct this study as outlined. I agree to conduct the study in compliance with all applicable regulations and guidelines, including E6R2 ICH, as stated in the protocol, and other information supplied to me.

Investigator Signature Date

Printed name: _____

Accepted for the Sponsor:

PI ıre PI Date

Printed name: PI MD, PI Clinical Science

BIOMARIN®

CLINICAL STUDY PROTOCOL

Study Title:	A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5
Protocol Number:	270-203
Active Investigational Product:	AAV5-hFVIII-SQ
European Union Drug Regulating Authorities Clinical Trials (EudraCT) Number:	2017-000662-29
Indication:	Hemophilia A
Sponsor:	BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949
Development Phase:	Phase 1/2
Sponsor's Responsible Medical Monitor:	PI MA MB BChir MSc BioMarin (UK) Ltd. 10 Bloomsbury Way London, WC1A 2SL
Study Design:	Single-arm, open-label
Duration of Subject Participation:	Up to 264 weeks
Dose:	6E13 vg/kg as single infusion
Study Population:	Males ≥ 18 years of age with severe hemophilia A and detectable pre-existing antibodies against AAV5 vector capsid
Date of Original Protocol:	29 September 2017
Date of Amendment 1:	5 October 2018

CLINICAL STUDY PROTOCOL AMENDMENT SUMMARY

Amendment 1

Date: 5 October 2018

RATIONALE AND SUMMARY OF CHANGES

A summary of major changes covered by Amendment 1 to the 270-203 protocol is provided below:

1. Language concerning the occurrence and management of infusion-related reactions has been added.

Rationale: Three subjects experienced medically important infusion-related reactions (all grade 2) associated with valoctocogene roxaparvovec administration that were reported as serious adverse events due to prolonged observation in the hospital. Two subjects were enrolled in Study 270-301 (Phase 3 open-label single-arm study in subjects with severe hemophilia A receiving the 6E13 vg/kg dose of BMN 270), and one subject was enrolled in 270-203. All of these infusion-related reactions were effectively managed clinically and resolved without any clinical sequelae.

The initial infusion rate will be changed from 4 mL/min to 1 mL/min and should be increased every 30 minutes by 1 mL/min to a maximum of 4 mL/min, provided that the subject is asymptomatic and tolerates the infusion. Adjustment/interruption of the infusion rate and/or duration may be required in the event of an adverse reaction occurring during the infusion. If necessary, anti-histamine, anti-pyretic, and/or corticosteroid administration is permitted prior to restarting an infusion interrupted by an infusion-related reaction.

To better elucidate the mechanisms of infusion-related hypersensitivity reactions, exploratory biomarker plasma samples collected at Baseline and at post-infusion study visits may also be used to assess the magnitude of biomarker changes. Samples will be used to assess acute phase response reactants, complement activation, inflammatory biomarkers, and IgE.

2. HIV-positive patients are now excluded from the study.

Rationale: An HIV-positive subject in Study 270-302 developed markedly elevated transaminase levels after receiving 4E13 vg/kg of BMN 270. The subject was receiving HAART treatment for his HIV infection, and it is hypothesized that the combination of BMN 270, one or more of his HAART medications, and/or unsuspected underlying hepatic disease may have contributed to the subject's elevated transaminase levels. Out of an abundance of caution for the long-term liver health of HIV-positive patients who are

receiving HAART and may be interested in receiving gene therapy, further enrollment of HIV-positive subjects will be suspended in 270-203.

3. Efavirenz, lamivudine, and experimental hemophilia treatments (emicizumab, fitusiran, and concizumab) have been added to the list of prohibited concomitant medications.

Rationale: The subject in 270-302 referenced above was receiving efavirenz and lamivudine as part of his HAART regimen, and these are considered the most potentially likely medications to have contributed to his elevated transaminase levels. Experimental hemophilia treatments are prohibited during the study as they could affect the assessment of FVIII levels in 270-203 subjects.

4. The exclusion criterion concerning liver test levels at Screening have been changed to require all assessed liver tests (ie, alanine aminotransferase, aspartate aminotransferase, alkaline phosphatase, gamma-glutamyltransferase, and total bilirubin) to be no higher than the 1.25 times the upper limit of normal for eligibility purposes.

Rationale: A subject with Gilbert's syndrome in Study 270-301 developed elevated transaminase levels after receiving 6E13 vg/kg of BMN 270. Although other subjects with Gilbert's syndrome or HIV infection have been safely treated in small numbers in BMN 270 clinical trials to-date, the purpose of this amendment is to focus development excluding groups of patients hypothesized to have potential risk factors for interactions with BMN 270. Restricting the range of acceptable baseline liver test levels will help ensure that entering subjects have the best chance to tolerate BMN 270 without experiencing potential hepatocellular injury subsequently.

5. The visit schedule after Week 26 has been expanded.

Rationale: The original protocol required subjects to return once every 3 months between Week 26 and Year 5. The expanded schedule is in line with the visit schedules used in other BMN 270 treatment studies (270-301, 270-302) and will allow for more granular assessment of BMN 270 efficacy and monitoring of subject safety.

6. An abbreviated visit schedule has been made available after Week 26 or after Week 52 for subjects who are considered to have not responded to BMN 270 therapy.

Rationale: Treatment failure—manifesting as either failure to achieve FVIII activity ≥5 IU/dL by Week 26/Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator—may, at the Investigator's discretion and after discussion with the Medical

Monitor, enable subjects to follow an abbreviated visit schedule after Week 26 or Week 52 of the study, as applicable and at the discretion of the Investigator.

Subjects who meet the "treatment failure" definition and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment according to the Schedules of Assessments until vector shedding has cleared.

7. An additional fasting serum sample will be drawn on Day 1, in case it is needed for future exploratory assessments.

Rationale: The additional sample will enable the conduct of exploratory assessments prior to BMN 270 infusion.

8. Additional details have been included concerning information to be collected as part of the medical history assessment at Screening.

Rationale: In subjects with a history of hepatitis B or hepatitis C infection, information on the specific treatments received for that infection should be collected. In addition, any previous pharmacokinetic data collected at the time the subject was receiving on-demand or prophylactic FVIII treatment should be collected. This additional history data could help explain a subject's FVIII response (or lack of FVIII response) to BMN 270.

9. Language in the inclusion criterion related to a subject's history of FVIII inhibitors has been clarified.

Rationale: The original language caused some issues of misinterpretation of Sponsor intent. The revised language removes the ambiguity.

10. Language concerning when to consider restarting FVIII prophylaxis following BMN 270 infusion has been modified.

Rationale: The decision for reinstitution of FVIII prophylaxis should be based on clinical grounds (eg, the advent of bleeding episodes), in consultation with the Medical Monitor. The need to reinstitute FVIII prophylaxis if the FVIII activity is below a certain level post-BMN 270 has not yet been clinically established but will be informed by results from this study.

11. An additional criterion to initiate therapeutic oral corticosteroids for elevated alanine aminotransferase (ALT) levels, following consultation with the Medical Monitor, of ALT > ULN and > 2x baseline value has been added.

Rationale: The addition will provide flexibility to initiate therapeutic oral corticosteroids, following consultation with the Medical Monitor, in a timely manner with respect to elevations in ALT levels.

12. Language has been added to include ABO testing at Baseline.

Rationale: ABO blood group results will be collected at Baseline to potentially correlate with FVIII activity level results in an exploratory manner. For subjects already enrolled in the study, ABO blood group results should be collected at the next regularly scheduled visit (or at least prior to the end of the study).

13. Clarified that the requirement for contraception use can end as early as Week 12, in the case that a subject has had 3 consecutive negative semen vector shedding assessments prior to that time point.

Rationale: Language in the protocol that stated that all subjects must continue with contraception for at least 26 weeks after infusion was inconsistent with the statement that semen sampling could be stopped as early as Week 12 if a subject had had 3 consecutive negative assessments. Revised language makes it clear that all subjects must remain on contraception and provide semen samples for assessment through Week 12, but after that timepoint, both contraception and semen sample assessment can be discontinued once 3 consecutive negative results have been obtained.

14. Added language to clarify that local laboratory testing is permitted at the Investigator's discretion when required to make clinical management decisions. Where possible, a matched sample for testing at the central laboratory should be taken at the same time.

Rationale: Local laboratory test results may be available more quickly compared with the central laboratory therefore to avoid potential delays in clinical management decisions, a local test may be performed at the Investigator's discretion. To provide a complete dataset of central laboratory results for reporting purposes, where possible, a matched sample for the central laboratory should also be taken from the subject at the same time as the local sample.

15. Added language that subjects will fast for at least 8 hours prior to collection of pre-infusion laboratory samples on the day of infusion.

Rationale: A fasting lipid panel is part of the infusion day laboratory assessment; as such, subjects need to fast for an adequate amount of time prior to the assessment to allow for accurate results.

16. Assessment of dermatologic and musculoskeletal systems has been added to the brief physical examination.

Rationale: Both organ systems are routinely assessed as part of comprehensive hemophilia care visits.

17. Testing of the exploratory samples for the Direct Thrombin Activity Test and TGA Assay has been clarified as optional.

Rationale: While exploratory samples for Direct Thrombin Activity Test and TGA Assay will be collected at the time points indicated in the protocol, analysis of these samples will be optional.

18. Specific testing of TNF-α and IL10a single nucleotide polymorphisms has been removed from biomarker testing.

Rationale: These biomarkers are designed to inform the risk of FVIII inhibitor development. There have not been any cases of FVIII inhibitor development observed in Study 270-201, so analyses of TNF-α and IL10a single nucleotide polymorphisms have not been indicated. FVIII inhibitors are not expected to develop in this study; however, in the event that inhibitors are observed, sufficient exploratory samples will have been collected to perform these tests.

19. The initial von Willebrand Factor antigen (VWF:Ag) assessment has been moved from the Screening visit to the Baseline visit.

Rationale: This was moved to only evaluate this laboratory test in individuals who have been deemed eligible to receive BMN 270.

20. It has been clarified that post-steroid testing for hepatitis B/C reactivation should be performed only in subjects who have a previous history of positive hepatitis B/C tests.

Rationale: The previous language in the protocol was unclear and may have led some investigators to think all subjects (including those with no prior evidence of viral hepatitis exposure) should be retested.

21. Subjects will be advised to abstain from blood or sperm donation after BMN 270 infusion until there is no further evidence of vector shedding.

Rationale: Vector shedding in the blood and semen following BMN 270 infusion has been observed, which could serve for vector transmission if subjects were to donate blood or semen following the BMN 270 infusion.

22. In the event of a positive Bethesda assay result during Years 3-5, an additional sample has been added to be collected within 4 weeks of the visit where the positive result was obtained.

Rationale: During Years 3-5, samples are being regularly collected every 6 weeks. However, to align with EMA guidance (which suggests that a confirmatory test on a second sample should be done within a month after a positive Bethesda assay result), language has been added to require the site to conduct an unscheduled visit within 4 weeks after the date when the positive Bethesda result was obtained.

23. Visit windows (±24 hours) have been added to the Week 1 visits (Day 2, Day 4, and Day 8).

Rationale: Visit windows will ease site and subject burden.

24. Testing of AAV5 antibody titers has been added to the Day 8 visit.
25. Clarified that hepatitis B testing at Screening should include hepatitis B surface antigen (HBsAg), hepatitis B surface antibody (HBsAb), and hepatitis B core antibody (HBcAb) testing.
26. The vector genome schematic figure has been updated.
27. The identity of the Medical Monitor has been updated.
28. Changes have been made to correct minor errors and for purposes of clarity and consistency.

Refer to Section 25 for a summary of revisions to the original protocol (dated 29 September 2017).

2 SYNOPSIS

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

TITLE OF STUDY:

A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5

PROTOCOL NUMBER:

270-203

STUDY SITES:

Approximately 2-3 sites in the United Kingdom

PHASE OF DEVELOPMENT:

Phase 1/2

STUDY RATIONALE:

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. The clinical phenotype of HA patients generally correlates tightly with the level of residual expression. Severe HA is classified as FVIII activity less than 1% of wild-type (< 1 IU/dL), moderate disease comprises 1-5% of wild-type activity, and the mild form is 5-40% activity. The clinical manifestations of severe HA are frequent spontaneous bleeding episodes, predominantly in joints and soft tissues, with a substantially increased risk of death from hemorrhage when the brain is involved.

Treatment of severe HA presently consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study (Berntorp, 2017) and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with on-demand-only therapy in the same studies, respectively. Continued repeated bleeding events compound

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death.

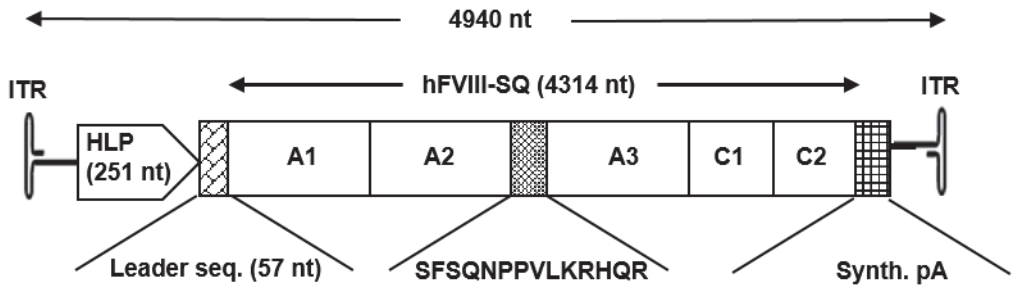
Chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) and bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half-life to maximum of 18 hours, leaving critical periods when FVIII activity levels are below the therapeutic range and leaving patients vulnerable to bleeding and concomitant sequelae. As such, despite currently available FVIII replacement therapies, a high unmet need remains for maintenance of a sustained, consistent FVIII level compatible with a normal and hemorrhage-free life.

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of active FVIII following a single intravenous infusion of a vector encoding the appropriate gene sequence. Hemophilia A is well-suited for a gene replacement approach because clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in minute amounts (200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and even modest increases in the level of FVIII (any increase of the plasma level by 2 ng/ml induces an increase in activity of 1%) can ameliorate the severe form of hemophilia A. Thus, relatively small changes in endogenous FVIII activity can result in clinically relevant improvements in disease phenotype. Moreover, the circulating FVIII response to gene transduction can be assessed using validated quantitative endpoints that are easily assayed using established laboratory techniques.

Several different gene transfer strategies for FVIII replacement have been evaluated, with adeno-associated viral (AAV) vectors used in most hemophilia clinical trials. AAV vectors have an excellent and well-defined safety profile, and can direct long-term transgene expression with tropism and promoter specificity for specific tissues, such as the liver (eg serotypes 2, 5, and 8). Indeed, an ongoing gene therapy clinical trial for a related disorder, hemophilia B, has established over 7 years of stable human factor IX (hFIX) expression at levels sufficient for conversion of their bleeding phenotype from severe to moderate or mild, following a single peripheral vein infusion of AAV8-hFIX vector (Nathwani, 2014). Several participants in this trial have been able to discontinue factor prophylaxis without suffering spontaneous hemorrhages, even when they undertook activities that previously resulted in bleeding. Thus, gene therapy treatment has resulted in a substantial improvement in their quality of life.

BMN 270 is an AAV5-based gene therapy vector that expresses the SQ form of hFVIII under the control of a hybrid human liver-specific promoter (Figure 1).

Figure 1. hFVIII-SQ Vector Genome Schematic



Legend –Note that schematic is not to scale; nt = nucleotides

BMN 270 is delivered by a single IV dose and is designed to achieve stable, potentially life-long expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.

BMN 270 is being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. Preliminary results from 270-201 have demonstrated that following gene transfer, sustained FVIII activity above 15% (15 IU/dL) up to a year's observation is achievable with a dose of 4-6E13 vg/kg with an acceptable safety profile (Pasi, 2017).

Subjects enrolled and infused in 270-201 were screened and negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes.

Current data describing the extent of neutralization or inhibited transduction has been inconsistent across studies, each employing one of several varying methodologies to evaluate both vector-specific antibodies and transduction inhibition (TI). Early studies using AAV2 for the treatment of hemophilia B described inhibited transduction in patients with neutralizing antibody titers of 2 or 3 (Manno, 2006), but more recent studies have described successful transduction in patients who, in retrospect, were determined to have pre-existing antibody to the vector capsid (Majowicz, 2017). Importantly, no drug-related adverse events were described in these patients. As the sensitivity of assays used to detect pre-existing antibody has increased greatly in recent years, the biological relevance of any given titer determination, and comparisons across studies using assays with differing titer sensitivities, requires additional study.

Data from a non-clinical study conducted by BioMarin in cynomolgus monkeys further demonstrated that transduction efficiency can vary greatly in the presence of pre-existing anti-AAV5 antibody. In non-human primates, the ability to attain FVIII levels in the face of measurable AAV5 antibody titers without significant safety risks supports the possibility of achieving a similar response in patients with AAV5 antibody positivity. The totality of available

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

data shows the effect of pre-existing immunogenicity to AVV delivery vectors is not completely understood, and indicates that some patients may benefit from AAV5 gene therapy who are currently excluded.

In this study, patients with severe HA and pre-existing antibodies against AAV5 will be enrolled to evaluate the clinical response to BMN 270 gene therapy. The aim is to evaluate the safety, tolerability, and efficacy of BMN 270 in patients with severe HA and pre-existing antibodies against AAV5 vector capsid at various levels of AAV5 antibody titers.

OBJECTIVES:

The primary objective of the study is to:

- Assess the safety of a single intravenous administration of BMN 270 in severe HA subjects with pre-existing antibody to AAV5 vector capsid, including development of FVIII neutralizing antibody

Secondary objectives of the study are to:

- Assess the efficacy of BMN 270 defined as FVIII activity at or above 5 IU/dL at Week 26
- Assess the impact of BMN 270 on usage of exogenous FVIII replacement therapy
- Assess the impact of BMN 270 on the number of bleeding episodes requiring exogenous FVIII therapy
- Evaluate the pharmacodynamics of FVIII expression following IV infusion of BMN 270
- Assess the impact of BMN 270 on patient-reported outcomes (PROs)

STUDY DESIGN AND PLAN:

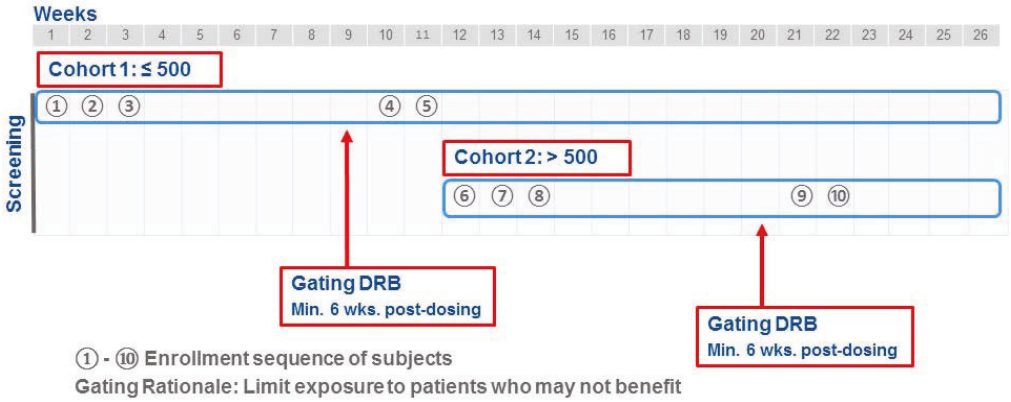
This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to the AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects may be enrolled at 2-3 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb titer ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb titer > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date.

The Data Review Board (DRB) will consist of the Principal Investigators and Sponsor's Medical Monitor, as well as hemophilia expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201. The DRB will review available safety and efficacy data

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

throughout the study and provide recommendations based on their review (Figure 2 represents one dosing schedule scenario):

Figure 2: 270-203 Dosing Schedule (One Possible Scenario)



Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from a minimum of 6 weeks post-infusion) reviewed. FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response, based on data from 270-201. Up to 6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness).

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DRB evaluation of safety and efficacy in treated subjects, with the following triggers that may potentially pause further enrollment:

- any related SAE;
- any related AE with a severity > CTCAE Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

Following a temporary halt of enrolment, the DRB may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed. Additional blood samples will be collected (tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine bead array (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.

Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DRB based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL. In subjects who experience recurring bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis.

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after either Week 26 or Week 52, as applicable and at the discretion of the Investigator.

The study analysis will be performed after all subjects have been followed for 26 weeks post-BMN 270 infusion, with presentation of safety, efficacy, and FVIII PD assessments. After the safety, efficacy, and FVIII PD analyses at 26 weeks post-BMN 270 infusion, long-term safety and efficacy will be assessed in all subjects for up to a total of 5 years post-infusion. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the DRB.

NUMBER OF SUBJECTS PLANNED:

Approximately 10 subjects

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

DIAGNOSIS AND ALL CRITERIA FOR INCLUSION AND EXCLUSION:

Individuals eligible to participate in this study must meet all of the following criteria:

1. Males ≥ 18 years of age with hemophilia A and residual FVIII levels ≤ 1 IU/dL as evidenced by medical history, at the time of signing the informed consent.
2. Detectable pre-existing antibodies against the AAV5 vector capsid as measured by AAV5 total antibody ELISA
3. Treated/exposed to FVIII concentrates or cryoprecipitate for a minimum of 150 exposure days (EDs)
4. Subject must have been on prophylactic FVIII replacement therapy for at least 12 months prior to study entry.
5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures.
6. No previous documented history of a detectable FVIII inhibitor, and results from a Bethesda assay or Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) (or less than 1.0 BU for laboratories with a historical lower sensitivity cutoff for inhibitor detection of 1.0 BU) on 2 consecutive occasions at least one week apart within the past 12 months (at least one of which should be tested at the central laboratory)
7. Sexually active participants must agree to use an acceptable method of effective contraception, either double-barrier contraception (ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with no detectable viral vector DNA.
8. Willing to abstain from consumption of alcohol for at least the first 26 weeks following BMN 270 infusion.

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Any evidence of active infection or any immunosuppressive disorder, including HIV infection.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

2. Significant liver dysfunction with any of the following abnormal laboratory results:
    - ALT (alanine aminotransferase) > 1.25x ULN;
    - AST (aspartate aminotransferase) > 1.25x ULN;
    - GGT (gamma-glutamyltransferase) > 1.25x ULN;
    - Total bilirubin > 1.25x ULN;
    - Alkaline phosphatase > 1.25x ULN; or
    - INR (international normalized ratio) ≥ 1.4

    Subjects whose liver laboratory assessments fall outside of these ranges may undergo repeat testing of the entire liver test panel within the same Screening window and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor.
3. Prior liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used
4. Evidence of any bleeding disorder not related to hemophilia A
5. Platelet count of < 100 x 10^9/L
6. Creatinine ≥ 1.5 mg/dL
7. Liver cirrhosis of any etiology as assessed by liver ultrasound
8. Chronic or active hepatitis B as evidenced by positive serology testing (hepatitis B surface antigen [HBsAg], hepatitis B surface antibody [HBsAb], and hepatitis B core antibody [HBcAb]) and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results .
9. Active Hepatitis C as evidenced by detectable HCV RNA, or currently on antiviral therapy
10. Active malignancy, except non-melanoma skin cancer
11. History of hepatic malignancy
12. History of arterial or venous thromboembolic events (eg, deep vein thrombosis, non-hemorrhagic stroke, pulmonary embolism, myocardial infarction, arterial embolus), with the exception of catheter-associated thrombosis for which anti-thrombotic treatment is not currently ongoing.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

13. Known inherited or acquired thrombophilia, including conditions associated with increased thromboembolic risk, such as atrial fibrillation.
14. A history of known inflammatory, connective tissue, or autoimmune disorders (eg, vasculitis).
15. Treatment with any Investigational Product within 30 days or 5 half-lives of the investigational product (whichever is longer) prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study
16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including possible corticosteroid treatment outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.
17. Prior treatment with any vector or gene transfer agent
18. Major surgery planned in the 26-week period following the infusion with BMN 270
19. Use of systemic immunosuppressive agents, not including corticosteroids, or live vaccines within 30 days before the BMN 270 infusion
20. Concurrent enrollment in another clinical study, unless it is an observational (non-interventional) clinical study that does not interfere with the requirements of the current protocol or have the potential to impact the evaluation of safety and efficacy of BMN 270 and with prior consultation with the Medical Monitor
21. Known allergy or hypersensitivity to investigational product formulation
22. Unwilling to receive blood or blood products for treatment of an adverse event and/or a bleed

INVESTIGATIONAL PRODUCT(S), DOSE, ROUTE AND REGIMEN:
Each subject will receive a single IV infusion of BMN 270 at 6E13 vg/kg. The volume of infusion will depend on the subject's weight.

REFERENCE THERAPY(IES), DOSE, ROUTE AND REGIMEN:
No reference therapy will be evaluated in this study.

DURATION OF TREATMENT:
BMN 270 is given as a single dose by IV infusion.

CRITERIA FOR EVALUATION:

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

Safety:

The following safety outcome measurements will be assessed:

- Incidence of adverse events (AEs), including serious AEs (SAEs)
- Change in clinical laboratory tests (serum chemistry and hematology)
- Change in vital signs
- Change in physical examination
- Vector shedding (blood, urine, semen, feces, saliva)
- Liver tests (LTs, including ALT, AST, GGT, total bilirubin, and alkaline phosphatase)
- Immune response to FVIII transgene product and AAV5 vector capsid

No major toxicity is expected based on 270-201 data and non-clinical studies. Each subject will have comprehensive surveillance monitoring of LTs (once per week for Weeks 1-26). During the long-term safety evaluation, LTs will be monitored every three months for up to 5 years post-infusion; the frequency and duration of LT testing may be changed based on discussion between the Medical Monitor and the Investigator and review of subject data.

There will be a detailed assessment of cellular and humoral responses to AAV5 vector capsid and FVIII.

Efficacy:

The efficacy measure will be to assess plasma FVIII activity. The efficacy goal is to achieve FVIII activity ≥ 5 IU/dL at 26 weeks post-BMN 270 administration. Other efficacy measures include assessing the impact of BMN 270 on the use of FVIII replacement therapy and on the number of bleeding episodes during the study. Subjects will be asked to keep a subject diary, provided by the sponsor, to record the relevant details.

Other efficacy endpoints:

- Change from baseline in the total score of HAEMO-QoL-A at Week 26 of the study post-BMN 270 infusion
- Change from baseline in the EQ-5D-5L score at Week 26 of the study post-BMN 270 infusion.
- Change from baseline in the Haemophilia Activities List (HAL) score at Week 26 of the study post-BMN 270 infusion.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

- Change from baseline in the Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) score at Week 26 of the study post-BMN 270 infusion.

Pharmacodynamics:

The FVIII antigen and activity level, as measured by a validated immunoassay and a validated FVIII activity assay, respectively, will be used for plasma profiles; FVIII antigen and activity will be used to determine PD parameters.

STATISTICAL METHODS:

Approximately 10 subjects may be dosed in the study. The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using longitudinal methods, such as a mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at $\geq$ 5 IU/dL at 26 weeks following study treatment. FVIII activity will be assessed weekly during the study period. Assessment of the true steady state of FVIII will require that FVIII activity is measured after a minimum of 72 hours (or 5x the known half-life of the FVIII replacement therapy administered) has elapsed since the last infusion of FVIII concentrates.

Analysis of neutralizing antibody response, other immunological parameters, and vector shedding will be primarily descriptive and involve both inter-subject and intra-subject comparisons across cohorts.

3 TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

4 LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviations

AAV	adeno-associated virus
ABR	annualized bleeding rate
ADL	activities of daily living
ADR	adverse drug reaction
AE	adverse event
ALT	alanine aminotransferase
APTT	activated partial thromboplastin time
ART	anti-retroviral therapy
AST	aspartate aminotransferase
BPV	BioMarin Pharmacovigilance
BU	Bethesda Unit
CFR	Code of Federal Regulations
CRA	clinical research associate
CRF	case report form
CSR	clinical study report
CTCAE	Common Terminology Criteria for Adverse Events
DRB	Data Review Board
EC	ethics committee
eCRF	electronic case report form
ED	exposure days
EOSI	events of special interest
ETV	early termination visit
EudraCT	European Union Drug Regulating Authorities Clinical Trials
FDA	Food and Drug Administration
FIH	first-in-human
FVIII	coagulation factor VIII
GCP	Good Clinical Practice
GGT	gamma-glutamyltransferase
HA	Hemophilia A
HAL	Haemophilia Activities List
HBcAb	hepatitis B core antibody
HBsAb	hepatitis B surface antibody
HBsAg	hepatitis B surface antigen

hFIX	human coagulation factor IX
hFVIII	human coagulation factor VIII
HIPAA	Health Insurance Portability and Accountability Act
IB	investigator brochure
ICF	informed consent form
ICH	International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
ICH E6 [R2]	ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6
IND	Investigational New Drug (application)
INR	international normalized ratio
IP	investigational product
IRB	institutional review board
IV	intravenous
LT	liver test
MedDRA	Medical Dictionary for Regulatory Activities
PBMC	peripheral blood mononuclear cells
PCR	polymerase chain reaction
PD	pharmacodynamics
PEG	polyethylene glycol
PK	Pharmacokinetics
PRO	patient-reported outcome
rhFVIII	recombinant human FVIII protein
SAE	serious adverse event
SAP	statistical analysis plan
SDV	source data verification
SoA	schedule(s) of activities
TGA	thrombin generation assay
ULN	upper limit of normal
vg	vector genomes
VWF:Ag	von Willebrand factor Antigen
WPAI+CIQ:HS	Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific

Definition of Terms:

Investigational Product (IP):

"A pharmaceutical form of an active ingredient or placebo being tested or used as a reference in a clinical trial, including a product with a marketing authorization when used or assembled (formulated or packaged) in a way different from the approved form, or when used for an unapproved indication, or when used to gain further information about an approved use" (from International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6 [ICH E6] (R2)]).

The terms "IP" and "study drug" may be used interchangeably in the protocol.

5 ETHICS

BioMarin Pharmaceutical Inc. (hereafter referred to as BioMarin or the Sponsor) conducts its studies according to the highest ethical and scientific standards. The following Sections articulate standards to which Investigators will be held accountable, as well as matters of compliance to document adherence to such standards.

5.1 Institutional Review Board or Independent Ethics Committee

Investigators are expected to interact with Ethics Committees (ECs) promptly, as required, during the course of the study. This includes, but is not limited to, providing appropriate documentation to support study initiation and maintaining appropriate flow of safety and other information during the course of the study and for study close-out activities. BioMarin (or designee) will assist Investigators with access to timely and accurate information and with assurance of prompt resolution of any queries.

Prior to initiating the study, the Investigator will obtain written confirmation that the institutional review board (IRB) or independent ethics committee (EC) is properly constituted and compliant with International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) and Good Clinical Practice (GCP) requirements, applicable laws, and local regulations. A copy of the confirmation from the IRB/EC will be provided to BioMarin Pharmaceutical Inc. (BioMarin) or its designee. The Investigator will provide the IRB/EC with all appropriate material, including the protocol, Investigator's Brochure (IB), the Informed Consent Form (ICF) including compensation procedures, and any other written information provided to the subjects, including all ICFs translated for patients who do not speak the local language at the clinical site. The study will not be initiated and Investigational Product (IP) supplies will not be shipped to the site until appropriate documents from the IRB/EC confirming unconditional approval of the protocol, the ICF and all subject recruitment materials are obtained in writing by the Investigator and copies are received at BioMarin or its designee. The approval document should refer to the study by protocol title and BioMarin protocol number (if possible), identify the documents reviewed, and include the date of the review and approval. BioMarin will ensure that the appropriate reports on the progress of the study are made to the IRB/EC and BioMarin by the Investigator in accordance with applicable guidance documents and governmental regulations.

5.2 Ethical Conduct of Study

It is expected that Investigators understand and comply with the protocol. This includes, but is not limited to: establishing and meeting enrollment commitments, including providing eligible subjects for study enrollment; adhering to adverse event reporting, diagnostic, or other procedures as specified in the protocol; and assuring appropriate compliance with study treatment administration and accountability.

This study will be conducted in accordance with the following:

- European Clinical Trial Directive 2001/20/EC and Good Clinical Practice Directive 2005/28/EC, for studies conducted within any European country
- US Code of Federal Regulations (CFR) Sections that address clinical research studies, and/or other national and local regulations, as applicable
- ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6(R2) (ICH E6R2)
- The ethical principles established by the Declaration of Helsinki

Specifically, this study is based on adequately performed laboratory and animal experimentation. The study will be conducted under a protocol reviewed and approved by an IRB/EC and will be conducted by scientifically and medically qualified persons. The benefits of the study are in proportion to the risks. The rights and welfare of the subjects will be respected and the investigators conducting the study do not find the hazards to outweigh the potential benefits. Each subject will provide written, informed consent before any study-related tests or evaluations are performed.

5.3 Subject Information and Informed Consent

A properly written and executed informed consent form (ICF), in compliance with ICH E6R2 (Section 4.8), United States Code of Federal Regulations (CFR) 21 CFR §50, European Clinical Trial Directive 2001/20/EC and Good Clinical Practice Directive 2005/28/EC, and other applicable local regulations, will be obtained for each subject prior to entering the subject into the study. The Investigator will prepare the ICF and provide the documents to BioMarin for approval prior to submission to the IRB/EC approval. BioMarin and the IRB/EC must approve the documents before they are implemented. A copy of the approved ICF and if applicable, a copy of the approved subject information sheet and all ICFs translated to a language other than the native language of the clinical site must also be received by BioMarin or designee prior to any study-specific procedures being performed.

6 INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

During administration of informed consent, expectations regarding participation in the study should be made clear to subjects. Subjects who are not willing and/or are not able to comply with all aspects of the study should not be encouraged to participate.

Prior to beginning the study, the Investigator at each site must provide to BioMarin or designee a fully executed and signed Statement of Investigator (SOI) form. A US Food and Drug Administration (FDA) Form FDA 1572 serves as an acceptable SOI form. If Form FDA 1572 may not be used in a particular region, the Investigator must provide a fully executed SOI on the form provided by the Sponsor. All Investigators and Sub-Investigators must be listed on Form FDA 1572 or its equivalent SOI. Financial Disclosure Forms must also be completed for all Investigators and Sub-Investigators listed on the Form FDA 1572 or SOI who will be directly involved in the treatment or evaluation of subjects in this study.

The study will be administered by and monitored by employees or representatives of BioMarin. Clinical Research Associates (CRAs) or trained designees will monitor each site on a periodic basis and perform verification of source documentation for each subject as well as other required review processes. BioMarin's Regulatory Affairs Department (or designee) will be responsible for the timely reporting of serious adverse events (SAEs) to appropriate regulatory authorities as required.

In multicenter studies, a Coordinating Investigator will be identified who will be responsible for study overview. The Coordinating Investigator will read the clinical study report (CSR) and confirm that it accurately describes the conduct and results of the study, to the best of his or her knowledge. The Coordinating Investigator will be chosen on the basis of active participation in the study, ability to interpret data, and willingness to review and sign the report in a specified timeframe. The identity of the Coordinating Investigator and a list of all Investigators participating in the study will be provided in the CSR.

Clinical Laboratory assessments will be performed at a nominated central laboratory. Bioanalytical samples will be sent to the appropriate specialty laboratories for testing. Refer to laboratory manual for more details.

7 INTRODUCTION

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males (Nathwani, 1992). It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. Clinical manifestations of severe FVIII deficiency are frequent unprovoked bleeding episodes in joints and soft tissues causing permanent disability and occasionally death mostly after brain hemorrhage. Treatment in Western countries (Berntorp, 2012) consists of intravenous injection of plasma-derived or recombinant FVIII protein concentrates at the time of a bleed to control it or prophylactically to prevent bleeding episodes. The short half-life for FVIII (12-18 hours) necessitates frequent infusions and makes this treatment prohibitively expensive for the majority of the world's hemophilia A patients. These individuals develop debilitating arthropathy and have a substantially increased risk of death from hemorrhage in life (Stonebraker, 2010). Chemical modification or bioengineering of FVIII may improve half-life to 18-19 hours (Kaufman, 2013). However, these extended half-life FVIII variants do not eliminate the need for lifelong FVIII protein administration (Hay, 2012).

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of human FVIII (hFVIII) following a single administration of vector. Hemophilia A is well-suited for this approach because its clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in low amounts (100-200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and a modest increase in the level of FVIII (a plasma level of 2 ng/ml protein leads to a 1% expression) can ameliorate the severe phenotype (Srivastava, 2013); thus, the therapeutic goal for gene therapy is a modest increase in hFVIII. Finally, the consequences of gene transfer can be assessed using validated quantitative endpoints that can be easily assayed in most clinical laboratories.

BMN 270 contains the cDNA for the B-domain-deleted SQ FVIII with a liver-specific HLP transcription promoter. The expression cassette is inserted between AAV2 ITRs, and this genome is packaged in the AAV5 capsid. A comprehensive review of BMN 270 is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.1 Nonclinical Studies

The nonclinical program supports a single IV infusion of BMN 270, the planned clinical route of administration, for the treatment of hemophilia A in male patients. This nonclinical program took into account the guidelines and reflection papers for gene therapy medicinal products under EMA Advanced Therapies as well as FDA guidance. The primary pharmacodynamics (PD), pharmacokinetics (PK), and toxicity of IV BMN 270 were characterized in a series of single dose studies in species that were vector permissive and responsive to the transgene including normal CD-1 mice, a B- and T-cell deficient mouse model of hemophilia A (B6;129S-*F8*^{tm1Kaz}/J x B6.129S6-*Rag2*^{tm1Fwa} N12; FVIII KO x Rag2), and normal cynomolgus and rhesus monkeys. Some PD studies evaluated additional PK, immunogenicity and toxicity endpoints.

The comparative pharmacodynamics of BMN 270 in cynomolgus monkeys with varying pre-existing AAV5 transduction inhibition (TI) titer and AAV5 TAb status was evaluated in study BMN270-16-021. BMN 270 was administered to 4 groups of monkeys, a control group (Group 1, n=3) that tested negative for both TI and AAV5 TAb, Group 2 (n=4) that was AAV5 TAb negative, and low TI titer (2-5) positive. Group 3 (n=4) was also AAV5 TAb negative, but had higher TI titers (5-10). Group 4 (n=5) tested positive for both AAV5 TAb and TI (TI titers were >5). Administration of BMN 270 by a single intravenous bolus injection was well-tolerated in cynomolgus monkeys regardless of baseline TI titer or TAb status. After dosing, all monkeys showed FVIII-SQ levels above the LLOQ, with the exception of two monkeys in the group that presented with both positive TI and TAb titers at baseline. Though these TAb+ monkeys, regardless of TI titers, showed a significant mean reduction in FVIII expression (68% less) compared to TAb negative monkeys, three of five monkeys showed detectable levels of FVIII-SQ, with one having levels similar to that observed in the TI and TAb negative control group. Monkeys that were TI+ but TAb-at baseline had FVIII expression levels that were similar to those of the TI and TAb negative control group.

Results of the nonclinical program to date are detailed in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.2 Previous Clinical Studies

Study 270-201 is an ongoing Phase 1/2, dose-escalation study to assess the safety, tolerability, and efficacy of BMN 270 in patients with severe hemophilia A (FVIII $\leq$1 IU/dL). Subjects received a single BMN 270 infusion and are to be followed for safety and efficacy for up to 5 years. A total of 15 subjects have been enrolled and dosed

with a single IV infusion of BMN 270 at one of 4 dose levels (6E12, 2E13, 4E13, and 6E13 vg/kg).

A comprehensive review of safety, efficacy, and immunogenicity results from 270-201 as of the latest data cut is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.3 Study Rationale

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. The clinical phenotype of HA patients generally correlates tightly with the level of residual expression. Severe HA is classified as FVIII activity less than 1% of wild-type (< 1 IU/dL), moderate disease comprises 1-5% of wild-type activity, and the mild form is 5-40% activity. The clinical manifestations of severe HA are frequent spontaneous bleeding episodes, predominantly in joints and soft tissues, with a substantially increased risk of death from hemorrhage when the brain is involved.

Treatment of severe HA presently consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study (Berntorp, 2017) and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with on-demand-only therapy in the same studies, respectively. Continued repeated bleeding events compound debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death.

Chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) and bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half-life to maximum of 18 hours, leaving critical periods when FVIII activity levels are below the therapeutic range and leaving patients vulnerable to bleeding and concomitant sequelae. As such, despite

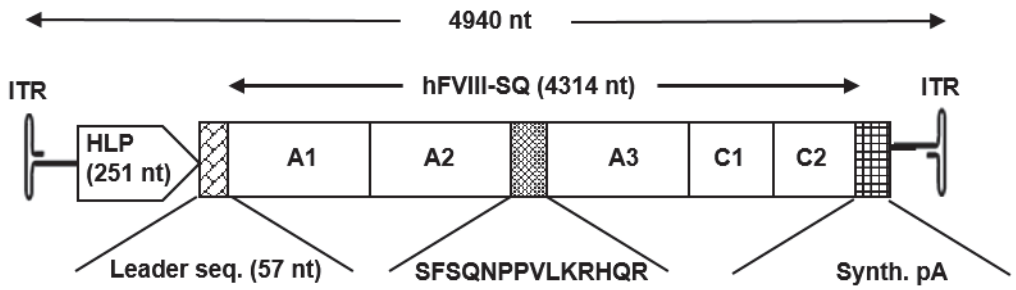
currently available FVIII replacement therapies, a high unmet need remains for maintenance of a sustained, consistent FVIII level compatible with a normal and hemorrhage-free life.

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of active FVIII following a single intravenous infusion of a vector encoding the appropriate gene sequence. Hemophilia A is well-suited for a gene replacement approach because clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in minute amounts (200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and even modest increases in the level of FVIII (any increase of the plasma level by 2 ng/ml induces an increase in activity of 1%) can ameliorate the severe form of hemophilia A. Thus, relatively small changes in endogenous FVIII activity can result in clinically relevant improvements in disease phenotype. Moreover, the circulating FVIII response to gene transduction can be assessed using validated quantitative endpoints that are easily assayed using established laboratory techniques.

Several different gene transfer strategies for FVIII replacement have been evaluated, with adeno-associated viral (AAV) vectors used in most hemophilia clinical trials. AAV vectors have an excellent and well-defined safety profile, and can direct long-term transgene expression with tropism and promoter specificity for specific tissues, such as the liver (eg serotypes 2, 5, and 8). Indeed, an ongoing gene therapy clinical trial for a related disorder, hemophilia B, has established over 7 years of stable human factor IX (hFIX) expression at levels sufficient for conversion of their bleeding phenotype from severe to moderate or mild, following a single peripheral vein infusion of AAV8-hFIX vector (Nathwani, 2014). Several participants in this trial have been able to discontinue factor prophylaxis without suffering spontaneous hemorrhages, even when they undertook activities that previously resulted in bleeding. Thus, gene therapy treatment has resulted in a substantial improvement in their quality of life.

BMN 270 is an AAV5-based gene therapy vector that expresses the SQ form of hFVIII under the control of a hybrid human liver-specific promoter (Figure 7.3.1).

Figure 7.3.1: hFVIII-SQ Vector Genome Schematic



Legend –Note that schematic is not to scale; nt = nucleotides

BMN 270 is delivered by a single IV dose and is designed to achieve stable, potentially life-long expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.

BMN 270 is being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. Preliminary results from 270-201 have demonstrated that following gene transfer, sustained FVIII activity above 15% (15 IU/dL) up to a year's observation is achievable with a dose of 4-6E13 vg/kg with an acceptable safety profile (Pasi, 2017). For additional information on preliminary data in 270-201, refer to the current version of the Investigator's Brochure.

Subjects enrolled and infused in 270-201 were screened and negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes.

Current data describing the extent of neutralization or inhibited transduction has been inconsistent across studies, each employing one of several varying methodologies to evaluate

both vector-specific antibodies and transduction inhibition (TI). Early studies using AAV2 for the treatment of hemophilia B described inhibited transduction in patients with neutralizing antibody titers of 2 or 3 (Manno, 2006), but more recent studies have described successful transduction in patients who, in retrospect, were determined to have pre-existing antibody to the vector capsid (Majowicz, 2017). Importantly, no drug-related adverse events were described in these patients. As the sensitivity of assays used to detect pre-existing antibody has increased greatly in recent years, the biological relevance of any given titer determination, and comparisons across studies using assays with differing titer sensitivities, requires additional study.

Data from a non-clinical study conducted by BioMarin in cynomolgus monkeys further demonstrated that transduction efficiency can vary greatly in the presence of pre-existing anti-AAV5 antibody. In non-human primates, the ability to attain FVIII levels in the face of measurable AAV5 antibody titers without significant safety risks supports the possibility of achieving a similar response in patients with AAV5 antibody positivity. The totality of available data shows the effect of pre-existing immunogenicity to AVV delivery vectors is not completely understood, and indicates that some patients may benefit from AAV5 gene therapy who are currently excluded.

In this study, patients with severe HA and pre-existing antibodies against AAV5 will be enrolled to evaluate the clinical response to BMN 270 gene therapy. The aim is to evaluate the safety, tolerability, and efficacy of BMN 270 in patients with severe HA and pre-existing antibodies against AAV5 vector capsid at various levels of AAV5 antibody titers.

7.4 Summary of Overall Risks and Benefits

The majority of subjects in the ongoing 270-201 clinical study who have received 4E13 or 6E13 vg/kg doses of BMN 270 have had Grade 1 asymptomatic elevations in ALT. For most subjects, the elevations have reached only slightly above the upper limit of normal (ULN). Based on the effectiveness of transient oral corticosteroid used to suppress a presumed Class 1 (cytotoxic T-cell) response in prior studies with hepatic transduction with AAV vectors (Mingozzi, 2013), subjects were treated with 7-32 weeks of oral corticosteroids prophylactically or in response to the elevations in ALT to ensure preservation of the transduced hepatocytes. Using this approach, no sustained loss of FVIII activity has been observed in subjects with ALT elevations, consistent with maintaining a high level of hepatocyte function. Moreover, the rise in ALT levels were not accompanied by significant or lasting aberrations in other liver tests such as AST, bilirubin or albumin, indicating that extent of toxicity is limited. There has been one HIV-positive subject in the ongoing 270-302 clinical study who experienced Grade 3 asymptomatic elevations in ALT and AST, which

has been attributed to an interaction between one or more of his antiretroviral therapy medications and/or unsuspected underlying hepatic disease with BMN 270. In addition, there has been one subject with Gilbert's syndrome in the ongoing 270-301 clinical study who has experienced Grade 3 asymptomatic elevations in ALT and AST. These cases have led to the exclusion of subsequent HIV-positive subjects and requirement of liver tests at Screening that are <1.25 times the upper limit of the normal range in the ongoing 270-301 and 270-302 clinical studies. Of note, two HIV-positive subjects in 270-301 and one presumed Gilbert's syndrome subject in 270-201 have received BMN 270 without experiencing any elevations in ALT to date. Overall, the literature and clinical experience with BMN 270 suggests that transient elevations in liver enzymes are expected following AAV-based gene therapy for the treatment for hemophilia A or B without any long-term concerns of hepatic injury (Manno, 2006; Nathwani, 2011; George, 2016; Miesbach, 2016; Pasi, 2017).

At the highest dose evaluated in 270-201 (6E13 vg/kg), the majority of subjects achieved FVIII levels above 50 IU/dL at 52 weeks post-infusion. Subjects in that cohort also reported markedly decreased bleeding episodes compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. Subjects at all dose levels continue to be followed. Subjects in 270-203, who have pre-existing immunity to the AAV5 vector capsid, may get no benefit from the study (in terms of increased FVIII activity) while possibly creating cross-reactive antibodies that may potentially preclude dosing with other serotypes.

As with any infused biological product, there is a potential risk of acute, systemic hypersensitivity reactions (including anaphylaxis) with BMN 270. No hypersensitivity reactions were observed during dosing of BMN 270 in the 270-201 clinical study, although one SAE of pyrexia was reported approximately 16 hours after the infusion in a subject in the 4E13 vg/kg cohort. The subject was treated with acetaminophen, and the fever resolved within 48 hours (see Investigator's Brochure for full details). Infusion-related reactions, including allergic reaction, maculopapular rash, and presyncope, have been reported from ongoing, actively dosing clinical studies of BMN 270, including this study. All of the infusion-related reactions were effectively managed clinically and resolved without any clinical sequelae. Refer to the Investigator's Brochure for additional details.

The current data available for BMN 270 does not yet permit comprehensive assessment of the benefit:risk profile of this investigational drug. Given the monitoring measures in place in the clinical protocol(s) to minimize the risk to subjects participating in the existing studies, the identified risks are justified by the anticipated benefits that may be afforded to subjects. Each subject in 270-203 will have a comprehensive surveillance plan that monitors LTs during the study, and elevations in LT will be addressed according to the guidelines set forth

in the protocol. Safety will be assessed by adverse event reporting and clinical laboratory assessments.

For additional information on findings in 270-201, refer to the current version of the IB.

8 STUDY OBJECTIVES

The primary objective of the study is to:

- Assess the safety of a single intravenous administration of BMN 270 in severe HA subjects with pre-existing antibody to AAV5 vector capsid, including development of FVIII neutralizing antibody

The secondary objectives of the study are to:

- Assess the efficacy of BMN 270 defined as FVIII activity at or above 5 IU/dL at Week 26
- Assess the impact of BMN 270 on usage of exogenous FVIII replacement therapy
- Assess the impact of BMN 270 on the number of bleeding episodes requiring exogenous FVIII therapy
- Evaluate the pharmacodynamics of FVIII expression following IV infusion of BMN 270
- Assess the impact of BMN 270 on patient-reported outcomes (PROs)

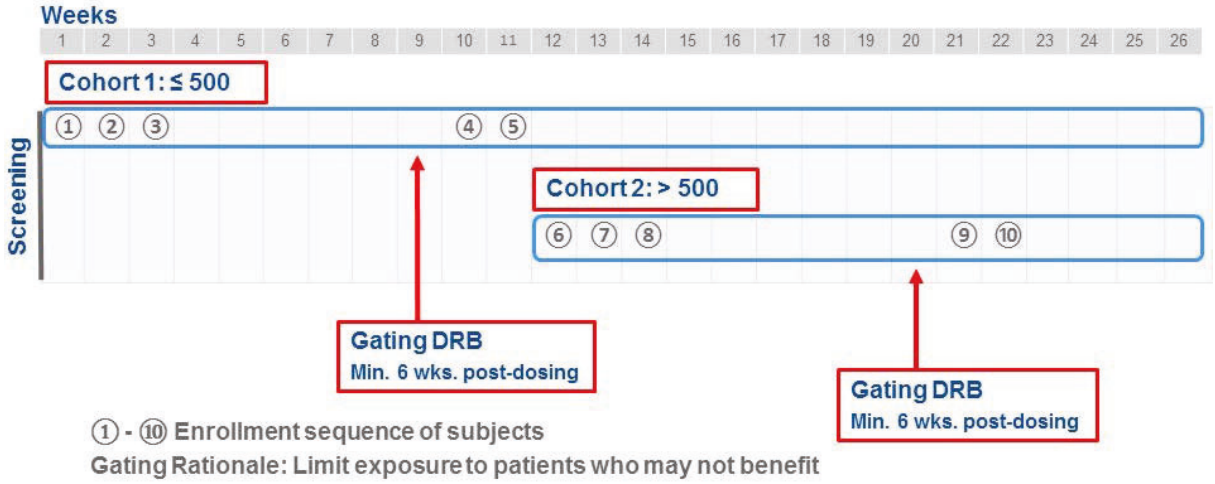
9 INVESTIGATIONAL PLAN

9.1 Overall Study Design and Plan

This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects may be enrolled at 2-3 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date.

The Data Review Board (DRB) will consist of the Principal Investigators and Sponsor's Medical Monitor, as well as hemophilia expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201. The DRB will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 9.1.1 represents one dosing schedule scenario):

Figure 9.1.1: 270-203 Dosing Schedule (One Scenario)



Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from a minimum of 6 weeks post-infusion) reviewed. Based on data from 270-201, FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time

point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response. Up to 6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness).

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DRB evaluation of safety and efficacy in treated subjects with the following triggers that may potentially pause further enrollment:

- any related SAE;
- any related AE with a severity > CTCAE Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects at 6 weeks post-BMN 270 infusion.

Following a temporary halt of enrolment, the DRB may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed. Additional blood samples will be collected (tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine bead array (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.

Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DRB based

on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL.

As the relationship between activity assay results of the BMN 270 gene product and bleeding remains to be established, Investigators should strive to minimize bias by avoiding consideration of FVIII activity levels by themselves or subjects in the reporting of bleeding episodes and FVIII usage.

In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis. Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after either Week 26 or Week 52, as applicable and at the discretion of the Investigator.

The study analysis will be performed after all subjects have been followed for 26 weeks post-BMN 270 infusion, with presentation of safety, efficacy, and FVIII PD assessments. After the safety, efficacy, and FVIII PD analyses at 26 weeks post-BMN 270 infusion, long-term safety and efficacy will be assessed in all subjects for up to a total of 5 years post-infusion. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the DRB.

A summary of all assessments is provided in the Schedule of Activities (SoA) in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5.

Table 9.1.1: Schedule of Activities

Assessment	Prior to BMN 270 Infusion			BMN 270 Infusion Visit (Day 1)[k]
	Screening (Day -28 to Day -1)	Smart Rescreening[i] (Day -28 to Day -1)	Baseline (Day -7 to Day -1)[h]	
Informed consent	X			
Demographics (age, sex, race, ethnicity)	X			
Medical History (including hemophilia A history, Hepatitis B, Hepatitis C, and HIV)	X			
Physical Examination[a]	X		X	X
Height and Weight[a]	X			
Vital Signs	X	X	X	X
Assessment of Adverse Events and Concomitant Medications	X	X	X	X
Documentation of bleeding episodes and FVIII usage for previous 12 months (by either subject or clinical information)	X	X	X	
Distribution of subject diaries and training in their use[l]	X			
Electrocardiogram	X			
Liver Ultrasound	X			
hFVIII Assays[b]	X	X[j]	X	
AAV5 TAb Assays[c]	X	X	X	X
AAV5 TI Assay[c]			X	X
Screen for Hepatitis B, Hepatitis C, HIV[d]	X			
Blood chemistry, hematology, and coagulation tests[e]	X	X	X	
Blood fasting lipid panel				X
Urine Tests[e]	X	X	X	
Liver Tests[e]	X	X	X	

Assessment	Prior to BMN 270 Infusion			BMN 270 Infusion Visit (Day 1)[k]
	Screening (Day -28 to Day -1)	Smart Rescreening[i] (Day -28 to Day -1)	Baseline (Day -7 to Day -1)[h]	
PBMC collection (for baseline determination of AAV5 and FVIII specific cellular immunity)			X	
Von Willebrand Factor Antigen (VWF:Ag)			X	
Direct Thrombin Activity Test			X	
Thrombin Generation Assay			X	
PCR of vector DNA in blood, saliva, urine, semen, and stools			X	X[k]
Biomarker testing[f]	X			
Exploratory biomarker assessments[g]			X	X
Cytokine bead array assay			X	
Hypersensitivity blood assessments[m]			X	(X)
Haemo-QOL-A assessment			X	
EQ-5D-5L assessment			X	
HAL assessment			X	
WPAI+CIQ:HS assessment			X	
BMN 270 Infusion				X

[a] Complete physical examination should be done at Screening. Brief physical examination may be done at Baseline and at the BMN 270 Infusion Visit.

[b] Includes baseline FVIII activity (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), coagulation exploratory assay, hFVIII inhibitor level (Bethesda assay with Nijmegen modification), hFVIII total antibody titer, and hFVIII antigen assay. Baseline activity should be assessed at trough (at least >72 hours after last dose of replacement FVIII therapy, or 5x the known half-life of the FVIII concentrates administered).

[c] Sample collection on the day of the infusion visit must be performed before the BMN 270 infusion is given. Screening, Smart Re-screening, and Infusion Day samples will be tested in an AAV5 Tab pre-screening assay specifically developed for enrollment purposes. Baseline and all post-dose samples will be tested in a different AAV5 TAb post-dose immunogenicity monitoring assay

[d] Subjects with documented negative results within the last 30 days do not need to be retested. Hepatitis B screening should include hepatitis B surface antigen (HBsAg), hepatitis B surface antibody (HBsAb), and hepatitis B core antibody (HBcAb).

[e] Detailed in the Section of laboratory assessments and liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. ABO blood typing assessment should be performed as part of the hematology assessment (at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study).

[f] Includes HLA genotyping and FVIII genotyping.

[g] Blood samples will be collected from subjects to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for those evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay, Direct Thrombin Activity test, and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[h] Should the screening visit occur within 30 days of the drug infusion, physical examination, blood chemistry, LTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline.

[i] Smart rescreening should only be performed if a subject has been determined to be eligible for the study and is unable to complete the Baseline assessments and Infusion prior to the closing of the original Screening window (28 + 14 days). Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once.

[j] Only the hFVIII inhibitor level (Bethesda assay with Nijmegen modification) assay must be done at smart rescreening.

[k] With the exception of the collection of samples for PCR vector DNA analysis, assessments on the day of infusion must be performed prior to the infusion. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit. On the day of the BMN 270 Infusion, vital signs will be monitored prior to the infusion, during the infusion every 15 minutes (± 5 minutes), and following the infusion hourly (± 5 minutes) for a total time of 24 hours during the subject's stay in the clinic. Shedding samples for PCR of vector DNA analysis (blood, saliva, urine, semen, stool) should be collected between 2 and 24 hours after the infusion has been completed.

[l] Diaries should be distributed to subjects who have consented to participate in the study and who have been determined to meet all study eligibility criteria.

[m] In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination, vital signs will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.

Table 9.1.2: Schedule of Activities – Post-Infusion Follow-Up (Week 1-16)

Assessment	Follow-Up After BMN 270 Infusion – Weeks*																	
	Week 1																	
	D2	D4	D8	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Study Day*	2	4	8	15	22	29	36	43	50	57	64	71	78	85	92	99	106	113
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight						X				X				X				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X		X		X		X						X		
Urine Tests[b]														X				
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII antibody titer				X		X		X		X		X		X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]	X[d]	X	X	X	X	X		X		X				X				X
Exploratory biomarker assessments[e]				X				X				X				X		
Haemo-QOL-A assessment														X				
EQ-5D-5L assessment														X				
HAL assessment														X				
WPAI+CIQ:HS assessment														X				
AAV5 TAb Assay	X		X	X		X		X		X		X		X		X		X
AAV5 TI Assay	X		X	X		X		X		X		X		X		X		X

Assessment	Follow-Up After BMN 270 Infusion – Weeks*																	
	Week 1																	
	D2	D4	D8	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Study Day*	2	4	8	15	22	29	36	43	50	57	64	71	78	85	92	99	106	113
Testing for reactivation of hepatitis B and hepatitis C																		X[f]
PBMC collection (for determination of AAV5 and FVIII specific immunity)	X			X		X		X		X		X		X		X		X
VWF:Ag						X				X				X				X
Direct Thrombin Activity test															X			
Cytokine bead array assay	X		X	X				X				X				X		

* Visit windows are ± 24 hours during Week 1 and ± 48 hours starting with the Week 2 visit.

[a] Brief physical examination should be done at all weekly visits.

[b] Detailed in the Section of laboratory assessments and liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ≥1.5x ULN or > ULN & > 2x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen assay. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Collection for each matrix to occur until at least 3 consecutive negative results are obtained. Collection and testing of semen samples must continue through Week 12, even if 3 consecutive negative results in that compartment have already been recorded. Separate collection on Day 2 is not required if vector shedding samples were collected on Day 1 post-BMN 270 infusion; if collection on Day 2 is needed, it must occur within 24 hours of the time of the BMN 270 infusion.

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay, Direct Thrombin Activity test, and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] Testing for reactivation of hepatitis B and hepatitis C at Week 16, for subjects with a past medical history of hepatitis B or hepatitis C prior to study entry, should be performed only in subjects who have not received therapeutic oral corticosteroids prior to Week 16; subjects who have received therapeutic oral corticosteroids will have hepatitis B and hepatitis C testing at the time points indicated in Table 9.1.6.

Table 9.1.3: Schedule of Activities – Post-Infusion Follow-Up (Week 17-32)

Assessment	Follow-Up After BMN 270 Infusion – Weeks*															
	17	**18**	**19**	**20**	**21**	**22**	**23**	**24**	**25**	**26**	**27[f]**	**28[f]**	**29[f]**	**30[f]**	**31[f]**	**32[f]**
Study Day*	120	127	134	141	148	155	162	169	176	183	190	197	204	211	218	225
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight[a]				X						X						X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X						X						X
Urine Tests[b]										X						
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII antibody titer				X				X		X						X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]				X				X		X						X
Exploratory biomarker assessments[e]		X				X				X						X
Haemo-QOL-A assessment										X						
EQ-5D-5L										X						
HAL										X						
WPAI+CIQ:HS										X						
AAV5 TAb Assay		X		X		X		X		X		X		X		X
AAV5 TI Assay		X		X		X		X		X		X		X		X

Assessment	Follow-Up After BMN 270 Infusion – Weeks*															
	17	**18**	**19**	**20**	**21**	**22**	**23**	**24**	**25**	**26**	**27[f]**	**28[f]**	**29[f]**	**30[f]**	**31[f]**	**32[f]**
Study Day[*]	120	127	134	141	148	155	162	169	176	183	190	197	204	211	218	225
PBMC collection (for determination of AAV5 and FVIII specific cellular immunity)		X		X		X		X		X		X		X		X
VWF:Ag				X						X						
Direct Thrombin Activity Test[e]										X						
Thrombin Generation Assay[e]				X				X		X						X
Cytokine bead array assay				X				X		X						X

[*] Visit windows are ± 48 hours.

[a] Brief physical examination should be done at all weekly visits except Week 26, where a complete physical examination should be performed. Weight should be recorded at Week 20 and Week 26.

[b] Detailed in the Section of laboratory assessments and liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs may be monitored more or less frequently (and in particular when ALT values are ≥ 1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ≥ 1.5x ULN or > ULN & > 2x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Collection for each matrix to occur until at least 3 consecutive negative results are obtained.

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay, Direct Thrombin Activity test, and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] Subjects who meet the definition of treatment failure to BMN 270 therapy after Week 26 (refer to Section 12.5.3) may omit Week 27-32 visits. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in all fluids must still provide samples at Week 32 (but do not need to do other scheduled assessments on that date).

Table 9.1.4: Schedule of Activities – Post-Infusion Follow-Up (Week 33-52)

Assessment	Year 1 – Weeks*											
	33[e]	34[e]	35[e]	36	38[e]	40[e]	42[e]	44	46[e]	48[e]	50[e]	52
Study Day[*]	232	239	246	253	267	281	295	309	323	337	351	365
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X
Weight[a]				X				X				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X				X				X
Urine Tests[b]				X								X
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]	X	X	X	X	X	X	X	X	X	X	X	X
AAV5 TAb Assay		X		X		X		X		X		X
AAV5 TI Assay		X		X		X		X		X		X
FVIII antibody titer				X				X				X
Exploratory biomarker assessments[d]				X		X		X		X		X
PBMC Collection (for determination of FVIII and Capsid specific cellular immunity)		X		X				X				X
VWF:Ag				X								X
Direct Thrombin Activity Test[d]				X								X
Thrombin Generation Assay[d]				X		X		X		X		X
Cytokine bead array assay				X				X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools				X		X		X		X		X
Haemo-QOL-A assessment												X
EQ-5D-5L												X

Assessment	Year 1 – Weeks*											
	33[e]	34[e]	35[e]	36	38[e]	40[e]	42[e]	44	46[e]	48[e]	50[e]	52
HAL												X
WPAI+CIQ:HS												X

* Visit windows are ± 48 hours through Week 36, then ±1 week until Week 52.

[a] Complete physical examination should be performed at Week 52; brief physical exam may be performed at other study visits. Weight should be recorded at Week 36 and every 4 weeks through Week 52.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ≥ 1.5x ULN or > ULN & > 2x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN or increased by > 10 U/L from prior assessment; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII protein assay. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay, Direct Thrombin Activity test, and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[e] Subjects who meet the definition of treatment failure to BMN 270 therapy after Week 26 (refer to Section 12.5.3) must attend the Week 36, Week 44, and Week 52 visits but may omit the visits at Weeks 33, 34, 35, 38, 40, 42, 46, 48, and 50. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in all fluids must still provide samples at Week 40 and Week 48, if necessary (but do not need to do other scheduled assessments on that date).

Table 9.1.5: Schedule of Events – Long-Term Follow-Up (Year 2 – Year 5)

Assessment	Years 2-5*	Year 2*	Years 3-5*	End of Year Visit				ETV
				Year 2	Year 3	Year 4	Year 5	
Study Week*	Q12W	Q4W[g]	Q6W[g]	W104	W156	W208	W260	
Physical examination[a]	X[a]			X[a]				X
Weight[a]	X[a]			X[a]				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X				X
Vital Signs	X			X				X
Blood chemistry, hematology, and coagulation tests[b]	X[b]			X[b]				X
Urine Tests[b]	X[b]			X[b]				X
Liver Tests[b]	X	X	X	X				X
FVIII assays[c]	X	X	X	X				X
AAV5 TAb Assay	X			X				X
AAV5 TI Assay	X			X				X
FVIII antibody titer	X			X				X
Exploratory biomarker assessments[e]	X			X				X
PBMC Collection (for determination of FVIII and Capsid specific cellular immunity)	X			X				X
VWF:Ag	X			X				X
Direct Thrombin Activity Test[e]	X			X				X
Thrombin Generation Assay[e]	X			X				X
Cytokine bead array assay	X			X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]	(X)[d]	(X)[d]	(X)[d]	(X)[d]				(X)[d]
Haemo-QOL-A assessment				X[f]				X
EQ-5D-5L				X[f]				X

Assessment	Years 2-5*	Year 2*	Years 3-5*	End of Year Visit				ETV
				Year 2	Year 3	Year 4	Year 5	
Study Week*	Q12W	Q4W[g]	Q6W[g]	W104	W156	W208	W260	
HAL				X[f]				X
WPAI+CIQ:HS				X[f]				X

* Visit windows are ± 2 weeks for visits in Years 2-5. The Q6W visits during Years 3-5 should restart after each End of Year visit (eg, the first Q6W visit during Year 3 should be ~6 weeks after the End of Year 2 visit).

[a] Brief physical examination should be performed at all visits during Years 2-5. Weight should be recorded at the second Q12W visit each year and at every End of Year visit during Years 2-5.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ≥ 1.5x ULN or > ULN & > 2x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN or increased by > 10 U/L from prior assessment; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. During Years 2-5 of the Post-Infusion Follow-Up period, urine tests and blood, chemistry, and coagulation tests should be performed at the second Q12W visit each year and at every End of Year visit.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII protein assay. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism. If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 3-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.

[d] Sample testing during Long-Term Follow-Up is not required if at least 3 consecutive samples were negative during the Post-Infusion Follow-Up period. Subjects who have not had 3 consecutive negative semen samples by Week 52 should continue to have PCR testing of semen every 4 weeks (during Year 2) or every 6 weeks (during Years 3-5) until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes

that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay, Direct Thrombin Activity test, and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] PRO assessments during Years 2-5 of Long-Term Follow-up should be performed at every End of Year visit.

[g] Subjects who meet the definition of treatment failure to BMN 270 therapy after Week 52 (refer to Section 12.6) may omit the Q4W and Q6W visits during Years 2-5, and must attend only the Q12W and End of Year visits. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in all fluids must still provide samples Q4W (during Year 2) or Q6W (during Years 3-5) until vector shedding has been cleared (but do not need to do other scheduled assessments at those visit dates).

Table 9.1.6: Therapeutic Corticosteroids for LT Elevations

	Steroid Treatment Period[b]								Post-Steroid Period[c]				
	Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Week 8[b]	Week 1	Week 2	Week 3	Week 4	Week 13
Therapeutic corticosteroids (dose in mg/day)[a]	60 mg	60 mg	40 mg	40 mg	40 mg	30 mg	20 mg	10 mg					
FVIII activity testing									X	X	X	X	
Liver testing									X	X	X	X	
Hepatitis B testing[d]						X			X				X
HCV Viral Load[d]						X			X				X

[a] Therapeutic oral corticosteroids may be initiated according to the parameters set out in Section 9.4.8.2.

[b] Following initiation or completion of steroid regimen, if a recurrence of ALT values ≥ 1.5x ULN is reported, steroid management decisions will based on discussions between the Investigator and Medical Monitor. Modification of the steroid regimen may take into consideration possible confounders for the ALT elevation, relationship between increases in ALT and FVIII activity, ALT/FVIII levels post steroid initiation, and adverse events related to steroid dosing. Guidance for tapering oral corticosteroid dosing can be found in Section 9.4.8.2.

[c] After discontinuation of oral corticosteroids, weekly labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values. If these assessments are already being done as part of normal study follow-up, they do not need to be duplicated.

[d] Should only be performed in subjects with a history of hepatitis B or hepatitis C prior to study entry.

9.2 Discussion of Study Design, Including Choice of Control Group

Study 270-203 is designed to be a Phase 1/2, single-arm, open-label study in hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid. Hemophilia A patients who provide written informed consent and meet the entry criteria will be eligible to enroll in the study.

Approximately 10 subjects may be enrolled at the 6E13 vg/kg BMN 270 dose. Subjects will be followed for 26 weeks post-BMN 270 infusion during which safety and efficacy assessments will be taken. After the final analysis at 26 weeks post-infusion, safety and efficacy will then continue to be assessed long-term for approximately a total of 5 years.

9.3 Selection of Study Population

Approximately 10 hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid may enroll into the study.

Additional criteria for participation in the study are provided in Section 9.3.1 and Section 9.3.2.

9.3.1 Inclusion Criteria

Individuals eligible to participate in this study must meet all of the following criteria:

1. Males ≥ 18 years of age with hemophilia A and residual FVIII levels ≤ 1 IU/dL as evidenced by medical history, at the time of signing the informed consent
2. Detectable pre-existing antibodies against the AAV5 vector capsid as measured by AAV5 total antibody ELISA
3. Treated/exposed to FVIII concentrates or cryoprecipitate for a minimum of 150 exposure days (EDs)
4. Subject must have been on prophylactic FVIII replacement therapy for at least 12 months prior to study entry.
5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures.
6. No previous documented history of a detectable FVIII inhibitor, and results from a Bethesda assay or Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) (or less than 1.0 BU for laboratories with a historical lower sensitivity cutoff for inhibitor detection of 1.0 BU) on 2 consecutive occasions at least one week apart within the past 12 months (at least one of which should be tested at the central laboratory)

7. Sexually active participants must agree to use an acceptable method of effective contraception, either double barrier contraception (ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with no detectable viral vector DNA.
8. Willing to abstain from consumption of alcohol for at least the first 26 weeks following BMN 270 infusion.

9.3.2 Exclusion Criteria

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Any evidence of active infection or any immunosuppressive disorder, including HIV infection.
2. Significant liver dysfunction with any of the following abnormal laboratory results:
    - ALT (alanine aminotransferase) > 1.25x ULN;
    - AST (aspartate aminotransferase) > 1.25x ULN;
    - GGT (gamma-glutamyltransferase) > 1.25x ULN;
    - Total bilirubin > 1.25x ULN;
    - Alkaline phosphatase > 1.25x ULN; or
    - INR (international normalized ratio) ≥ 1.4

Subjects whose laboratory assessments fall outside of these ranges may undergo repeat testing of the entire liver test panel within the same Screening window and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor.

1. Prior liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used
2. Evidence of any bleeding disorder not related to hemophilia A
3. Platelet count of < 100 x 10^9/L
4. Creatinine ≥ 1.5 mg/dL
5. Liver cirrhosis of any etiology as assessed by liver ultrasound

6. Chronic or active hepatitis B as evidenced by positive serology testing (hepatitis B surface antigen [HBsAg], hepatitis B surface antibody [HBsAb], and hepatitis B core antibody [HBcAb]) and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results in the Laboratory Manual.
7. Active Hepatitis C as evidenced by detectable HCV RNA, or currently on antiviral therapy
8. Active malignancy, except non-melanoma skin cancer
9. History of hepatic malignancy
10. History of arterial or venous thromboembolic events (eg, deep vein thrombosis, non-hemorrhagic stroke, pulmonary embolism, myocardial infarction, arterial embolus), with the exception of catheter-associated thrombosis for which anti-thrombotic treatment is not currently ongoing.
11. Known inherited or acquired thrombophilia, including conditions associated with increased thromboembolic risk, such as atrial fibrillation.
12. A history of known inflammatory, connective tissue, or autoimmune disorders (eg, vasculitis).
13. Treatment with any Investigational Product within 30 days or 5 half-lives of the investigational product (whichever is longer) prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study
14. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including possible corticosteroid treatment outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.
15. Prior treatment with any vector or gene transfer agent
16. Major surgery planned in the 26-week period following the infusion with BMN 270
17. Use of systemic immunosuppressive agents, not including corticosteroids, or live vaccines within 30 days before the BMN 270 infusion
18. Concurrent enrollment in another clinical study, unless it is an observational (non-interventional) clinical study that does not interfere with the requirements of the current protocol or have the potential to impact the evaluation of safety and efficacy of BMN 270 and with prior consultation with the Medical Monitor
19. Known allergy or hypersensitivity to investigational product formulation
20. Unwilling to receive blood or blood products for treatment of an adverse event and/or a bleed

9.3.3 Removal of Subjects from Treatment or Assessment

Subjects may withdraw their consent to participate in the study at any time without prejudice. The Investigator must withdraw from the study any subject who requests to be withdrawn. Such subjects will always be asked about the reason(s) for withdrawal. The Investigator will discuss with the subject appropriate procedures for withdrawal from the study. The Investigator should ask the subject's consent to perform the procedures listed under the early termination visit. Should a subject withdraw from the study, all efforts will be made to complete and report the observations as thoroughly as possible up to the date of the withdrawal.

A subject's participation in the study may be discontinued at any time at the discretion of BioMarin or of the Investigator and in accordance with his/her clinical judgment. When possible, the tests and evaluations listed for the termination visit should be carried out and every effort will be made to gather follow-up safety data if possible.

BioMarin must be notified of all subject withdrawals as soon as possible. BioMarin also reserves the right to discontinue the study at any time for either clinical or administrative reasons and to discontinue participation by an individual Investigator or site for poor enrollment or noncompliance.

Reasons for which the Investigator or BioMarin may withdraw a subject from the study include, but are not limited to, the following:

- Subject requires medication or medical procedure prohibited by the protocol
- Subject was erroneously enrolled into the study or does not meet entry criteria and not yet been dosed with BMN 270; subjects who do not meet entry criteria but who erroneously receive BMN 270 should remain in the study for safety monitoring
- Subject is lost to follow-up

If a subject fails to return for scheduled visits, a documented effort must be made to determine the reason. If the subject cannot be reached by telephone, a certified letter should be sent to the subject requesting contact with the Investigator. This information should be recorded in the study records.

The Investigator or designee must explain to each subject, before enrollment into the study, that for evaluation of study results, the subject's protected health information obtained during the study may be shared with the study Sponsor, regulatory agencies, and IRB/EC It is the Investigator's (or designee's) responsibility to obtain written permission to use protected health information per country-specific regulations, such as the Health Insurance Portability

and Accountability Act (HIPAA) in the US, from each subject. If permission to use protected health information is withdrawn, it is the Investigator's responsibility to obtain a written request, to ensure that no further data will be collected from the subject and the subject will be removed from the study.

9.3.3.1 Study Safety Evaluation Criteria

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DRB evaluation of safety and efficacy in treated subjects with the following triggers for pausing further enrollment:

- any related SAE;
- an related AE with a severity > Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

Additionally, the DRB should consider whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction. Following a temporary halt of enrolment, the DRB may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

If a Grade 4 or Grade 5 adverse event assessed as related to study drug occurs in a study subject, further enrollment into the trial will be halted until a full safety review of the data by the DRB has taken place. Relevant reporting and discussion with the Sponsor and the DRB will take place before resumption of dosing.

If any of the following events occur in a subject in the study who has received BMN 270 infusion, further enrollment into the trial will be temporarily put on hold pending prompt evaluation by the DRB.

1. Liver dysfunction (criteria do not apply to ALT elevations with an extra-hepatic etiology):
    - ALT >5x ULN, for more than 2 weeks
    - ALT >3x ULN and (total bilirubin >2x ULN **or** INR >1.5)
    - ALT >3x ULN with signs and symptoms of liver dysfunction
2. The occurrence of an AE of hepatic failure.
3. The detection of high titer neutralizing antibodies (>5 BU) to hFVIII following BMN 270 infusion in two subjects.
4. The occurrence of any cancer (except non-melanoma skin cancer) at any point after BMN 270 infusion.

5. The occurrence of a thromboembolic event with FVIII activity > 150 IU/dL in one subject.

If any of the following events occurs in a subject in the study who has received BMN 270 infusion, a prompt evaluation by the DRB will be required. Further enrollment into the trial will continue while DRB evaluation is ongoing, unless deemed otherwise by the DRB.

1. Acute hypersensitivity assessed as related to BMN 270
3. The detection of high titer neutralizing antibodies (>5 BU) to hFVIII following BMN 270 infusion in one subject
4. Occurrence of a thromboembolic event

9.3.4 Subject Identification and Replacement of Subjects

Each subject will be assigned a unique subject identifier. This unique identifier will be on all eCRF pages. In legal jurisdictions where use of initials is not permitted, a substitute identifier will be used.

Subjects who withdraw from the study after receiving BMN 270 will not be replaced.

9.3.5 Duration of Subject Participation

The duration of participation for each subject will be approximately 264 weeks. This includes 4 weeks of screening, 1 day of BMN 270 infusion, 26 weeks of Post-Infusion Follow-Up, and 234 weeks of Long-Term Follow-Up.

9.4 Treatments

9.4.1 Treatments Administered

BioMarin and/or its designee will provide the study site with a supply of IP sufficient for the completion of the study. BioMarin is responsible for shipping study drug to clinical sites.

9.4.2 Identity of Investigational Product

9.4.2.1 Product Characteristics and Labeling

BMN 270 is a sterile, clear, colorless-to-pale yellow solution for IV infusion and is supplied in a 2 mL polypropylene cryovial. Each vial contains 1.1 mL of AAV5-hFVIII-SQ at a concentration of 2E13 vector genomes per mL in a pH 7.4 phosphate buffer.

The study drug is labelled according to the particulars approved by the relevant regulatory agencies.

9.4.3 Storage

At the study site, all IP must be stored under the conditions specified in the Pharmacy Manual in a secure area accessible only to the designated pharmacists and clinical site personnel. All IP must be stored and inventoried and the inventories must be carefully and accurately documented according to applicable state, federal and local regulations, ICH GCP, and study procedures.

9.4.4 Directions for Administration

On the day of infusion, the subject will come to the infusion site, where a physical examination will be performed by the Investigator or designee. If the subject is found to have an active acute illness at the time of planned infusion, then the infusion should be deferred until the illness has resolved; screening procedures may require repetition if outside the specified window. An IV catheter or butterfly needle will be inserted into a suitable peripheral vein (eg, the median cubital vein) and flushed with saline. FVIII replacement therapy will not be given since venipuncture is a minimally invasive procedure in these individuals under ordinary conditions.

BMN 270 will be prepared and infused as a pure solution over a dose-dependent time. Prepared drug will be kept at room temperature prior to administration. An electric syringe pump will be used to infuse through an in-line, low protein binding 0.22 micron filter. BMN 270 will be infused through the catheter using an appropriate infusion pump at an initial rate of 1 mL/min. The infusion rate should be increased every 30 minutes by 1 mL/min up to a maximum of 4 mL/min, provided that the subject's clinical condition permits such an increase. Of note, the IP has been shown to be stable at room temperature for 7.5 hours following completion of product thaw. Vital signs (pulse, blood pressure, respiration rate and temperature) should be monitored at 15 minute (±5 minutes) intervals throughout the period of the infusion.

As with any infused biological product, there is a potential risk of acute, systemic hypersensitivity reactions (including anaphylaxis) with BMN 270. Dosing will be administered at a qualified infusion site, with appropriate resuscitation equipment and medication available and easily accessible.

Clinical staff administering BMN 270 should be trained appropriately in recognizing and managing the signs and symptoms associated with potential hypersensitivity, anaphylactic, and anaphylactoid reactions. Additionally, the Investigator should be familiar with Sampson's criteria for defining anaphylaxis (Sampson, 2006; Appendix 1).

Should symptoms of potential hypersensitivity occur, the infusion may be slowed or halted at the Investigator's discretion, with consideration of the subject's clinical condition. If the infusion is halted, it should only be restarted if the Investigator considers it safe and appropriate to do so. Antihistamines, anti-pyretic, and/or corticosteroid administration is permitted prior to restarting an interrupted infusion by an infusion-related reaction. At the restart, the infusion rate may be adjusted (ie, to a slower rate [minimum of 1 mL/min], with the rate increased every 30 minutes by 1 mL/min up to a maximum rate of 4 mL/min, if the subject's clinical condition permits such an increase) with careful monitoring of the subject.

In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.

Following completion of the infusion, vital signs will be monitored hourly (± 5 minutes). If the vital signs are stable the catheter will be removed 8 hours after the infusion. Hemostasis at the puncture site will be established by applying pressure according to standard protocol for infusing FVIII concentrates. Subjects will remain in the clinic for at least 24 hours to observe for any immediate toxicity of the procedure; in-patient observation can be extended beyond 24 hours if needed per Investigator discretion. After 24 hours, subjects will be discharged from the clinic unless toxicity has been observed in which case the stay in the clinic may be extended or the subject may transfer to a separate facility based on the evaluation and judgment of the Principal Investigator after consultation with the Medical Monitor.

Prior to discharging subjects from the clinic, the Investigator or designee should instruct subjects how to recognize signs and symptoms of potential (delayed) hypersensitivity reactions and anaphylaxis, and to contact a medical practitioner or seek emergency care in case of such an event.

9.4.5 Method of Assigning Subjects to Treatment Groups

Subjects who meet all eligibility criteria (refer to Section 9.3.1 and Section 9.3.2) may be enrolled into the study. Approval by the Medical Monitor will be required prior to enrollment of each study subject. Upon their enrollment into the study, subjects will be assigned a unique subject number.

9.4.6 Selection of Dose Used in the Study

Data from an ongoing first in human study (270-201) indicates that following single escalated doses of BMN 270 (6E12, 2E13, 4E13, 6E13 vg/kg), dose-related increases in FVIII activity were observed, with concurrent improvements in bleeding episodes and exogenous FVIII utilization, particularly at the 4E13 and 6E13 vg/kg dose levels. At all dose levels, BMN 270 is considered to be well-tolerated with mild increases in ALT as the most common adverse event. Please refer to the IB for detailed efficacy and safety data. The 6E13 vg/kg dose has been selected for this study to maximize the likelihood of transduction in the face of pre-existing AAV5 antibodies.

9.4.6.1 Selection of Timing of Dose for Each Subject

9.4.7 Blinding

This is an open-label study.

9.4.8 Prior and Concomitant Medications

All prescription and over-the-counter medications (including dietary and herbal supplements) taken by a subject for 30 days before Screening will be recorded on the designated eCRF. The Investigator may prescribe additional medications, deemed necessary to provide adequate prophylactic or supportive care, during the study, as long as the prescribed medication is not prohibited by the protocol. In the event of an emergency, any needed medications may be prescribed without prior approval, but the Medical Monitor must be notified of the use of any contraindicated medications immediately thereafter. Any concomitant medications added or discontinued during the study should be recorded on the eCRF. Medications should, whenever possible, not be recorded in the electronic database with a frequency of PRN.

The following medications are prohibited starting 30 days before Screening and through the end of the study, and the Sponsor must be notified if a subject receives any of these during the study:

- Any investigational therapy

- Systemic immunosuppressive agents, except for corticosteroids
- Emicizumab
- Fitusiran
- Concizumab
- Efavirenz
- Lamivudine

Subjects who begin antiretroviral therapy following BMN 270 infusion and during their participation in 270-203 must have their antiretroviral medication regimen approved by the Investigator and Medical Monitor prior to starting antiretroviral treatment.

The following medications should be avoided, starting 30 days prior to and for at least 26 weeks after BMN 270 infusion and minimized throughout the remaining duration of the study.

- Alcohol
- Herbal and natural remedies and dietary supplements
- Medications which may be hepatotoxic

Vaccines should also be avoided during this period, but in particular during the first 26 weeks unless clinically indicated.

The following medications should be avoided during oral corticosteroid therapy:

- Vaccines
- NSAIDs

9.4.8.1 Concomitant Hemophilia Treatments

Subjects on prophylactic FVIII therapy will discontinue their regular treatment regimen starting 4 weeks after the day of infusion and switch to an “on-demand” schedule. FVIII replacement therapy can always be taken as needed by the subject for treatment of an acute bleeding episode; the subject must carefully record his treatment and bleeding episodes in his diary. Prophylactic FVIII can be used on a case-by-case basis and in consultation with the Medical Monitor to prevent bleeding in extenuating circumstances (eg, peri-operative).

In addition, information on FVIII usage and bleeding episodes by medical history will be collected from subjects for the 12-month period immediately preceding study enrollment.

9.4.8.2 Therapeutic Glucocorticoid Treatment for Elevated Hepatic Transaminases

Therapeutic oral corticosteroids (prednisone or converted equivalent) should be initiated when either of the following occurs post-BMN 270 infusion in any subject and after consultation with the Medical Monitor:

- ALT ≥ 1.5x ULN or ALT > ULN & > 2x baseline value in 2 consecutive assessments within 72 hours and alternative etiologies have been ruled out, or ALT ≥ 3x ULN in 2 consecutive assessments within 48 hours (refer to Table 9.7.6.3.2)
    - Whenever possible, a confirmatory lab draw for ALT should be performed, along with FVIII activity, prior to initiating oral corticosteroids.
    - Corticosteroids may be delayed if elevations in ALT are clearly not related to BMN 270 (eg, elevated in ALT with concurrent increase in CPK due to intensive exercise)

The prescribed regimen for therapeutic oral corticosteroids is detailed in Table 9.1.6. Changes to the corticosteroid regimen should be made as follows:

Table 9.4.8.2.1: Adjustments to Corticosteroid Regimen

Tapering Corticosteroid Dose	Subject has been receiving oral corticosteroids <3 weeks	Corticosteroids may be discontinued **if**: • ALT <1.5x ULN or ALT < ULN & < 2x baseline value; and • FVIII levels > 20 IU/dL and within 10% of the pre-decline FVIII levels; and • There is no concern for adrenal insufficiency post-withdrawal
	Subject has been receiving oral corticosteroids ≥3 weeks	Corticosteroids may be tapered by 10 mg weekly **if**: • ALT <1.5x ULN or ALT < ULN & < 2x baseline value; and • FVIII levels > 20 IU/dL and within 10% of the pre-decline FVIII levels; and • There is no concern for adrenal insufficiency post-withdrawal
Increasing Corticosteroid Dose	If ALT level is increasing or FVIII level is decreasing while on oral corticosteroids, any increases in oral corticosteroid dosing should be made only upon consultation with the Medical Monitor	

For any scenarios that are not accounted for in the above table, a discussion should take place between the Investigator and Medical Monitor regarding corticosteroid dose adjustments.

After discontinuation of oral corticosteroids, labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values.

Following initiation or completion of therapeutic oral corticosteroids, if ALT elevation ≥1.5x ULN or ALT ≥ ULN & ≥ 2x baseline value is reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor.

Modification of the corticosteroid regimen may take into consideration possible confounders for the ALT elevation and impact on FVIII expression.

Management and monitoring of reactions to corticosteroids should be determined by the Investigator's clinical judgment in consultation with the Sponsor's Medical Monitor. This includes the contraindicated use of NSAIDs during corticosteroid treatment and specific monitoring not already covered by the SoA. The use of COX-2 inhibitors, while not contraindicated during corticosteroid treatment, should be limited, if possible. Practical management to prevent complications related to oral corticosteroid therapy may be undertaken at the discretion of the Investigator (eg, evaluation of glucose intolerance, hyperlipidemia etc.). Hepatitis B status and HCV viral load will be rechecked 6 weeks after the start of oral corticosteroid treatment and then 1 week and 13 weeks after the completion of oral corticosteroid treatment in subjects with a history of hepatitis B or hepatitis C. All adverse events (including any adverse events suspected to be caused by or related to corticosteroid use) should be reported as outlined in Section 10 of the protocol.

9.4.9 Treatment Compliance

Study drug will be administered to subjects at the dosing facility by a qualified health care professional. The quantity dispensed, returned, used, lost, etc. must be recorded on a dispensing log. Sites will be instructed to return or destroy all used and unused study drug containers.

9.5 Investigational Product Accountability

The Investigator or designee is responsible for maintaining accurate records (including dates and quantities) of IP(s) received and IP lost or accidentally or deliberately destroyed. The Investigator or designee must retain all unused or expired study supplies until the study monitor (on-site CRA) has confirmed the accountability data, if allowed by local SOPs.

9.5.1 Return and Disposition of Clinical Supplies

Unused study drug must be kept in a secure location for accountability and reconciliation by the study monitor. The Investigator or designee must provide an explanation for any destroyed or missing study drug or study materials (or must be referenced in their institution SOPs).

Unused study drug may be destroyed on site, per the site's standard operating procedures, but only after BioMarin has granted approval for drug destruction. The monitor must account for all study drug in a formal reconciliation process prior to study drug destruction. All study drug destroyed on site must be documented. Documentation must be provided to BioMarin or

designee and retained in the Investigator study files. If a site is unable to destroy study drug appropriately, the site can return unused study drug to BioMarin upon request. The return of study drug or study drug materials must be accounted for on a Study Drug Return Form provided by BioMarin.

All study drug and related materials should be stored, inventoried, reconciled, and destroyed or returned according to applicable state and federal regulations and study procedures. For additional information, please refer to the Study Pharmacy Manual.

9.6 Dietary or Other Protocol Restrictions

There are no dietary or other protocol restrictions for this study. Alcohol should be avoided for the first 26 weeks of the study, and particularly within 48 hours prior to lab work.

Subjects should be advised to abstain from any blood or sperm donation after BMN 270 infusion, until there is no further evidence of vector shedding from PCR analysis of samples.

9.7 Efficacy and Safety Variables

9.7.1 Efficacy and Safety Measurements Assessed

The SoA (Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5) describe the timing of required evaluations.

9.7.2 Efficacy Variables

9.7.2.1 FVIII Activity

Efficacy (response to treatment) will be defined as FVIII activity ≥ 5 IU/dL at Week 26 following BMN 270 infusion.

Values for FVIII activity will be excluded from analysis if obtained within 72 hours since the last infusion of exogenous FVIII protein concentrates. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; The 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy.

In the event of an FVIII activity level decline during the study:

- If FVIII activity has declined at least 20% from the peak but less than 35% and has declined for at least 2 consecutive assessments, FVIII activity and LTs should be repeated every 7 days until FVIII activity is stable or increasing

- If FVIII activity has declined >35% from the peak and has declined for at least 2 consecutive assessments, FVIII activity and LTs should be repeated every 72 hours until FVIII activity is stable or increasing.

Note that fluctuations in FVIII activity are common, and if no clear trend indicating a decline in FVIII activity is observed, then this additional testing may be deferred (upon consultation between the Investigator and the Medical Monitor) until either a more clear trend of decline has been demonstrated or until the FVIII activity levels stabilize or increase.

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after either Week 26 or Week 52, as applicable and at the discretion of the Investigator.

Details on collecting FVIII activity samples are included in the Laboratory Manual.

9.7.2.2 Factor VIII Replacement Therapy/Bleeding Episodes

Additional efficacy variables are:

- Change of annualized utilization (IU/kg) of exogenous FVIII replacement therapy during Week 5 to Week 26 post BMN 270 infusion from the baseline utilization of exogenous FVIII replacement therapy.
- Change of ABRs requiring exogenous FVIII replacement treatment during Week 5 to Week 26 of the study post BMN 270 infusion from the baseline ABR.

During the study, subjects will be asked at each study visit to report the use of factor replacement therapy and the number of bleeding episodes since the previous visit. This information will be captured on the subject's diary or other subject records.

Subjects are strongly encouraged to immediately consult Investigator for guidance regarding exogenous FVIII administration for suspected bleeds or bleeding episodes within the first 6 weeks post BMN 270 infusion.

In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis.

9.7.2.3 Patient-Reported Outcomes (PRO)

The Haemo-QoL-A questionnaire is a validated hemophilia-specific health-related quality of life questionnaire for adults (Rentz, 2008). It consists of 41 questions covering six domains (Physical Functioning, Role Functioning, Worry, Consequences of Bleeding, Emotional

Impact and Treatment Concerns). Items are answered on a 6-point Likert-type scale, ranging from 0 (None of the time) to 5 (All of the time). Higher scores mean better health-related quality of life or less impairment for a particular subscale (Haemo-QoL Study Group, 2017). Details regarding the Haemo-QoL-A assessment will be included in the Study Reference Manual.

The EQ-5D-5L instrument is a self-reported questionnaire designed to measure general health status (The EuroQol Group, 1990) (Brooks, 1996). The EQ-5D-5L is composed of 2-parts: a descriptive system that assesses 5 levels of perceived problems (mobility, self-care, usual activities, pain/discomfort, and anxiety/depression) in 5 dimensions and the EQ visual analogue scale (EQ VAS) assessment for overall health. A sample copy of the EQ-5D-5L and additional information are provided in the Study Reference Manual.

The Haemophilia Activities List (HAL) measures the impact of hemophilia on self-perceived functional abilities in adults (van Genderen, 2006). The instrument consists of multiple domains including lying/sitting/kneeling/standing, leg and arm function, use of transportation, self-care, household tasks, and leisure activities where subjects are asked to rate their level of difficulty with activities of daily living on a 6-point Likert-type scale from 1 (Impossible) to 6 (Never). For some items, subjects are given the choice to answer 'Not applicable'. A sample copy of the HAL and additional information are provided in the Study Reference Manual.

The Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) instrument is designed to measure the effect of disease symptom severity on work productivity and classroom productivity (if applicable) (Recht, 2014). The WPAI+CIQ:HS questionnaire yields scores related to work/classroom absenteeism, reduced on-the-job effectiveness, overall work/classroom impairment, and activity impairment. WPAI+CIQ:HS outcomes are expressed as impairment percentages, with higher numbers indicating greater impairment and less productivity (Reilly, 2002). A sample copy of the WPAI+CIQ:HS and additional information are provided in the Study Reference Manual.

9.7.3 Immunogenicity

Immunogenicity assays will be performed on plasma and PBMCs. The assays will include detection of anti-AAV5 vector capsid and anti-FVIII total antibodies, as well as determination of neutralizing antibodies against FVIII (FVIII inhibitors) and against the AAV5 vector capsid (Transduction Inhibitors, TI). FVIII Inhibitors will be assessed using the Bethesda assay with Nijmegen modification. Any abnormality of the liver parameters will

lead to a retrospective immunogenicity assessment to evaluate FVIII-and vector capsid-specific cellular immunogenicity. FVIII- and vector capsid-specific cellular immunity will be assessed by stimulated cytokine secretion using an ELISpot assay performed on collected PBMCs.

9.7.4 Pharmacodynamics

The FVIII protein concentration and activity level as measured by a validated immunoassay and by a validated FVIII activity assay, respectively, will be used for plasma profiles; FVIII protein and activity will be used to determine PD parameters.

9.7.5 Exploratory Assessments

A cytokine bead array assay assessment will be performed at Baseline and then weekly through Week 26.

In addition, blood samples will be collected from subjects at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5 to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV5 gene transfer, and to develop assays used for these evaluations. Subject may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A.

All biomarker samples collected in this study may be used for exploratory biomarker research, including evaluation of additional biomarkers not specifically listed in the protocol. In addition, samples collected for other purposes in this study may be used for exploratory research once testing for the primary purpose has been completed.

9.7.6 Safety Variables

Safety in this study will be determined from evaluation of AEs, clinical laboratory assessments with a particular attention to the liver function, vital signs assessments, physical examinations, and immunogenicity.

9.7.6.1 Adverse Events

The determination, evaluation and reporting of AEs will be performed as outlined in Section 10.

9.7.6.2 Clinical Laboratory Assessments

The scheduled clinical laboratory tests are listed in Table 9.7.6.2.1. Refer to the Study Reference Manual for instructions on obtaining and shipping samples. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the

central laboratory. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample.

Any abnormal test results determined to be clinically significant by the Investigator should be repeated (at the Investigator's discretion) until: (1) the cause of the abnormality is determined; (2) the value returns to baseline or to within normal limits; or (3) the Investigator determines that the abnormal value is no longer clinically significant.

All abnormal clinical laboratory results should be initialed and dated by an Investigator, along with a comment regarding whether or not the result is clinically significant. Each clinically significant laboratory result should be recorded as an adverse event.

The diagnosis, if known, associated with abnormalities in clinical laboratory tests that are considered clinically significant by the Investigator will be recorded on the AE eCRF.

Table 9.7.6.2.1: Clinical Laboratory Tests

Blood Chemistry	Hematology	Urine Tests	Coagulation Screen including:
Albumin	Hemoglobin	Appearance	APTT
BUN	Hematocrit	Color	PT/INR
Calcium	WBC count	pH	TT
Chloride	RBC count	Specific gravity	
Total cholesterol	Platelet count	Ketones	
CPK	Differential cell count	Protein	
Creatinine	RBC indices (MCV and MCH)	Glucose	
CRP	ABO blood typing*	Bilirubin	
Glucose		Nitrite	
Phosphorus		Urobilinogen	
Potassium		Hemoglobin	
Total protein			
Sodium			
Uric Acid			

BUN, blood urea nitrogen; CPK, creatinine phosphokinase; CRP, C-reactive protein; PT, prothrombin time; APTT, activated partial thromboplastin time; RBC, red blood cell; WBC, white blood cell; TT, thrombin time; INR, international normalized ratio; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin.

* ABO blood typing assessment should be performed as part of the hematology assessment (at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study).

In addition to scheduled clinical laboratory assessments, a fasting blood lipid panel (including triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) will be assessed at the BMN 270 infusion visit. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit.

In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.

9.7.6.3 Liver and Hepatitis Testing

Subjects will be screened for evidence of previous or active hepatitis B or hepatitis C infection at Screening; hepatitis B screening should include HBsAg, HBsAb, and HBcAb. Subjects with documented results showing an absence of active hepatitis B or hepatitis C infection (as measured by positive surface antigen for hepatitis B or positive RNA testing for hepatitis C) 30 days prior to providing signed informed consent do not need to repeat those tests during the screening period.

Evidence of ongoing hepatitis B or hepatitis C infection is exclusionary. Subjects with history of hepatitis B or hepatitis C infection prior to study entry will be tested for hepatitis B and hepatitis C reactivation at Week 16. Subjects with a history of hepatitis B or hepatitis C will be asked for information about the treatments received as part of their medical history assessment at Screening.

Subjects with a previous history of hepatitis B or hepatitis C who receive therapeutic oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.6.

A liver ultrasound and liver tests (LTs) during Screening will identify any significant hepatic dysfunction.

Liver tests will be monitored on a regular basis; at each time point specified in the SoA, the following LTs should be assessed:

Table 9.7.6.3.1: Liver Tests

Liver Tests (LTs)			
Alkaline Phosphatase	AST (SGOT)	Total Bilirubin	LDH
ALT (SGPT)	Direct Bilirubin	GGT	

ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma-glutamyltransferase; LDH, lactate dehydrogenase; SGOT, serum glutamic-oxaloacetic transaminase; SGPT, serum glutamic-pyruvic transaminase

Elevated ALT levels (above the upper limit of normal range) should be evaluated according to the following plan:

Table 9.7.6.3.2: Evaluation of ALT Elevations

ALT Level	Work-Up
Above ULN and <1.5x ULN	• Continue to monitor LTs and FVIII per protocol (repeat within 7 days if next protocol scheduled visit is >7 days from the time of the reported ALT elevation) • Consider evaluation to rule out alternative etiology (eg, concomitant medications, viral or autoimmune hepatitis, alcohol use, recreational drug use, special diets, strenuous exercise, prior and/or concurrent illnesses, exposure to environmental and/or industrial chemicals, etc.) (refer to Table 9.7.6.3.3) • If ALT is > ULN & > 2x baseline in 2 consecutive assessments within 72 hours and alternative etiologies have been ruled out, start oral corticosteroids upon consultation with the Medical Monitor (refer to Section 9.4.8.2)
1.5 - <3x ULN	• Repeat LTs and FVIII within 72 hours • Continue to monitor LTs weekly until ALT is stable or improving • Evaluate and rule out alternative etiologies (as above) • Consult with Medical Monitor • If ALT is ≥ 1.5x ULN in 2 consecutive assessments within 72 hours and alternative etiologies have been ruled out, start oral corticosteroids (refer to Section 9.4.8.2)
≥3x ULN	• Consult with Medical Monitor • Evaluate and rule out alternative etiologies (as above) • Repeat LTs and FVIII within 48 hours, and continue with monitoring of LTs at least twice weekly for as long as the subject's ALT remains ≥ 3x ULN • If ≥3x ULN in 2 consecutive assessments within 48 hours, start oral corticosteroids (refer to Section 9.4.8.2) • Obtain other possibly relevant laboratory evaluations (albumin, PT/INR, CRP, etc.) • Obtain complete blood count with differential to assess for eosinophilia • Obtain PBMC to evaluate potential immune response (prior to starting oral corticosteroids) • If no improvement in 14 days, consider gastroenterology and/or hepatology consult, abdominal workup, imaging (including MRI or ultrasound), and/or liver biopsy as appropriate

When ruling out alternative viral or autoimmune hepatitis as part of the elevated ALT workup, the following tests should be performed:

Table 9.7.6.3.3: Viral and Autoimmune Hepatitis Testing

Viral Hepatitis Workup Testing	Autoimmune Hepatitis Workup Testing
Hepatitis A	Smooth muscle antibody
Hepatitis B	Mitochondrial antibody
Hepatitis C	Liver/kidney microsomal antibodies
Hepatitis E	Antinuclear antibody (ANA) HEP-2
Cytomegalovirus (CMV)	
Epstein-Barr virus (EBV)	
Herpes simplex virus (HSV) 1 & 2	

9.7.6.4 HIV Testing

HIV testing will be performed at Screening. Subjects with documented negative results within the last 30 days prior to screening do not need to be retested.

9.7.6.5 Vital Signs, Physical Examinations and Other Observations Related to Safety

Vital signs will include seated systolic and diastolic blood pressure, heart rate, respiration rate, and temperature. Any clinically significant change in vital signs will be recorded as an AE.

Systolic blood pressure, diastolic blood pressure, heart rate, respiration rate, and temperature will be assessed at Screening, Baseline, and at the beginning of each visit during the Post-Infusion Follow-Up and Long-Term Follow-Up periods. On the day of the BMN 270 Infusion, vital signs will be monitored prior to infusion, during the infusion every 15 minutes (± 5 minutes), following the infusion hourly (± 5 minutes) for at least 8 hours during the subject's stay in the clinic. Any abnormal vital sign assessments should be repeated, and both values should be recorded in the eCRF.

A complete physical examination should be performed at Screening, Week 26, Week 52, and at the End of Year visits. A complete physical examination will include general appearance (head, eyes, ears, nose, and throat), cardiovascular, dermatologic, lymphatic, respiratory, gastrointestinal, genitourinary, musculoskeletal, and neurologic systems.

At other visits, brief physical examinations may be performed at the discretion of the Investigator based on the subject's clinical condition. A brief physical examination will include general appearance, cardiovascular, dermatologic, respiratory, gastrointestinal, musculoskeletal, and neurologic assessments. Particular attention should be given to signs of bleeding, as well as assessing possible hemarthroses.

Height will be recorded at Screening only. Weight will be recorded at Screening and then every 4 weeks thereafter through Week 20, at Week 26, every 4 weeks through Week 52, and then at the second Q12W visit each year and at every End of Years visit during Years 2-5.

9.7.6.6 Vector Shedding

During the Post-Infusion Follow-Up period, subjects will undergo testing of various bodily samples to look for evidence of vector shedding for possible viral transmission. Bodily fluids will be tested by polymerase chain reaction (PCR). Fluids tested will include:

- Blood
- Saliva
- Semen
- Urine
- Stool

Vector shedding will also be extensively studied in the present clinical trial, at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5. Testing will continue until at least 3 negative results are obtained. Testing of semen will continue at least through Week 12, even if 3 consecutive negative results have been recorded in that compartment prior to that time point. Subjects who have not had 3 consecutive negative semen samples by Week 26 should continue to have PCR testing in semen every 4 weeks until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor). Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule (refer to Section 12.5.3) but who have not cleared vector shedding from all fluids must still provide samples for assessment until vector shedding has cleared: at Weeks 32, 36, 40, 44, 48, and 52 (during Year 1), every 4 weeks (during Year 2), or every 6 weeks (during Years 3-5).

Samples may be fractionated prior to shedding analysis in order to better characterize the presence, structure, and location of vector DNA and/or vector capsid within each matrix. If needed, the fractionation may be performed with samples collected specifically for shedding analysis (saliva, blood, semen, urine, feces). Alternatively, the vector DNA characterization during shedding analysis may utilize already fractionated exploratory samples obtained from the above biofluids, such as exploratory plasma samples, exploratory PBMC samples, and red blood cells recovered during PBMC/plasma isolations.

Fractionation of semen to collect purified sperm separately from non-sperm cells may be performed in parallel at any visit where semen samples are collected. The shedding analysis

of a fractionated semen sample will only be performed if vector DNA was detected in the whole semen sample for the same visit. Fractionation of semen during shedding analysis may be stopped if purified sperm tested positive for vector DNA on at least three visits, or if purified sperm tested negative for vector DNA on at least three consecutive visits.

Contraception use may need to be extended beyond 26 weeks in individual subjects based on observed vector shedding in semen. After 26 weeks, subjects may stop contraception use only if they have had 3 consecutive negative semen samples (upon consultation between the Investigator and Medical Monitor).

Details for sample collection and storage are provided in the Laboratory Manual.

10 REPORTING ADVERSE EVENTS

10.1 Safety Parameters and Definitions

Safety assessments will consist of monitoring and recording protocol-defined AEs and SAEs; measurement of protocol-specified hematology, clinical chemistry, and urinalysis variables; measurement of protocol-specified vital signs; and other protocol-defined events of special interest that are deemed critical to the safety evaluation of the study drug.

10.1.1 Adverse Events

For this protocol, an adverse event (AE) is any untoward medical occurrence in a subject, temporally associated with the use of study intervention, whether or not considered related to the study intervention.

An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of study intervention.

Events meeting the AE definition include:

- Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition.
- New conditions detected or diagnosed after study intervention administration even though it may have been present before the start of the study.
- Signs, symptoms, or the clinical sequelae of a suspected drug-drug interaction.

Events not meeting the AE definition include:

- Any clinically significant abnormal laboratory findings or other abnormal safety assessments which are associated with the underlying disease, unless judged by the investigator to be more severe than expected for the subject's condition.
- The disease/disorder being studied or expected progression, signs, or symptoms of the disease/disorder being studied, unless more severe than expected for the subject's condition.
- Medical or surgical procedure (eg, endoscopy, appendectomy): the condition that leads to the procedure is the AE.
- Situations in which an untoward medical occurrence did not occur (social and/or convenience admission to a hospital).
- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

10.1.1.1 Bleeding and Suspected Bleeding Events

All bleeding events and suspected bleeding events, regardless of the need for exogenous FVIII therapy as treatment, should be captured in subject diaries and recorded on the designated bleeding eCRF. Bleeding events and suspected bleeding events should not be reported as adverse events, with the following exception:

- All bleeding events and suspected bleeding events which meet one or more of the criteria for being serious (refer to Section 10.1) should be reported as serious adverse events (whether or not they are bleeding events that are normal sequelae of hemophilia, and whether or not they required exogenous FVIII as treatment).

10.2 Serious Adverse Events

A serious adverse event (SAE) is any untoward medical occurrence that, at any dose:

- Results in death
- Is life-threatening

  Note: Life-threatening refers to an event in which the subject was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.
- Requires inpatient hospitalization or prolongation of existing hospitalizationNote: In general, hospitalization signifies that the subject has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization, the event is serious.

  Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE.
- Results in persistent or significant disability or incapacity
- Is a congenital anomaly or birth defect in the child or fetus of a subject exposed to IP prior to conception or during pregnancy
- Is an important medical event or reaction – that, based on medical judgment, may jeopardize the subject or require medical/surgical intervention to prevent one of the other outcomes listed above (eg, anaphylaxis)

10.2.1 Events of Special Interest (EOSI)

The following EOSI need to be reported to the Sponsor within 24 hours of site awareness, irrespective of seriousness, severity or causality:

- Elevation of ALT ≥1.5x ULN or ALT > ULN & >2x baseline value, regardless of whether that elevation triggers an initiation or modification of oral corticosteroid treatment
- Thromboembolic event
- Systemic hypersensitivity, anaphylactic, or anaphylactoid reactions (refer to Appendix 1)

10.3 Methods and Timing for Capturing and Assessing Safety Parameters

10.3.1 Adverse Event Reporting Period

The study AE reporting period is as follows: After informed consent but prior to initiation of study drug, only SAEs associated with any protocol-imposed interventions will be collected. After informed consent is obtained and following infusion of study drug, the reporting period for all non-serious AEs and SAEs begins and continues for approximately 5 years or until study discontinuation/termination, whichever is longer. The criteria for determining, and the reporting of SAEs is provided in Section 10.1.

10.3.2 Eliciting Adverse Events

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and non-leading verbal questioning of the subject is the preferred method to inquire about AE occurrences. The Investigator will record all relevant AE/SAE/EOSI information in the subject's medical record and AE Case Report Form (eCRF).

10.3.3 Assessment of Seriousness, Severity, and Causality

The Investigator responsible for the care of the subject or medically qualified designee will assess AEs for severity, relationship to study drug, and seriousness (refer to Section 10.2 for SAE definitions). These assessments must be made by a study clinician with the training and authority to make a diagnosis (eg, MD/DO, physician's assistant, nurse practitioner, or DDS).

10.3.3.1 Seriousness

The Investigator will assess if an AE should be classified as "serious" based on the seriousness criteria enumerated in Section 10.2. Seriousness serves as a guide for defining regulatory reporting obligations.

10.3.3.2 Severity

Severity (as in mild, moderate, or severe headache) is not equivalent to seriousness, which is based on subject/event outcome or action criteria usually associated with events that pose a threat to a subject's life or functioning. The Investigator will determine the severity of each AE, SAE and EOSI using the NCI CTCAE v4.03. Adverse events that do not have a corresponding CTCAE term will be assessed according to the general guidelines for grading used in the CTCAE v4.03 as stated in Table 10.3.3.2.1.

Table 10.3.3.2.1: Adverse Event Grading (Severity) Scale

Grade	Description	
1	Mild: asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated	
2	Moderate: minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living (ADL)[a]	
3	Severe or medically significant but not immediately life-threatening: hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL[b]	
4	Life threatening consequences; urgent intervention indicated	Grade 4 and 5 AEs should always be reported as SAEs
5	Death related to AE	

[a] Instrumental ADL refer to the following examples: preparing meals, shopping for groceries or clothes, using the telephone, managing money.

[b] Self-care ADL refer to the following examples: bathing, dressing and undressing, feeding oneself, using the toilet, taking medications, not bedridden.

10.3.3.3 Causality

The Investigator will determine the relationship of an AE to the study drug and will record it on the source documents and AE eCRF. To ensure consistency of causality assessments, Investigators should apply the guidance in Table 10.3.3.3.1.

Table 10.3.3.3.1: Causality Attribution Guidance

Relationship	Description
Not Related	• Exposure to the IP has not occurred **OR** • The administration of the IP and the occurrence of the AE are not reasonably related in time **OR**

Relationship	Description
	• The AE is considered likely to be related to an etiology other than the use of the IP; that is, there are no facts, evidence, or arguments to suggest a causal relationship to the IP.
Related	• The administration of the IP and the occurrence of the AE are reasonably related in time **AND** • The AE could possibly be explained by factors or causes other than exposure to the IP **OR** • The administration of IP and the occurrence of the AE are reasonably related in time **AND** • The AE is more likely explained by exposure to the IP than by other factors or causes.

Factors suggestive of a causal relationship could include (but are not limited to):

- Plausible temporal relationship
- Absence of alternative explanations
- Rarity of event in a given patient or disease state
- Absence of event prior to study drug exposure
- Consistency with study product pharmacology
- Known relationship to underlying mechanism of study drug action
- Similarity to adverse reactions seen with related drug products
- Abatement of AE with discontinuation of study drug, and/or recurrence of AE with reintroduction of study drug

The Investigator's assessment of causality for individual AE reports is part of the study documentation process. Regardless of the Investigator's assessment of causality for individual AE reports, the Sponsor will promptly evaluate all reported SAEs against cumulative product experience to identify and expeditiously communicate possible new safety findings to Investigators and applicable regulatory authorities.

10.4 Procedures for Recording Adverse Events

10.4.1 Recording Adverse Events on a eCRF

Investigators should use precise medical terminology when recording AEs or SAEs on the AE eCRF. Avoid colloquialisms and abbreviations.

Record only one diagnosis, sign, or symptom per event field on the AE eCRF (eg, nausea and vomiting should not be recorded in the same entry, but as 2 separate entries).

In order to classify AEs and diseases, preferred terms will be assigned by the Sponsor to the original terms entered on the AE eCRF, using MedDRA (Medical Dictionary for Regulatory Activities) terminology.

10.4.1.1 Diagnosis versus Signs and Symptoms

The Investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE. Using accepted medical terminology, enter the diagnosis (if known). If not known, enter sign(s) and/or symptom(s). If a diagnosis subsequently becomes available, then this diagnosis should be entered on the AE (or SAE, as appropriate) eCRF, replacing the original entries where appropriate.

10.4.1.2 Adverse Events Occurring Secondary to Other Events

In general, AEs occurring secondary to other events (eg, cascade events) should be identified by their primary cause. For example, if severe diarrhea is known to have resulted in dehydration, it is sufficient to record only diarrhea as an AE or SAE on the AE eCRF. However, medically important events that may be linked and/or separated in time should be recorded as independent events on the AE eCRF. For example, if severe hemorrhage leads to renal failure, both events should be recorded separately on the AE eCRF.

10.4.1.3 Persistent or Recurrent Adverse Events

A persistent AE (duration of adverse event > 7 days) is one that extends continuously, without resolution, between subject evaluation time points. Events that change in severity necessitate the recording of an additional AE. AEs that do not have a change in severity should be recorded only once on the eCRF.

A recurrent AE is one that occurs and resolves between subject evaluation time points, but then subsequently recurs. All recurrences of the AE should be recorded on the AE eCRF. For example, if a subject has an adverse event of ALT increased that subsequently resolves,

but the subject's ALT increases again, that should be reported as two adverse events – the initial ALT increase, and the second ALT increase.

10.4.1.4 Abnormal Laboratory Values

Laboratory test results (including any local FVIII activity or liver test results) will be recorded on the laboratory results pages of the eCRF, or appear on electronically produced laboratory reports submitted directly from the central laboratory, if applicable.

Any laboratory result abnormality fulfilling the criteria for a SAE or EOSI should be reported as such, and recorded in the AE eCRF.

Any laboratory result abnormality of CTCAE Grade 4 or 5 should be recorded as an SAE in the AE eCRF.

A clinical laboratory abnormality is considered clinically significant and should be documented as an AE if not refuted by a repeat test to confirm the abnormality and **any** one or more of the following conditions is met:

- Accompanied by clinical symptoms
- Requiring a change in concomitant therapy (eg, addition of, interruption of, discontinuation of, or any other change in a concomitant medication, therapy or treatment).
- The abnormality suggests a disease and/or organ toxicity
- The abnormality is of a degree that requires active management (eg, change of dose, discontinuation of study drug, more frequent follow-up assessments, further diagnostic investigation, etc.)

This applies to any protocol and non-protocol specified safety and efficacy laboratory result from tests performed after the first dose of study medication that falls outside the laboratory reference range and meets the clinical significance criteria.

This does not apply to any abnormal laboratory result that falls outside the laboratory reference range but that does not meet the clinical significance criteria (these will be analyzed and reported as laboratory abnormalities), those that are considered AEs of the type explicitly exempted by the protocol, or those which are a result of an AE that has already been reported.

For purposes of this study, laboratory tests showing a decreased level of FVIII activity should not be reported as adverse events unless there is an impact to clinical outcomes (eg, increased rate of bleeding, worsening of joint disease).

10.4.1.5 Pre-existing Conditions

A pre-existing condition is one that is present prior to administration of BMN 270. Such conditions should be recorded as medical history on the appropriate eCRF.

A pre-existing condition should be recorded as an AE or SAE during the study **only** if the frequency, intensity, or character of the condition worsens during the study period. It is important to convey the concept that a pre-existing condition has changed by including applicable language in the verbatim description of the event (eg, *more frequent* headaches).

10.4.1.6 General Physical Examination Findings

At screening, any clinically significant abnormality should be recorded as a pre-existing condition (refer to Section 10.4.1.5). During the study, any new clinically significant findings and/or abnormalities discovered on physical examination that meet the definition of an AE (or an SAE) must be recorded and documented as an AE or SAE on the AE eCRF.

10.4.1.7 Hospitalization, Prolonged Hospitalization, or Surgery

Any AE that results in hospitalization or prolonged hospitalization should be documented and reported as an SAE unless specifically instructed otherwise in this protocol (refer to Section 10.2).

There are some hospitalization scenarios that do not require reporting as an SAE when there is no occurrence of an AE. These scenarios include planned hospitalizations or prolonged hospitalizations to:

- Perform a protocol-mandated efficacy measurement
- Undergo a diagnostic or elective surgical procedure for a pre-existing medical condition that has not worsened
- Insert an in-dwelling IV catheter (such as a Port-a-Cath or other brand, if applicable) for administration of study drug or FVIII replacement therapy
- Receive scheduled therapy (study drug or otherwise) for the study indication

10.4.1.8 Deaths

All deaths that occur during the AE reporting period (refer to Section 10.3.1), regardless of attribution, will be recorded on the AE eCRF and expeditiously reported to the Sponsor as an SAE.

When recording a death, the event or condition that caused or contributed to the fatal outcome should be recorded as the single medical concept on the eCRF. If the cause of death

is unknown and cannot be ascertained at the time of reporting, record “Unexplained Death” or “Death of Unknown Cause” on the AE eCRF.

10.4.1.9 Pregnancy

Although not an AE per se, pregnancy in the partner of a subject taking trial medication should be reported expeditiously to the Sponsor to facilitate outcome monitoring by the Sponsor. Pregnancy in partner should be reported during the period up to 5 years after viral infusion.

Pregnancy in a partner should be reported within 24 hours of the site becoming aware of the pregnancy by entering the information on the Pregnancy eCRF and submitting to BPV within 24 hours of the site becoming aware of the event. The Investigator must make every effort to follow the subject’s partner (with that partner’s consent) through resolution of the pregnancy (delivery or termination) and to report the resolution on the Pregnancy Follow-up eCRF. In the event of pregnancy in the partner of a study subject, the Investigator should make every reasonable attempt to obtain the woman’s consent for release of protected health information.

Abortion, whether therapeutic or spontaneous, should always be classified as an SAE (as the Sponsor considers these to be medically significant), recorded on the AE eCRF, and expeditiously reported to the Sponsor as an SAE.

10.5 Reporting Requirements

10.5.1 Expedited Reporting Requirements

All SAEs and EOSI that occur during the course of the AE Reporting Period (refer to Section 10.3.1), whether or not considered related to study drug, must be reported by entering the information in the AE eCRF and submitting to BPV within 24 hours of the site becoming aware of the event. Investigators should not wait to collect information that fully documents the event before notifying BPV of an SAE. BioMarin may be required to report certain SAEs to regulatory authorities within 7 calendar days of being notified about the event; therefore, it is important that Investigators submit any information requested by BioMarin as soon as it becomes available. IND safety reports will be submitted within 7 calendar days for unexpected fatal or life-threatening unexpected suspected adverse reactions (SUSARs) and within 15 calendar days for other non-life-threatening SUSARs.

The Sponsor is responsible for identifying, preparing and reporting all SUSARs to the relevant competent authorities, ethics committees and Investigators in accordance with the requirements identified in the Clinical Trials Regulations.

If the EDC is unavailable, all SAEs should be reported to BPV by completing the SAE Report Form and faxing or emailing the completed form to BPV within 24 hours of the site becoming aware of the event. Once the EDC is available, the information should be entered in the AE eCRF.

10.5.2 Institutional Review Board or Independent Ethics Committee Reporting Requirements

Reporting of SAEs to the Independent Ethics Committee (EC) or Institutional Review Board (IRB) will be done in compliance with the standard operating procedures and policies of the IRB/EC and with applicable regulatory requirements. Adequate documentation must be obtained by BioMarin showing that the IRB/EC was properly and promptly notified as required.

10.6 Follow-up of Subjects after Adverse Events

After the initial AE/SAE/EOSI report, the Investigator is required to proactively follow each subject at subsequent visits/contacts. All AEs/SAEs/EOSI will be followed until resolution, stabilization, the event is otherwise explained, or the subject is lost to follow-up. Resolution of AEs/SAEs/EOSI (with dates) should be documented on the AE eCRF and submitted to BioMarin Pharmacovigilance and in the subject's medical record to facilitate source data verification.

For some SAEs and EOSI, the Sponsor may follow up by telephone, fax, electronic mail, and/or a monitoring visit to obtain additional case details (eg, hospital discharge summary, consultant report, or autopsy report) deemed necessary to appropriately evaluate the SAE or EOSI report.

10.7 Post-Study Adverse Events

At the last scheduled visit, the Investigator should instruct each subject to report, to the Investigator and/or to BPV directly, any subsequent SAEs that the subject's personal physician(s) believes might be related to prior study drug.

The Investigator should notify the study Sponsor of any death or SAE occurring at any time after a subject has discontinued or terminated study participation, if the Investigator believes that the death or SAE may have been related to prior study drug. The Sponsor should also be notified if the Investigator should become aware of the development of cancer or of a congenital anomaly in a subsequently conceived offspring of a subject that participated in this study.

10.8 Urgent Safety Measures

The regulations governing clinical trials state that the Sponsor and Investigator are required to take appropriate urgent safety measures to protect subjects against any immediate hazards that may affect the safety of subjects, and that the appropriate regulatory bodies should be notified according to their respective regulations. According to the European Union (EU) Clinical Trial Directive 2001/20/EC, "…in the light of the circumstances, notably the occurrence of any new event relating to the conduct of the trial or the development of the investigational medicinal product where that new event is likely to affect the safety of the patients, the Sponsor and the Investigator shall take appropriate urgent safety measures to protect the patients against any immediate hazard. The Sponsor shall forthwith inform the competent authorities of those new events and the measures taken and shall ensure that the IRB/EC is notified at the same time."

The reporting period for these events which may require the implementation of urgent safety measures is the period from the time of signing of the ICF through the completion of the last study visit or at the Early Termination Visit (ETV). Investigators are required to report any events which may require the implementation of urgent safety measures to BioMarin within 24 hours.

Examples of situations that may require urgent safety measures include discovery of the following:

- Lack of study scientific value, or detrimental study conduct or management
- Discovery that the quality or safety of the IP does not meet established safety requirements

10.9 BioMarin Pharmacovigilance Contact Information

Contact information for BioMarin Pharmacovigilance is as follows:

BioMarin Pharmaceutical Inc.

Address	105 Digital Drive Novato, CA 94949
Phone:	PI
Fax:	PI
E-mail:	drugsafety@bmrn.com

The Investigator is encouraged to discuss with the medical monitor any AEs for which the issue of seriousness is unclear or questioned. Contact information for the medical monitor is as follows:

Name:	PI, MA MB BChir MSc
Address:	Biomarin (UK) Ltd. 10 Bloomsbury Way London WC1A 2SL
Phone	PI (office) PI (mobile)
E-mail:	PI

11 APPROPRIATENESS OF MEASUREMENTS

The measures of efficacy to be used in this study are standard, ie, widely used and generally recognized as reliable, accurate, and relevant (able to discriminate between effective and ineffective agents). The measures of safety used in this study are routine clinical and laboratory procedures.

The chromogenic substrate FVIII assay and the one-stage clotting FVIII assay are both validated and utilize CE marked reagents. The exploratory FVIII activity assay will be used for exploratory purposes only.

12 STUDY PROCEDURES

12.1 Prestudy

An ICF must be signed and dated by the subject, the Investigator or designee and witness (if required) before any study-related procedures are performed.

12.2 Screening Visit(s)

Screening assessments should be performed within 28 days of BMN 270 infusion (and must be performed within 42 days prior to BMN 270 infusion), while baseline assessments will take place within 7 days prior to BMN 270 infusion (Day 1). Should the screening visit occur within 30 days of the drug infusion, physical examination, vital signs, blood chemistry, LTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline.

The following procedures will be performed during the Screening Period:

- Demographics (age, sex, race, ethnicity)
- Full medical history, including hemophilia A history, Hepatitis B, Hepatitis C, and HIV. Subjects with a history of hepatitis B or hepatitis C will be asked for information about the treatments received. Any prior pharmacokinetics information obtained while the subject was receiving prophylactic or on-demand FVIII therapy prior to the study should also be collected.
- Complete Physical Examination
- Height and weight
- Vital Signs (systolic and diastolic blood pressure, heart rate, respiration rate, and temperature)
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information) for the previous 12 months
- Distribution of subject diaries and training in diary completion
- Electrocardiogram
- Liver Ultrasound
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay
    - hFVIII coagulation activity exploratory assay (collected but not tested prior to enrollment)

- hFVIII inhibitors (Bethesda assay with Nijmegen modification)
- hFVIII total antibody assay (collected but not tested prior to enrollment)
- hFVIII antigen assay (collected but not tested prior to enrollment)

- Blood sample for AAV5 total antibody (TAb) assay
  - Screening sample will be tested in an AAV5 Tab pre-screening assay specifically developed for enrolment purposes.
- Screen for Hepatitis B, Hepatitis C, and HIV if required (subjects with documented negative results 30 days prior to informed consent being obtained do not need to be retested)
  - Hepatitis B screening should include HBsAg, HBsAb, and HBcAb
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)
- Blood samples for Biomarker testing (may include HLA genotyping and FVIII genotyping status)

12.2.1 "Smart Rescreening" Visit

Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once.

If a patient has to be screened again because the original assessments have fallen out of the 28 + 14 day period allowed for Screening (refer to Section 12.2), then only the following assessments need to be performed (rather than the full list indicated in Section 12.2) for the patient to be successfully re-screened for the study:

- Vital signs
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- hFVIII Assays (only the hFVIII inhibitor level (Bethesda assay with Nijmegen modification))
- AAV5 Total Antibody assay

- Smart Re-screening sample will be tested in an AAV5 Tab pre-screening assay specifically developed for enrolment purposes.

- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)

12.3 Baseline Visit

Baseline values will be recorded from 1 to 7 days prior to the infusion visit. The following procedures will be performed during the Baseline Period:

- Brief physical examination
- Vital signs
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay
    - hFVIII coagulation activity exploratory assay
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
    - hFVIII total antibody assay
    - hFVIII antigen assay
- Von Willebrand Factor Antigen (VWF:Ag)
- Blood sample for AAV5 Total Antibody assay
    - Baseline sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
    - ABO blood typing assessment should be performed as part of the hematology assessment (at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study).
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)
- PBMC collection for CTL baseline

- Direct Thrombin test
- Blood sample for AAV5 TI assay
- Thrombin Generation Assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools
- Exploratory biomarker assessments
- Cytokine bead array assay
- Hypersensitivity blood assessments
- Haemo-QoL-A assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.4 Treatment Visit/BMN 270 Infusion Visit (Day 1)

There will be one infusion visit for each subject. Subjects will remain in the clinic for at least 24 hours for the BMN 270 Infusion Visit. The following procedures will be performed during the BMN 270 Infusion Visit:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Blood sample for AAV5 TAb Assay (sample collected pre-infusion for analysis)
    - Infusion Day sample will be tested in an AAV5 Tab pre-screening assay specifically developed for enrolment purposes.
- Blood sample for AAV5 TI assay (sample collected pre-infusion for analysis)
- Fasting blood sample for future exploratory analysis (sample collected pre-infusion)
- Fasting lipid panel (blood triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) (sample collected pre-infusion)
    - Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit.
- BMN 270 Infusion
- Vital Signs

- Vital signs will be recorded prior to BMN 270 infusion and then every 15 minutes (± 5 minutes) during BMN 270 infusion. Following infusion, vital signs will be monitored every 1 hour (± 5 minutes) for at least 8 hours during the subject's stay in the clinic.

- PCR of vector DNA in blood, saliva, urine, semen, and stools
  - Collection of samples for PCR testing should occur between 2 and 24 hours after the BMN 270 infusion has been completed

In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.

12.5 BMN 270 Infusion Follow-Up Visits

12.5.1 Week 1

During Week 1, the subject will be assessed on Study Day 2, Study Day 4, and Study Day 8.

12.5.1.1 Week 1, Study Day 2

On Study Day 2, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
- PCR of vector DNA in blood, saliva, urine, semen, and stools
  - Separate collection on Day 2 is not required if vector shedding samples were collected on Day 1 post-BMN 270 infusion; if collection on Day 2 is needed, it must occur within 24 hours of the time of the BMN 270 infusion.

- Blood sample for AAV5 TAb Assay (sample collected pre-infusion for analysis)
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection
- Cytokine bead array assay

12.5.1.2 Week 1, Study Day 4

On Study Day 4, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
- PCR of vector DNA in blood, saliva, urine, semen, and stools

12.5.1.3 Week 1, Study Day 8

On Study Day 8, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- Samples for FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level
    - hFVIII antigen assay

- PCR of vector DNA in blood, saliva, urine, semen, and stools
- Cytokine bead array assay

12.5.2 Weeks 2-26

After Week 1 (Day 8), subjects will return to the study site once a week (± 48 hours) during Weeks 2-26.

12.5.2.1 Once per week (Weeks 2 through 26)

The following procedures will be performed once per week from Weeks 1 through 26:

- Brief physical examination (complete physical examination at Week 26)
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are >1.5x ULN) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN..
- Samples for FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level
    - hFVIII antigen assay

12.5.2.2 Every Other Week (Weeks 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, and 26)

The following procedures will be performed every other week (at Weeks 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, and 26):

- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection

12.5.2.3 Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26

The following procedures will be performed at Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26:

- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Collection to occur until at least 3 consecutive negative results are obtained. Semen samples should continue to be collected and tested through Week 12, even if 3 consecutive negative results in that compartment have been recorded prior to that time point.

12.5.2.4 Weeks 2, 4, 6, 8, 10, 12, 16, 20, and 26

The following procedures will be performed at Weeks 2, 4, 6, 8, 10, 12, 16, 20, and 26:

- hFVIII total antibody assay

12.5.2.5 Weeks 2, 4, 6, 8, 14, 20, and 26

The following procedures will be performed at Weeks 2, 4, 6, 8, 14, 20, and 26:

- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)

12.5.2.6 Weeks 2, 6, 10, 14, 18, 22, and 26

The following procedures will be performed at Weeks 2, 6, 10, 14, 18, 22, and 26:

- Exploratory biomarker assessments

12.5.2.7 Weeks 2, 6, 10, 14, 20, 24, and 26

The following procedures will be performed at Weeks 2, 6, 10, 14, 20, 22, and 26:

- Cytokine bead array assay

12.5.2.8 Weeks 4, 8, 12, 16, 20, and 26

The following procedures will be performed at Weeks 4, 8, 12, 16, 20, and 26:

- Weight
- VWF:Ag

12.5.2.9 Weeks 12 and 26

The following procedures will be performed at Weeks 12 and 26:

- Urine tests (to be performed locally)
- Haemo-QoL-A QoL assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)

- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.5.2.10 Weeks 13 and 26

The following procedure will be performed at Weeks 13 and 26:

- Direct thrombin activity test

12.5.2.11 Week 16

The following procedure will be performed at Week 16:

- Testing for reactivation of hepatitis B and hepatitis C (only in subjects with evidence of prior exposure to hepatitis B and/or hepatitis C)
    - Subjects who receive therapeutic oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.6.

12.5.2.12 Weeks 20, 24, and 26

The following procedure will be performed at Weeks 20, 24, and 26:

- Thrombin Generation Assay

12.5.3 Post-Infusion Follow-Up – Weeks 27-52

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 26 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 26 of the study by attending only the Week 36, 44, and 52 visits during the remainder of Year 1. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in all fluids must still provide samples at Week 32, Week 40, and Week 48, if necessary (but do not need to do other scheduled assessments on that date).

During Weeks 27-36, subjects will return to the study site weekly (± 48 hours). During Weeks 37-52, subjects will return to the study site every 2 weeks (Week 38, 40, 42, 44, 46, 48, 50, and 52) (± 1 week). At these visits, the following procedures will be completed:

12.5.3.1 Every Visit

At every visit (Weeks 27-36, 38, 40, 42, 44, 46, 48, 50, and 52), the following procedures will be performed:

- Physical examination
    - Brief physical examination should be performed at all weeks except Week 52, when a complete physical examination should be performed
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
        - FVIII coagulation activity exploratory assay
        - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
        - FVIII protein assay

12.5.3.2 Weeks 28, 30, 32, 34, 36, 44, and 52

At Weeks 28, 30, 32, 34, 36, 44, and 52, the following procedure will be performed:

- PBMC collection

12.5.3.3 Weeks 32, 36, 44, and 52

At Weeks 32, 36, 44, and 52, the following procedures will be performed:

- Weight
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.4.8.2.1)
- FVIII antibody titer
- AAV5 TAb Assay
- AAV5 TI Assay

- Cytokine bead array assay

12.5.3.4 Weeks 32, 36, 40, 44, 48, and 52

At Weeks 32, 36, 40, 44, 48, and 52, the following procedures will be performed:

- Exploratory biomarker assessments
- TGA Assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Sample testing to occur until at least 3 consecutive negative sample results have been obtained. Subjects who have not had 3 consecutive negative semen samples by Week 52 should continue to have PCR testing of semen every 4 weeks until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).
    - Subjects considered to be treatment failures must continue to provide samples for PCR assessment at these timepoints until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints)

12.5.3.5 Week 36 and 52

At Weeks 36 and 52, the following procedures will be performed:

- Urine Tests (refer to Table 9.7.6.2.1)
- Direct Thrombin test
- VWF:Ag

12.5.3.6 Week 52

At Week 52, the following procedures will be performed:

- Haemo-QoL-A assessment
- EQ-5D-5L
- HAL
- WPAI+CIQ:HS

12.6 Post-Infusion Follow-Up – Years 2-5

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion

with the Medical Monitor, follow an abbreviated visit schedule after Week 52 of the study by attending only the Q12W and End of Year visits during Years 2-5.

Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every 4 weeks (during Year 2) or every 6 weeks (during Years 3-5) until vector shedding has cleared (but do not have to perform other scheduled assessments at those study visits).

During Years 2-5 of Post-Infusion Follow-up, the following procedures will be completed:

12.6.1 Year 2 – Every 4 Weeks (not required for treatment failure)

During Year 2, every 4 weeks (±2 weeks), the following procedures will be performed:

- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
    - FVIII protein assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)
    - Sample testing during Year 2 is not required if at least 3 consecutive samples are negative during the Post-Infusion Follow-Up period in Weeks 1-52. Subjects who have not had 3 consecutive negative semen samples by Week 52 should continue to have PCR testing of semen every 4 weeks during Years 2 until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).

- Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every 4 weeks during Year 2 until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints).

12.6.2 Years 3-5 – Every 6 Weeks (not required for treatment failure)

During Years 3-5, every 6 weeks (± 2 weeks), the following procedures will be performed:

- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
- Liver Tests (refer to Table 9.7.6.3.1)
  - LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
- FVIII Assays
  - FVIII activity level (chromogenic substrate FVIII assay)
  - FVIII activity level (one-stage clotting FVIII assay)
  - FVIII coagulation activity exploratory assay
  - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
    - If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 3-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.
  - FVIII protein assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)
  - Sample testing during Years 3-5 is not required if at least 3 consecutive samples are clear by the end of Year 2. Subjects who have not had 3 consecutive negative semen samples by the end of Year 2 should continue to have PCR testing of semen every 6 weeks during Years 3-5 until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).

- Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every 6 weeks during Years 3-5 until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints).

12.6.3 Years 2-5 – Every 12 Weeks and End of Year Visits (required for all subjects)

During Years 2-5, subjects will be asked to return to the study site for visits at the following study weeks (±2 weeks):

- Year 2 – Week 64, Week 76, Week 88, Week 104
- Year 3 – Week 116, Week 128, Week 140, Week 156
- Year 4 – Week 168, Week 180, Week 192, Week 208
- Year 5 – Week 220, Week 232, Week 244, Week 260

For each of these years, the last study visit listed (Week 104, Week 156, Week 208, and Week 260) will serve as an End of Year visit.

At the every 12 week and End of Year visits, the following procedures will be performed:

- Brief physical examination (at every 12 week visits) or complete physical examination (at End of Year visits)
- Weight (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
- Liver Tests (refer to Table 9.7.6.3.1)
  - LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
- FVIII Assays
  - FVIII activity level (chromogenic substrate FVIII assay)
  - FVIII activity level (one-stage clotting FVIII assay)
  - FVIII coagulation activity exploratory assay
  - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level

  - If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 3-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.
- FVIII protein assay

- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1) (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Urine Tests (refer to Table 9.7.6.2.1) (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Vital Signs
- AAV5 TAb Assay
- AAV5 TI Assay
- FVIII antibody titer
- Haemo-QoL-A assessment (at End of Year visits only)
- EQ-5D-5L (at End of Year visits only)
- HAL (at End of Year visits only)
- WPAI+CIQ:HS (at End of Year visits only)
- Exploratory biomarker assessments
- PBMC collection
- VWF:Ag
- Direct Thrombin test
- TGA Assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)
  - Sample testing during Years 2-5 is not required if at least 3 consecutive samples are negative during the Post-Infusion Follow-Up period in Weeks 1-52. Subjects who have not had 3 consecutive negative semen samples by Week 52 should continue to have PCR testing of semen every 4 weeks (during Year 2) or every 6 weeks (during Years 3-5) until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).

12.7 Early Termination Visit

The Early Termination visit will occur on the date the subject withdraws from the study, even if the date does not correspond to a protocol-specific visit.

If a subject leaves the study prior to the Week 260 visit, the subject will be asked to return to the study site and complete an Early Termination visit. At the Early Termination visit, as many of the following assessments as possible should be done:

- Complete physical examination
- Weight
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Vital signs
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay
    - hFVIII coagulation activity exploratory assay
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
    - hFVIII antigen assay
- hFVIII total antibody assay
- Exploratory biomarker assessments
- Blood sample for AAV5 Total Antibody assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection for CTL baseline
- VWF:Ag
- Direct Thrombin test
- Thrombin Generation Assay
- Haemo-QoL-A QoL assessment
- EQ-5D-5L

- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire
- PCR of vector DNA in blood, saliva, urine, semen, and stool
    - Sample testing at the Early Termination Visit is not required if at least 3 consecutive samples are clear during the period of the subject's participation in the study.

12.8 End of Study

The study will end after the last subject yet to complete the last Long-Term Follow-Up visit (Week 260) does so, has transferred to another BMN 270 study, is withdrawn from the study, or discontinues from the study. BioMarin reserves the right to discontinue the study any time for clinical or administrative reasons and to discontinue participation of an individual Investigator or site for clinical or administrative reasons, including, but not limited to, poor enrollment or noncompliance with procedures of the protocol or GCP. In addition, the study may be terminated if, in the opinion of BioMarin, the safety of the study subjects may be compromised.

13 DATA QUALITY ASSURANCE

BioMarin personnel or designees will visit the study site prior to initiation of the study to review with the site personnel information about the IP, protocol and other regulatory document requirements, any applicable randomization procedures, source document requirements, CRFs, monitoring requirements, and procedures for reporting AEs, including SAEs.

At visits during and after the study, a CRA will monitor the site for compliance with regulatory documentation, with a focus on accurate and complete recording of data on CRFs from source documents, adherence to protocol, randomization (if applicable), SAE reporting and drug accountability records.

Sites will enter study data into eCRFs into the study EDC system. Data Quality Control will be performed by BioMarin Clinical Data Management or designee through implementation of quality control checks specified in the study data management plan and edit check specifications.

14 STATISTICAL METHODS AND DETERMINATION OF SAMPLE SIZE

14.1 Statistical and Analytical Plans

The statistical analysis plan (SAP) will provide additional details on the planned statistical analysis. Unless otherwise stated, all analyses will be performed using SAS.

14.1.1 Interim Analyses

No interim analysis is planned.

14.1.2 Procedures for Accounting for Missing, Unused and Spurious Data

Because the completeness of the data affects the integrity and accuracy of the final study analysis, every effort should be made to ensure complete, accurate, and timely data collection and, therefore, avoid missing data.

Sensitivity analyses will be conducted to assess the impact of missing data on the primary efficacy endpoint analysis. Additional details regarding the handling of missing data will be provided in the SAP.

14.2 Efficacy Analysis

The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using longitudinal methods, such as mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at $\geq$ 5 IU/dL at 26 weeks following study treatment. FVIII activity will be assessed weekly during the study period. We can only assess the true steady state of FVIII produced from BMN 270 after a minimum of 72 hours (or 5x the known half-life of the FVIII replacement therapy administered) has elapsed since the last infusion of FVIII concentrates.

14.3 Safety Analysis

The Medical Dictionary for Regulatory Activities terminology (MedDRA) will be used by the Sponsor to assign system organ class and preferred term classification to events and diseases, based on the original terms entered on the eCRF.

All AEs will be coded using the current version of MedDRA. The incidence of AEs will be summarized by system organ class, preferred term, relationship to study drug, and severity. A by-subject listing will be provided for those subjects who experience a serious AE (SAE),

including death, or experience an AE associated with early withdrawal from the study or study drug.

Clinical laboratory data will be summarized by the type of laboratory test. For each clinical laboratory test, descriptive statistics will be provided on Baseline as well as all subsequent visits. Descriptive statistics for physical examination results and vital signs will also be provided.

Analysis of neutralizing antibody response and other immunological parameters as well as vector shedding will be primarily descriptive and involve both inter-subject and intra-subject comparisons across doses.

Detailed statistical methods will be provided in the SAP.

14.4 Determination of Sample Size

The sample size is based upon clinical considerations and is sufficient to detect a strong clinical efficacy signal. Approximately 10 subjects may be dosed in the study.

14.5 Analysis Populations

The efficacy analysis set will be comprised of all subjects who have received the BMN 270 infusion.

The safety population will consist of all subjects who receive BMN 270 infusion during the study.

14.6 Changes in the Conduct of the Study or Planned Analyses

Only BioMarin may modify the protocol. Any change in study conduct considered necessary by the Investigator will be made only after consultation with BioMarin, who will then issue a formal protocol amendment to implement the change. The only exception is when an Investigator considers that a subject's safety is compromised without immediate action. In these circumstances, immediate approval of the chairman of the IRB/EC must be sought, and the Investigator should inform BioMarin and the full IRB/EC within 2 working days after the emergency occurs.

With the exception of minor administrative or typographical changes, the IRB/EC must review and approve all protocol amendments. Protocol amendments that have an impact on subject risk or the study objectives, or require revision of the ICF, must receive approval from the IRB/EC prior to their implementation.

When a protocol amendment substantially alters the study design or the potential risks or burden to subjects, the ICF will be amended and approved by BioMarin and the IRB/EC, and all active subjects must again provide informed consent.

15 DATA REVIEW BOARD

The Data Review Board (DRB) will consist of the Principal Investigators and Sponsor's Medical Monitor, as well as hemophilia expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201. The DRB will review available safety and efficacy data during the study on an ongoing basis to determine, based on emerging data and the benefit/risk profile, whether further enrollment should continue. The DRB will meet weekly until all subjects have completed Week 26, and then monthly thereafter.

Duties of the DRB include:

- Conducting an ongoing review of individual subject safety and efficacy data during the study;
- Recommending whether to proceed with enrollment of subjects at a different gating schedules based on emerging data from 270-203 and the overall risk/benefit analysis of BMN 270;
- If applicable, considering whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction.
- Making other recommendations on the conduct and reporting of the trial based on their evaluation of clinical data including institution of any pause or stopping stages.

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DRB determination of safety and efficacy in treated subjects with the following triggers for pausing further enrollment:

- any related SAE;
- any related AE with a severity > Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

16 COSTS, COMPENSATION, AND SUBJECT INJURY

There will be no charge to study subjects to be in this study. BioMarin will pay all costs of tests, procedures, and treatments that are part of this study. In addition, after IRB/EC approval, BioMarin may reimburse the reasonable cost of travel for study-related visits in accordance with BioMarin's travel and reimbursement policy. BioMarin will not pay for any hospitalizations, tests, or treatments for medical problems of any sort related solely to the study subject's disease. Costs associated with such hospitalizations, tests, and treatments should be billed and collected in the way that such costs are usually billed and collected outside the study.

The Investigator should contact BioMarin immediately upon notification that a study subject has been injured by the study drug or by procedures performed as part of the study. Any subject who experiences a study-related injury should be instructed by the Investigator to seek immediate medical treatment at a pre-specified medical institution if possible, or at the closest medical treatment facility if necessary. The subject should be given the name of a person to contact to seek further information about, and assistance with, treatment for study-related injuries. The treating physician should bill the subject's health insurance company or other third party payer for the cost of this medical treatment. If the cost of the medical treatment is not covered by health insurance or another third party that usually pays these costs, then either BioMarin or the institution may pay for reasonable and necessary medical services to treat the injuries caused by the study drug or study procedures. In some jurisdictions, BioMarin is obligated by law to pay for study-related injuries without prior recourse to third party payer billing and/or regardless of fault. If this is the case, BioMarin will comply with the law.

17 CASE REPORT FORMS AND SOURCE DOCUMENTS

Electronic case report forms will be provided for each subject. The Investigator must review and electronically sign the completed eCRF casebook to verify its accuracy.

eCRFs must be completed using a web–based application developed and validated. Study site personnel will be trained on the application and will enter the clinical data from source documentation. Unless explicitly allowed in the eCRF instructions, blank data fields are not acceptable.

In the event of an entry error, or if new information becomes available, the value will be corrected by deselecting the erroneous response and then selecting or entering the factual response. In compliance with ICH GCP Guidelines and 21 CFR Part 11, the system will require the personnel making the correction to enter a reason for changing the value. The documented audit trail will include the reason for the change, the original value, the new value, the time of the correction and the identity of the operator.

BioMarin's policy is that study data on the eCRFs must be verifiable to the source data, which necessitates access to all original recordings, laboratory reports, and subject records. In addition, all source data should be attributable (signed and dated). The Investigator must therefore agree to allow direct access to all source data. Subjects must also allow access to their medical records, and subjects will be informed of this and will confirm their agreement when giving informed consent. If direct source document verification of study data by the site monitor is prohibited by institutional policy or local law, then the Investigator must make available facilities and/or personnel to allow GCP-compliant source verification to occur. Examples of such methods include certified copies of records which have study data visible but sensitive information redacted, or other GCP-compliant means agreed between the Investigator and the Sponsor.

A site monitor designated by BioMarin will compare the eCRFs with the original source documents at the study site and evaluate them for completeness and accuracy before designating them as "Source Data Verified" (SDV). If an error is discovered at any time or a clarification is needed, the site monitor, or designee, will create an electronic query on the associated field. Site personnel will then answer the query by either correcting the data or responding to the query. The site monitor will then review the response and determine either to close the query or re–query the site if the response does not fully address the question. This process is repeated until all open queries have been answered and closed.

Before a subject's eCRF casebook can be locked, data fields must be source data verified and all queries closed. Refer to the Study Monitoring Plan for details on which fields must be source data verified. The Investigator will then electronically sign the casebook, specifying that the information on the eCRFs is accurate and complete. The Data Manager, or designee, will then set the status of the forms, visits, and the entire casebook to Locked. Upon completion of the CSR, an electronic copy of each site's casebooks will be copied to a compact disk (CD) and sent to each site for retention with other study documents.

18 STUDY MONITORING AND AUDITING

Qualified individuals designated by BioMarin will monitor all aspects of the study according to GCP and standard operating procedures for compliance with applicable government regulations. The Investigator agrees to allow these monitors direct access to the clinical supplies, dispensing, and storage areas and to the clinical files, including original medical records, of the study subjects, and, if requested, agrees to assist the monitors. The Investigator and staff are responsible for being present or available for consultation during routinely scheduled site visits conducted by BioMarin or its designees.

Members of BioMarin's GCP Compliance Department or designees may conduct an audit of a clinical site at any time before, during, or after completion of the study. The Investigator will be informed if an audit is to take place and advised as to the scope of the audit. Representatives of the FDA or other Regulatory Agencies may also conduct an audit of the study. If informed of such an inspection, the Investigator should notify BioMarin immediately. The Investigator will ensure that the auditors have access to the clinical supplies, study site facilities, original source documentation, and all study files.

19 RETENTION OF RECORDS

The Investigator must retain all study records required by BioMarin and by the applicable regulations in a secure and safe facility. The Investigator must consult a BioMarin representative before disposal of any study records, and must notify BioMarin of any change in the location, disposition or custody of the study files. The Investigator /institution must take measures to prevent accidental or premature destruction of essential documents, that is, documents that individually and collectively permit evaluation of the conduct of a study and the quality of the data produced, including paper copies of study records (eg, subject charts) as well as any original source documents that are electronic as required by applicable regulatory requirements.

All study records must be retained for at least 2 years after the last approval of a marketing application in the US or an ICH region and until (1) there are no pending or contemplated marketing applications in the U.S. or an ICH region or (2) at least 2 years have elapsed since the formal discontinuation of clinical development of the IP. The Investigator /institution should retain subject identifiers for at least 15 years after the completion or discontinuation of the study. Subject files and other source data must be kept for the maximum period of time permitted by the hospital, institution or private practice, but not less than 15 years. These documents should be retained for a longer period, however, if required by the applicable regulatory requirements or by a BioMarin agreement. BioMarin must be notified and will assist with retention should Investigator /institution be unable to continue maintenance of subject files for the full 15 years. It is the responsibility of BioMarin to inform the Investigator /institution as to when these documents no longer need to be retained.

20 USE OF INFORMATION AND PUBLICATION

BioMarin recognizes the importance of communicating medical study data and therefore encourages the publication of these data in reputable, peer-reviewed scientific journals and at seminars or conferences. The details of the processes of producing and reviewing reports, manuscripts, and presentations based on the data from this study will be described in the Clinical Trial Agreement between BioMarin and the Investigator/Institution. Consideration for authorship of all publications will be based on compliance with the Uniform Requirements for Manuscripts Submitted to Biomedical Journals ("Uniform Requirements") of the International Committee of Medical Journal Editors (ICMJE) (http://www.icmje.org/ethical_1author.html) and good publication practices (GPP).

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22 INVESTIGATOR RESPONSIBILITIES

22.1 Conduct of Study and Protection of Human Subjects

In accordance with FDA Form 1572 and/or principles of ICH E6 GCP, the Investigator will ensure that:

- He or she will conduct the study in accordance with the relevant, current protocol and will only make changes in a protocol after notifying the sponsor, except when necessary to protect the safety, rights, or welfare of subjects.
- He or she will personally conduct or supervise the study.
- He or she will inform any potential subjects, or any persons used as controls, that the drugs are being used for investigational purposes, and he or she will ensure that the requirements relating to obtaining informed consent in 21 CFR Part 50 and/or ICH E6 Sections 2.9 and 4.8 are met. As well, he or she will ensure that IRB/EC review and approval in 21 CFR Part 56 and/or ICH E6 Section 2.6 are met.
- He or she will report to the sponsor adverse experiences that occur in the course of the investigation in accordance with 21 CFR 312.64 and/or ICH E6 Section 4.11.
- He or she has read and understands the information in the Investigator's Brochure, including potential risks and side effects of the drug.
- His or her staff and all persons who assist in the conduct of the study are informed about their obligations in meeting the above commitments
- Adequate and accurate records in accordance with 21 CFR 312.62 and/or ICH E6 Section 4.9 are kept, and those records are available for inspection in accordance with 21 CFR 312.68 and/or ICH E6 Section 4.9.7.
- The IRB/EC complies with the requirements of 21 CFR Part 56, ICH E6 Section 3.0, and other applicable regulations, and conducts initial and ongoing reviews and approvals of the study. He or she will also ensure that any change in research activity and all problems involving risks to human subjects or others are reported to the IRB/EC. Additionally, he or she will not make any changes in the research without IRB/EC approval, except where necessary to eliminate apparent immediate hazards to human subjects.
- He or she agrees to comply with all other requirements regarding the obligations of clinical Investigators and all other pertinent requirements in 21 CFR Part 312 and/or ICH E6.

23 SIGNATURE PAGE

Protocol Title: A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5

Protocol Number: 270-203 Amendment 1

I have read the foregoing protocol and agree to conduct this study as outlined. I agree to conduct the study in compliance with all applicable regulations and guidelines, including E6R2 ICH, as stated in the protocol, and other information supplied to me.

Investigator Signature Date

Printed name: _____

Accepted for the Sponsor:

DocuSigned by:
PI
Signer Name: PI
Signing Reason: I approve this document
Signing Time: 10/11/2018 2:42:32 AM PDT
D81F76E32EA74F83864C63F8E75F1EBE

Medical Monitor Signature Date

Printed name: PI MA MB BChir MSc PI, Clinical Science

24 APPENDIX 1: SAMPSON'S ANAPHYLAXIS CRITERIA

According to the National Institute of Allergy and Infectious Disease/Food Allergy and Anaphylaxis Network (NIAID/FAAN) Second Symposium on the definition and management of anaphylaxis, anaphylaxis is highly likely when any one of the following 3 criteria are fulfilled:

1. Acute onset of an illness (minutes to several hours) with involvement of the skin, mucosal tissue, or both (eg, generalized hives, pruritus or flushing, swollen lips-tongue-uvula)
2. AND AT LEAST ONE OF THE FOLLOWING
    a. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced peak expiratory flow [PEF], hypoxemia)
    b. Reduced blood pressure (BP) or associated symptoms of end-organ dysfunction (eg, hypotonia [collapse], syncope, incontinence)
3. Two or more of the following that occur rapidly after exposure *to a likely allergen for that patient* (minutes to several hours):
    a. Involvement of the skin-mucosal tissue (eg, generalized hives, itch-flush, swollen lips-tongue-uvula)
    b. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced PEF, hypoxemia)
    c. Reduced BP or associated symptoms (eg, hypotonia [collapse], syncope, incontinence)
    d. Persistent gastrointestinal symptoms (eg, crampy abdominal pain, vomiting)
4. Reduced BP after exposure to *known allergen for that patient* (minutes to several hours):
    a. Infants and children: low systolic blood pressure (age specific) or greater than 30% decrease is systolic BP
    b. Adults: systolic BP of less than 90 mmHg or greater than 30% decrease from that person's baseline.

Source: Sampson, 2006.

25 PROTOCOL AMENDMENT TEXT REVISIONS

The following table summarizes the revisions made to the protocol and relates the changes to the appropriate rationale (see pages 2-7). Added text is indicated by underlined font and deleted text is indicated by ~~strikethrough~~ font.

Section No./Title	Revision	Rationale
2/Synopsis (Study Rationale)	*The vector genome schematic figure has been updated.* BMN 270 is ~~currently~~ being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. Preliminary results from 270-201 have demonstrated that following gene transfer, sustained FVIII activity above ~~5%~~ ~~(5~~15% (15 IU/dL) up to a year's observation is achievable ~~at doses~~with a dose of 4-6E13 vg/kg with an acceptable safety profile (Pasi, ~~2016). The majority of subjects achieved FVIII levels within the normal range for FVIII, with few to no bleeding episodes and discontinuation of FVIII prophylaxis.~~2017).	26, 28
2/Synopsis (Study Design and Plan)	This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to the AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects ~~will~~may be enrolled at 2-3 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb titer ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb titer > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date. The Data Review Board (DRB) will consist of the ~~2~~ Principal Investigators and Sponsor's Medical Monitor, as well as hemophilia expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201. The DRB will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 2 represents one dosing schedule scenario): ...	1, 5, 6, 10, 28

Section No./Title	Revision	Rationale
	Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed ~~and~~. Additional blood samples will be collected (tryptase, C3, C3a, C4, C5, ~~and C5a~~C5a, and cytokine bead array, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine bead array (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction~~)~~. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor. Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DRB based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL. ~~Throughout the study, subjects with FVIII activity below 5 IU/dL may be monitored more frequently at the discretion of the Investigator and the Medical Monitor.~~ In subjects who ~~show an initial response to BMN 270 but who later have FVIII activity decline to < 5 IU/dL~~experience recurring bleeding episodes, the Investigator and Medical Monitor will ~~review the subject's FVIII activity levels and~~ discuss whether to resume prior FVIII prophylaxis. ~~In addition, the investigator will notify the subject of his FVIII activity levels and will discuss with the subject the risk of bleeding and when (and if) prior FVIII prophylaxis will be resumed.~~ Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after either Week 26 or Week 52, as applicable and at the discretion of the Investigator.	
2/Synopsis (Inclusion and Exclusion Criteria)	Individuals eligible to participate in this study must meet all of the following criteria: 6. No previous documented history of a detectable FVIII inhibitor, and results from a Bethesda assay or Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) (or less than 1.0 BU for laboratories with a historical lower sensitivity cutoff for inhibitor detection of 1.0 BU) on 2 consecutive occasions ~~(~~at least one week apart within the	2, 4, 9, 13, 25, 28

Section No./Title	Revision	Rationale
	~~most recent~~past 12 months (at least one of which should be tested at the central laboratory) ~~at least one week apart within the past 12 months~~ ~~7. HIV positive patients may be enrolled, only if the patient has a CD4 count > 200/mm3 and an undetectable viral load.~~ 8. Sexually active participants must agree to use an acceptable method of effective contraception, either double-barrier contraception ~~for at least 6 months~~(ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion~~, which may include hormonal contraception for a female partner. After 6 months~~; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with no detectable viral vector DNA. Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study: 1. Any evidence of active infection or any immunosuppressive disorder, ~~except for~~including HIV infection~~ as described in the inclusion criterion above~~. 2. Significant liver dysfunction with any of the following abnormal laboratory results: o ALT (alanine ~~transaminase) or AST >2X~~aminotransferase) > 1.25x ULN; o AST (aspartate aminotransferase) > 1.25x ULN; o GGT (gamma-glutamyltransferase) > 1.25x ULN; o Total bilirubin >~~2X~~ 1.25x ULN; o Alkaline phosphatase >~~2X~~ 1.25x ULN; or o INR (international normalized ratio) ≥ 1.4 Subjects whose liver laboratory assessments fall outside of these ranges may undergo repeat testing of the entire liver test panel within the same Screening window and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor. ~~In addition, subjects with abnormal laboratory results related to confirmed benign liver conditions (eg, Gilbert's syndrome) are considered eligible for the study notwithstanding their abnormal laboratory results and may be enrolled after discussion with the Medical Monitor.~~ 8. Chronic or active hepatitis B as evidenced by positive serology testing (hepatitis B surface antigen [HBsAg], hepatitis B surface antibody [HBsAb], and hepatitis B core antibody [HBcAb]) and confirmatory HBV DNA testing.	

Section No./Title	Revision	Rationale
	Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results ~~in the Laboratory Manual~~. 15. Treatment with any Investigational Product within 30 days or 5 half-lives of the investigational product (whichever is longer) prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study.	
7.3/Study Rationale	*The vector genome schematic figure has been updated.* BMN 270 is~~ currently~~ being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. Preliminary results from 270-201 have demonstrated that following gene transfer, sustained FVIII activity above ~~5% (5~~15% (15 IU/dL) up to a year's observation is achievable ~~at doses~~with a dose of 4-6E13 vg/kg with an acceptable safety profile (Pasi, ~~2016). The majority~~2017). For additional information on preliminary data in 270-201, refer to the current version of ~~subjects achieved FVIII levels within the normal range for FVIII, with few to no bleeding episodes and discontinuation of FVIII prophylaxis~~the Investigator's Brochure.	26, 28
7.4/Summary of Overall Risks and Benefits	The majority of subjects in the ongoing 270-201 clinical study who have received 4E13 or 6E13 vg/kg doses of BMN 270 have had Grade 1 asymptomatic elevations in ALT. For most subjects, the elevations have reached only slightly above the upper limit of normal (ULN). Based on the effectiveness of transient oral corticosteroid ~~use~~used to suppress a presumed Class 1 (cytotoxic T-cell) response~~ to hepatocytes transduced~~ in prior studies with hepatic transduction with AAV vectors (Mingozzi, 2013), subjects were treated with 7-32 weeks of oral corticosteroids prophylactically or in response to the elevations in ALT to ensure preservation of the transduced hepatocytes. Using this approach, no sustained loss of FVIII activity has been observed in subjects with ALT elevations, consistent with maintaining a high level of hepatocyte function. Moreover, the rise in ALT levels were not accompanied by significant or lasting aberrations in other liver ~~function tests such as AST, bilirubin or albumin, indicating that extent of toxicity is limited. Overall, the literature~~tests such as AST, bilirubin or albumin, indicating that extent of toxicity is limited. There has been one HIV-positive subject in the ongoing 270-302 clinical study who experienced Grade 3 asymptomatic elevations in ALT and AST, which has been attributed to an interaction between one or more of his antiretroviral therapy medications	1, 4, 28

Section No./Title	Revision	Rationale
	and/or unsuspected underlying hepatic disease with BMN 270. In addition, there has been one subject with Gilbert's syndrome in the ongoing 270-301 clinical study who has experienced Grade 3 asymptomatic elevations in ALT and AST. These cases have led to the exclusion of subsequent HIV-positive subjects and requirement of liver tests at Screening that are <1.25 times the upper limit of the normal range in the ongoing 270-301 and 270-302 clinical studies. Of note, two HIV-positive subjects in 270-301 and one presumed Gilbert's syndrome subject in 270-201 have received BMN 270 without experiencing any elevations in ALT to date. Overall, the literature and clinical experience with BMN 270 suggests that transient elevations in liver enzymes are expected following AAV-based gene therapy for the treatment for hemophilia A or B without any long-term concerns of hepatic injury (Manno, 2006; Nathwani, 2011; George, 2016; Miesbach, 2016; Pasi, 2017). ... As with any infused biological product, there is a potential risk of acute, systemic hypersensitivity reactions (including anaphylaxis) with BMN 270. No hypersensitivity reactions were observed during dosing of BMN 270 in the 270-201 clinical study, although one SAE of pyrexia was reported approximately 16 hours after the infusion in a subject in the 4E13 vg/kg cohort. The subject was treated with acetaminophen, and the fever resolved within 48 hours (see Investigator's Brochure for full details). Infusion-related reactions, including allergic reaction, maculopapular rash, and presyncope, have been reported from ongoing, actively dosing clinical studies of BMN 270, including this study. All of the infusion-related reactions were effectively managed clinically and resolved without any clinical sequelae. Refer to the Investigator's Brochure for additional details.	
9.1/Overall Study Design and Plan	This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects ~~will~~may be enrolled at 2-3 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date. The Data Review Board (DRB) will consist of the ~~2~~ Principal Investigators and Sponsor's Medical Monitor, as well as hemophilia expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201. The DRB will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 9.1.1 represents one dosing schedule scenario):	1, 5, 6, 10, 28

Section No./Title	Revision	Rationale
	... Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed ~~and~~. Additional blood samples will be collected (tryptase, C3, C3a, C4, C5, ~~and C5a~~C5a, and cytokine bead array, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine bead array (and possible additional exploratory testing) will be collected between 8--24 hours after the reaction~~)~~. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor. Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DRB based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL. ~~Throughout the study, subjects with FVIII activity below 5 IU/dL may be monitored more frequently at the discretion of the Investigator and the Medical Monitor. In subjects who show an initial response to BMN 270 but who later have FVIII activity decline to < 5 IU/dL, the investigator and medical monitor will review the subject's FVIII activity levels and discuss whether to resume prior FVIII prophylaxis. In addition, the investigator will notify the subject of his FVIII activity levels and will discuss with the subject the risk of bleeding and when (and if) prior FVIII prophylaxis will be resumed.~~ As the relationship between activity assay results of the BMN 270 gene product and bleeding remains to be established, Investigators should strive to minimize bias by avoiding consideration of FVIII activity levels by themselves or subjects in the reporting of bleeding episodes and FVIII usage.	

Section No./Title	Revision	Rationale
	In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis. Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after either Week 26 or Week 52, as applicable and at the discretion of the Investigator.	
Table 9.1.1-9.1.5 (Schedules of Activities)	*Tables 9.1.1, 9.1.2, and 9.1.3 have been updated (and their footnotes updated) consistent with the changes elsewhere in the protocol. Tables 9.1.4 and 9.1.5 (and their footnotes) have been added as part of this amendment.*	1, 5, 6, 7, 8, 11, 12, 13, 15, 17, 18, 19, 20, 22, 23, 24, 25, 28
Table 9.1.1 (Footnotes)	[b] Includes baseline FVIII activity (chromogenic substrate FVIII assay and one-stage ~~clot~~clotting FVIII assay), coagulation exploratory assay, hFVIII inhibitor level (Bethesda assay with Nijmegen modification), hFVIII total antibody titer, and hFVIII antigen assay. Baseline activity should be assessed at trough (at least >72 hours after last dose of replacement FVIII therapy, or 5x the known half-life of the FVIII ~~replacement therapy~~concentrates administered). [d] Subjects with documented negative results within the last 30 days do not need to be retested. Hepatitis B screening should include hepatitis B surface antigen (HBsAg), hepatitis B surface antibody (HBsAb), and hepatitis B core antibody (HBcAb). [e] Detailed in the Section of laboratory assessments and liver ~~function~~ tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. ABO blood typing assessment should be performed as part of the hematology assessment (at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study). [f] Includes HLA genotyping and FVIII genotyping~~; may include TNF□ and IL10a single nucleotide polymorphisms~~ [g] Blood samples will be collected from subjects to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A ~~disease. The exploratory genetic/genomic testing on these samples is optional~~, coagulation, and/or AAV gene transfer, and to develop assays used for those evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above,	1, 12, 14, 15, 17, 25, 28

Section No./Title	Revision	Rationale
	testing of these samples (including those for TGA assay, Direct Thrombin Activity test, and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor. [i] Smart rescreening should only be performed if a subject has been determined to be eligible for the study and is unable to complete the Baseline assessments and Infusion prior to the closing of the original Screening window (28 + 14 days). Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once. ~~Any screening assessment has not been done at the Screening, it must be done during Smart Rescreening.~~ [k] With the exception of the collection of samples for PCR vector DNA analysis, assessments on the day of infusion must be performed prior to the infusion. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit. On the day of the BMN 270 Infusion, vital signs will be monitored prior to the infusion, during the infusion every 15 minutes (± 5 minutes), and following the infusion hourly (± 5 minutes) for a total time of 24 hours during the subject's stay in the clinic. Shedding samples for PCR of vector DNA analysis (blood, saliva, urine, semen, stool) should be collected between 2 and 24 hours after the infusion has been completed. [m] In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination, vital signs will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, ~~and C5a) and one sample for IgE between 8-24 hours after the reaction, if possible.~~C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.	
Table 9.1.2 (Footnotes)	[*] Visit windows are ± 24 hours during Week 1 and ± 48 hours starting with the Week 2 visit. [b] Detailed in the Section of laboratory assessments and liver ~~function~~ tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. ~~LFTs~~LTs may be monitored more or less frequently (and in particular when ALT values are ~~>~~ ≥ 1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but ~~LFTs~~LTs will be	11, 17, 20, 23, 28

Section No./Title	Revision	Rationale
	monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ~~>~~ ≥ 1.5x ULN or > ULN & > 2x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and ~~LFTs~~LTs during any study week may be performed if: (1) the ALT has increased to above ULN; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ~~liver enzymes~~ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. [c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage ~~clot~~clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen assay. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism. [d] Collection for each matrix to occur until at least 3 consecutive negative results are obtained. Collection and testing of semen samples must continue through Week 12, even if 3 consecutive negative results in that compartment have already been recorded. Separate collection on Day 2 is not required if vector shedding samples were collected on Day 1 post-BMN 270 infusion; if collection on Day 2 is needed, it must occur within 24 hours of the time of the BMN 270 infusion. [e] Blood samples will be collected ~~from subjects~~ to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A ~~disease.~~, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing ~~on these samples is optional~~of these samples (including those for TGA assay, Direct Thrombin Activity test, and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.	

Section No./Title	Revision	Rationale
	[f] Testing for reactivation of hepatitis B and hepatitis C at Week 16, for subjects with a past medical history of hepatitis B or hepatitis C prior to study entry, should be performed only in subjects ~~with evidence of prior exposure and~~ who have not received therapeutic oral corticosteroids prior to Week 16; subjects who have received therapeutic oral corticosteroids will have hepatitis B and hepatitis C testing at the time points indicated in Table 9.1.6.	
Table 9.1.3 (Footnotes)	[*] Visit windows are ± 48 hours ~~through Week 26, and then ± 2 weeks through the end of Year 5~~. [a] Brief physical examination should be done at all weekly visits except Week 26, where a complete physical examination should be performed. Weight should be recorded at ~~every 4 weeks and Week 26. After Week 26, complete physical examination should be done at Week 52 and then yearly, brief physical examination at other visits~~Week 20 and Week 26. [b] Detailed in the Section of laboratory assessments and liver ~~function~~ tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. ~~LFTs~~LTs may be monitored more or less frequently (and in particular when ALT values are ~~>~~≥ 1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but ~~LFTs~~LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ~~>~~≥ 1.5x ULN or > ULN & > 2x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and ~~LFTs~~LTs during any study week may be performed if: (1) the ALT has increased to above ULN; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ~~liver enzymes~~ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. [c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage ~~elot~~clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be	5, 6, 11, 28

Section No./Title	Revision	Rationale
	considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism. [d] Collection for each matrix to occur until at least 3 consecutive negative results are obtained. [f] Subjects who meet the definition of treatment failure to BMN 270 therapy after Week 26 (refer to Section 12.5.3) may omit Week 27-32 visits. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in all fluids must still provide samples at Week 32 (but do not need to do other scheduled assessments on that date).	
Table 9.1.6 (Footnotes)	[d] Should only be performed in subjects with a history of hepatitis B or hepatitis C prior to study entry.	20
9.2/Discussion of Study Design	Approximately 10 subjects ~~will~~ may be enrolled at the 6E13 vg/kg BMN 270 dose. Subjects will be followed for 26 weeks post-BMN 270 infusion during which safety and efficacy assessments will be taken. After the final analysis at 26 weeks post-infusion, safety and efficacy will then continue to be assessed long-term for approximately a total of 5 years.	28
9.3.1/Inclusion Criteria	Individuals eligible to participate in this study must meet all of the following criteria: 6. No previous documented history of a detectable FVIII inhibitor, and results from a Bethesda assay or Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) (or less than 1.0 BU for laboratories with a historical lower sensitivity cutoff for inhibitor detection of 1.0 BU) on 2 consecutive occasions (at least one week apart within the ~~most recent~~past 12 months (at least one of which should be tested at the central laboratory) ~~at least one week apart within the past 12 months~~ ~~7. HIV positive patients may be enrolled, only if the patient has a CD4 count > 200/mm3 and an undetectable viral load.~~ 8. Sexually active participants must agree to use an acceptable method of effective contraception, either double-barrier contraception ~~for at least 6 months~~(ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion~~, which may include hormonal contraception for a female partner. After 6 months~~; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with no detectable viral vector DNA.	2, 9, 13, 28

Section No./Title	Revision	Rationale
9.3.2/Exclusion Criteria	Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study: 1. Any evidence of active infection or any immunosuppressive disorder, ~~except for~~including HIV infection~~ as described in the inclusion criterion above~~. 2. Significant liver dysfunction with any of the following abnormal laboratory results: o ALT (alanine ~~transaminase) or AST >2X~~aminotransferase) > 1.25x ULN; o AST (aspartate aminotransferase) > 1.25x ULN; o GGT (gamma-glutamyltransferase) > 1.25x ULN; o Total bilirubin >~~2X~~ 1.25x ULN; o Alkaline phosphatase >~~2X~~ 1.25x ULN; or o INR (international normalized ratio) ≥ 1.4 Subjects whose liver laboratory assessments fall outside of these ranges may undergo repeat testing of the entire liver test panel within the same Screening window and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor. ~~In addition, subjects with abnormal laboratory results related to confirmed benign liver conditions (eg, Gilbert's syndrome) are considered eligible for the study notwithstanding their abnormal laboratory results and may be enrolled after discussion with the Medical Monitor.~~ 8. Chronic or active hepatitis B as evidenced by positive serology testing (hepatitis B surface antigen [HBsAg], hepatitis B surface antibody [HBsAb], and hepatitis B core antibody [HBcAb]) and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results ~~in the Laboratory Manual~~. 15. Treatment with any Investigational Product within 30 days or 5 half-lives of the investigational product (whichever is longer) prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study.	2, 4, 25, 28
9.4.2.1/Product Characteristics and Labeling	~~The study drug label includes the following information: lot number, required storage conditions, a precautionary statement, expiry date, study number, and BioMarin name and address. Labelling will be done according to country specific requirements.~~	28

Section No./Title	Revision	Rationale
	The study drug is labelled according to the particulars approved by the relevant regulatory agencies.	
9.4.4/Directions for Administration	~~After~~On the day of infusion, the subject will come to the infusion site, where a physical examination will be performed by the Investigator or designee~~, subjects will be admitted on the day of BMN 270 infusion~~. If the subject is found to have an active acute illness at the time of planned infusion, then the infusion should be deferred until the illness has resolved; screening procedures may require repetition if outside the specified window. An IV catheter or butterfly needle will be inserted into a suitable peripheral vein (eg, the median cubital vein) and flushed with saline. FVIII replacement therapy will not be given since venipuncture is a minimally invasive procedure in these individuals under ordinary conditions. BMN 270 will be prepared and infused as a pure solution over a dose-dependent time. Prepared drug will be kept at room temperature prior to administration. An electric syringe pump will be used to infuse through an in-line, low protein binding 0.22 micron filter. BMN 270 will be infused through the catheter using an appropriate infusion pump at ~~a constant~~an initial rate of 1 mL/min. The infusion rate should be increased every 30 minutes by 1 mL/min up to a maximum of 4 mL/min ~~while monitoring the~~, provided that the subject's clinical condition permits such an increase. Of note, the IP has been shown to be stable at room temperature for 7.5 hours following completion of product thaw. Vital signs (pulse, blood pressure, respiration rate and temperature) should be monitored at 15 minute (±5 minutes) intervals~~. The anticipated maximum interval from initiation of thawing of BMN 270 to completion of the infusion is 4 hours, although the IP has been shown to be stable at room temperature for 6 hours.~~ throughout the period of the infusion. As with any infused biological product, there is a potential risk of acute, systemic hypersensitivity reactions (including anaphylaxis) with BMN 270. Dosing will be administered at a qualified infusion site, with appropriate resuscitation equipment and medication available and easily accessible. Clinical staff administering BMN 270 should be trained appropriately in recognizing and managing the signs and symptoms associated with potential hypersensitivity, anaphylactic, and anaphylactoid reactions. Additionally, the Investigator should be familiar with Sampson's criteria for defining anaphylaxis (Sampson, 2006; Appendix 1). Should symptoms of potential hypersensitivity occur, the infusion may be slowed or halted at the Investigator's discretion, with consideration of the subject's clinical condition. If the infusion is halted, it should only be restarted if the Investigator considers it safe and appropriate to do so. Antihistamines, anti-pyretic, and/or corticosteroid administration is permitted prior to restarting an interrupted infusion by an infusion-related reaction. At the restart, the infusion rate may be adjusted (ie, to a slower rate [minimum of 1 mL/min], with the rate increased every 30 minutes by	1, 28

Section No./Title	Revision	Rationale
	1 mL/min up to a maximum rate of 4 mL/min, if the subject's clinical condition permits such an increase) with careful monitoring of the subject. In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor. ... Prior to discharging subjects from the clinic, the Investigator or designee should instruct subjects how to recognize signs and symptoms of potential (delayed) hypersensitivity reactions and anaphylaxis, and to contact a medical practitioner or seek emergency care in case of such an event.	
9.4.5/Method of Assigning Subjects to Treatment Groups	Subjects who meet all eligibility criteria (refer to Section 9.3.1 and Section 9.3.2) may be enrolled into the study. Approval by the Medical Monitor will be required prior to enrollment of each study subject. Upon their enrollment into the study, subjects will be assigned a unique subject number ~~by the Sponsor~~.	28
9.4.8/Prior and Concomitant Medications	The following medications are prohibited starting 30 days before Screening and through the end of the study, and the Sponsor must be notified if a subject receives any of these during the study: • Any investigational therapy • Systemic immunosuppressive agents, except for corticosteroids • Emicizumab • Fitusiran • Concizumab • Efavirenz	2, 3, 28

Section No./Title	Revision	Rationale
	• Lamivudine Subjects who begin antiretroviral therapy following BMN 270 infusion and during their participation in 270-203 must have their antiretroviral medication regimen approved by the Investigator and Medical Monitor prior to starting antiretroviral treatment.	
9.4.8.1/Concomitant Hemophilia Treatments	Subjects on prophylactic FVIII therapy will discontinue their regular treatment regimen starting 4 weeks ~~following~~after the day of infusion ~~or after FVIII activity has reached at least 5 IU/dL (whichever is earlier)~~ and switch to an "on-demand" schedule.	10
9.4.8.2/Therapeutic Glucocorticoid Treatment for Elevated Hepatic Transaminases	Therapeutic oral corticosteroids (prednisone or converted equivalent) should be initiated when either of the following occurs post-BMN 270 infusion in any subject and after consultation with the Medical Monitor ~~(or their designee if consultation is required outside of the Medical Monitor's waking hours)~~: • ALT ≥ 1.5x ULN or ALT > ULN & > 2x baseline value in 2 consecutive assessments within 72 hours and alternative etiologies have been ruled out, or ALT ≥ 3x ULN in 2 consecutive assessments within 48 hours (refer to Table 9.7.6.3.2) ~~In addition, if FVIII activity drops > 50% at any time post-BMN 270 infusion, a course of therapeutic oral corticosteroids should be considered upon consultation between the Investigator and the Medical Monitor.~~ ... Following initiation or completion of therapeutic oral corticosteroids, if ALT elevation ≥1.5x ULN or ALT ≥ ULN & ≥ 2x baseline value is reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor. Modification of the corticosteroid regimen may take into consideration possible confounders for the ALT elevation and impact on FVIII expression. Management and monitoring of reactions to corticosteroids should be determined by the Investigator's clinical judgment in consultation with the Sponsor's Medical Monitor. This includes the contraindicated use of NSAIDs during corticosteroid treatment and specific monitoring not already covered by the SoA. The use of COX-2 inhibitors, while not contraindicated during corticosteroid treatment, should be limited, if possible. Practical management to prevent complications related to oral corticosteroid therapy may be undertaken at the discretion of the Investigator (eg, evaluation of glucose intolerance, hyperlipidemia etc.). Hepatitis B status and HCV viral load will be rechecked 6 weeks after the start of oral corticosteroid treatment and then 1 week and 13 weeks after the completion of oral	10, 11, 20, 28

Section No./Title	Revision	Rationale
	corticosteroid treatment~~.~~ in subjects with a history of hepatitis B or hepatitis C. All adverse events (including any adverse events suspected to be caused by or related to corticosteroid use) should be reported as outlined in Section 10 of the protocol.	
Table 9.4.8.2.1/ Adjustments to Corticosteroid Regimen	Table 9.4.8.2.1 has been updated to reflect changes in the text of this section.	11, 28
~~Section 9.4.9/ Monitoring of HIV-Positive Subjects~~	~~HIV-positive subjects may be enrolled in 270-203 if the subject has a CD4 count > 200/mm3 and an undetectable viral load.~~ ~~Subjects should continue anti-retroviral therapy (ART) as prescribed and follow routine monitoring of CD4 count and viral load (Angus, 2016). No alterations in the monitoring are indicated for enrolled immunocompetent HIV-positive subjects who receive corticosteroids as part of their enrollment in 270-203.~~	2
9.4.10/Treatment Compliance	Study drug will be administered to subjects at the dosing facility by a qualified health care professional. The quantity dispensed, returned, used, lost, etc. must be recorded on ~~the~~a dispensing log ~~provided for the study~~. Sites will be instructed to return or destroy all used and unused study drug containers.	28
9.5/IP Accountability	The Investigator or designee is responsible for maintaining accurate records (including dates and quantities) of IP(s) received and IP lost or accidentally or deliberately destroyed. The Investigator or designee must retain all unused or expired study supplies until the study monitor (on-site CRA) has confirmed the accountability data, if allowed by local SOPs.	28
9.5.1/Return and Disposition of Clinical Supplies	Unused study drug must be kept in a secure location for accountability and reconciliation by the study monitor. The Investigator or designee must provide an explanation for any destroyed or missing study drug or study materials~~.~~ (or must be referenced in their institution SOPs).	28
9.6/Dietary or Other Restrictions	Subjects should be advised to abstain from any blood or sperm donation after BMN 270 infusion, until there is no further evidence of vector shedding from PCR analysis of samples.	21
9.7.2.1/FVIII Activity	Values for FVIII activity will be excluded from analysis if obtained within 72 hours since the last infusion of exogenous FVIII protein concentrates ~~(or within 5x the known half-life of the FVIII concentrates administered).~~ If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements	6, 10, 28

Section No./Title	Revision	Rationale
	when a 72-hour interval without FVIII use is achieved; The 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. ~~In subjects who show an initial response to BMN 270 but who later have FVIII activity decline to < 5 IU/dL, the investigator and medical monitor will review the subject's FVIII activity levels and discuss whether to resume prior FVIII prophylaxis. In addition, the investigator will notify the subject of his FVIII activity levels and will discuss with the subject the risk of bleeding and when (and if) prior FVIII prophylaxis will be resumed.~~ ... Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after either Week 26 or Week 52, as applicable and at the discretion of the Investigator.	
9.7.2.2/FVIII Replacement Therapy/Bleeding Episodes	Additional efficacy variables are: • Change of annualized utilization (IU/kg) of exogenous FVIII replacement therapy during Week 5 to Week 26 post BMN 270 infusion from the baseline utilization of exogenous FVIII replacement therapy~~ calculated using subjects' historical medical records during the year prior to the enrollment.~~ • Change of ABRs requiring exogenous FVIII replacement treatment during Week 5 to Week 26 of the study post BMN 270 infusion from the baseline ABR~~ calculated using subjects' historical medical records during the year prior to the enrollment.~~ ... In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis.	10, 28
9.7.5/Exploratory Assessments	In addition, blood samples will be collected from subjects at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5 to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV5 gene transfer, and to develop assays used for these	28

Section No./Title	Revision	Rationale
	evaluations. Subject may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A ~~is optional~~.	
9.7.6.2/Clinical Laboratory Assessments	The scheduled clinical laboratory tests are listed in Table 9.7.6.2.1. Refer to the Study Reference Manual for instructions on obtaining and shipping samples. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample. ... In addition to scheduled clinical laboratory assessments, a fasting blood lipid panel (including triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) will be assessed at the BMN 270 infusion visit. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit. In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.	1, 14, 15, 28
Table 9.7.6.2.1/ Clinical Lab Tests	Table 9.7.6.2.1 has been updated to reflect changes in the text of the protocol amendment.	12
9.7.6.3/Liver and Hepatitis Testing	Subjects will be screened for evidence of previous or active hepatitis B or hepatitis C infection at Screening~~.~~; hepatitis B screening should include HBsAg, HBsAb, and HBcAb. Subjects with documented results showing an absence of active hepatitis B or hepatitis C infection (as measured by positive surface antigen for hepatitis B or positive RNA testing for hepatitis C) 30 days prior to providing signed informed consent do not need to repeat those tests during the screening period.	20, 25, 28

Section No./Title	Revision	Rationale
	Evidence of ongoing hepatitis B or hepatitis C infection is exclusionary. ~~Subjects who have cleared a Hepatitis B infection or are seronegative do not need to receive the~~ Subjects with history of hepatitis B ~~vaccination. Subjects with evidence of~~ or hepatitis C infection prior ~~exposure~~to study entry will be tested for hepatitis B and hepatitis C reactivation at Week 16. Subjects with a history of hepatitis B or hepatitis C will be asked for information about the treatments received as part of their medical history assessment at Screening. Subjects with a previous history of hepatitis B or hepatitis C who receive therapeutic oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.46.	
Section 9.7.6.3 (Tables)	Table 9.7.6.3.1 (Liver Tests), Table 9.7.6.3.2 (Evaluation of ALT Elevations), and Table 9.7.6.3.3 (Viral and Autoimmune Hepatitis Workup Testing) have been updated to reflect changes made as part of this amendment.	4, 28
Section 9.7.6.5/Vital Signs, Physical Exams and Other Safety Observations	A complete physical examination should be performed at Screening, Week 26, Week 52, and ~~then yearly until the end of the study. At other visits, brief physical examinations may be performed at the discretion of the Investigator based on the subject's clinical condition. Particular attention should be given to signs of bleeding, as well as assessing possible hemarthroses.~~ at the End of Year visits. A complete physical examination will include general appearance (head, eyes, ears, nose, and throat), cardiovascular, dermatologic, lymphatic, respiratory, gastrointestinal, genitourinary, musculoskeletal, and neurologic systems. At other visits, brief physical examinations may be performed at the discretion of the Investigator based on the subject's clinical condition. A brief physical examination will include general appearance, cardiovascular, dermatologic, respiratory, gastrointestinal, musculoskeletal, and neurologic assessments. Particular attention should be given to signs of bleeding, as well as assessing possible hemarthroses. Height will be recorded at Screening only. Weight will be recorded at Screening and then every 4 weeks thereafter through Week 20, at Week 26, every 4 weeks through Week 52, and then ~~every 3 months thereafter until~~at the second Q12W visit each year and at every End of ~~the study~~Years visit during Years 2-5.	5, 16, 28
9.7.6.6/Vector Shedding	Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule (refer to Section 12.5.3) but who have not cleared vector shedding from all fluids must still provide samples for assessment until vector shedding has cleared: at Weeks 32, 36, 40, 44, 48, and 52 (during Year 1), every 4 weeks (during Year 2), or every 6 weeks (during Years 3-5).	6, 13, 28

Section No./Title	Revision	Rationale
	Samples may be fractionated prior to shedding analysis in order to better characterize the presence, structure, and location of vector DNA and/or vector capsid within each matrix. If needed, the fractionation ~~will~~may be performed with samples collected specifically for shedding analysis (saliva, blood, semen, urine, ~~stool~~feces). Alternatively, the vector DNA characterization during shedding analysis may utilize already fractionated exploratory samples obtained from the above biofluids, such as exploratory plasma samples, exploratory PBMC samples, and red blood cells recovered during PBMC/plasma isolations. ... Contraception use may need to be extended beyond 26 weeks in individual subjects based on observed vector shedding in semen. After 26 weeks, subjects may stop contraception use only if they have had 3 consecutive negative semen samples~~.~~ (upon consultation between the Investigator and Medical Monitor).	
10.2.1/EOSI	The following EOSI need to be reported to the Sponsor within 24 hours of site awareness, irrespective of seriousness, severity or causality: • Elevation of ALT ~~>~~≥ 1.5x ULN or ALT > ULN & >2x baseline value, regardless of whether that elevation triggers an initiation or modification of oral corticosteroid treatment • ~~Drug-related anaphylactoid or~~ Systemic hypersensitivity, anaphylactic, or anaphylactoid reactions (refer to Appendix 1)	1, 11
10.9/Contact Information	Contact information for the medical monitor is as follows: ~~Name: PI MD~~ ~~Address 105 Digital Drive~~ ~~Novato, CA 94949~~ ~~Phone: PI~~ ~~E-mail: PI~~ Name: PI MA MB BChir MSc Address: Biomarin (UK) Ltd. 10 Bloomsbury Way London WC1A 2SL Phone: PI (office)	27

Section No./Title	Revision	Rationale
	PI (mobile) E-mail: PI	
12.2/Screening	The following procedures will be performed during the Screening Period: • Full medical history, including hemophilia A history, Hepatitis B, Hepatitis C, and HIV. Subjects with a history of hepatitis B or hepatitis C will be asked for information about the treatments received. Any prior pharmacokinetics information obtained while the subject was receiving prophylactic or on-demand FVIII therapy prior to the study should also be collected. • Screen for Hepatitis B, Hepatitis C, and HIV if required (subjects with documented negative results 30 days prior to informed consent being obtained do not need to be retested) ○ Hepatitis B screening should include HBsAg, HBsAb, and HBcAb • ~~Von Willebrand Factor Antigen (VWF:Ag)~~ • Blood samples for Biomarker testing (may include HLA genotyping~~,~~ and FVIII genotyping status~~, TNFα and IL10a single nucleotide polymorphisms~~)	8, 18, 19, 25
12.3/Baseline	The following procedures will be performed during the Baseline Period: • Samples for hFVIII Assays ○ Baseline FVIII activity – chromogenic substrate FVIII assay ○ Baseline FVIII activity level – one-stage ~~elot~~clotting FVIII assay ○ hFVIII coagulation activity exploratory assay ~~(collected but not tested prior to enrollment)~~ ○ hFVIII inhibitors (Bethesda assay with Nijmegen modification) ○ hFVIII total antibody assay ~~(collected but not tested prior to enrollment)~~ ○ hFVIII antigen assay ~~(collected but not tested prior to enrollment)~~ • Von Willebrand Factor Antigen (VWF:Ag) • Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)	12, 19, 28

Section No./Title	Revision	Rationale
	○ ABO blood typing assessment should be performed as part of the hematology assessment (at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study).	
12.4/Treatment Visit/ Day 1	The following procedures will be performed during the BMN 270 Infusion Visit: • Fasting blood sample for future exploratory analysis (sample collected pre-infusion) • Fasting lipid panel (blood triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) (sample collected pre-infusion) ○ Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit. • PCR of vector DNA in blood, saliva, urine, semen, and stools ○ Collection of samples for PCR testing should occur between 2 and 24 hours after the BMN 270 infusion has been completed In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.	1, 7, 15, 28
12.5.1.1/Day 2	On Study Day 2, the following assessments will be performed: • PCR of vector DNA in blood, saliva, urine, semen, and stools ○ Separate collection on Day 2 is not required if vector shedding samples were collected on Day 1 post-BMN 270 infusion; if collection on Day 2 is needed, it must occur within 24 hours of the time of the BMN 270 infusion.	28

Section No./Title	Revision	Rationale
12.5.1.3/Day 8	On Study Day 8, the following assessments will be performed: • Blood sample for AAV5 TAb Assay ○ Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay • Blood sample for AAV5 TI assay	24
12.5.2.3/Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26	The following procedures will be performed at Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26: • PCR of vector DNA in blood, saliva, urine, semen, and stools ○ Collection to occur until at least 3 consecutive negative results are obtained. Semen samples should continue to be collected and tested through Week 12, even if 3 consecutive negative results in that compartment have been recorded prior to that time point.	28
12.5.2.7/Weeks 2, 6, 10, 14, 20, 24, and 26	The following procedures will be performed at Weeks 2, 6, 10, 14, 20, 22, and 26: • Cytokine bead array assay	1
12.5.2.11/Week 16	The following procedure will be performed at Week 16: • Testing for reactivation of hepatitis B and hepatitis C (only in subjects with evidence of prior exposure ~~will be tested for~~to hepatitis B and/or hepatitis C ~~reactivation at Week 16.~~	20
12.5.3/Weeks 27-~~260~~ 52	~~Every 3 months (Weeks 39, 52, 65, 78, 91, 104, 117, 130, 143, 156, 169, 182, 195, 208, 221, 234, 247, and 260)~~ Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 26 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 26 of the study by attending only the Week 36, 44, and 52 visits during the remainder of Year 1. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in all fluids must still provide samples at Week 32, Week 40, and Week 48, if necessary (but do not need to do other scheduled assessments on that date). During Weeks 27-36, subjects will return to the study site weekly (± 48 hours). During Weeks 37-52, subjects will return to the study site every 2 weeks (Week 38, 40, 42, 44, 46, 48, 50, and 52) (± 1 week). At these visits, the following procedures will be completed:	5, 6

Section No./Title	Revision	Rationale
12.5.3.1/Every Visit	At every visit (Weeks 27-36, 38, 40, 42, 44, 46, 48, 50, and 52), the following procedures will be performed ~~every 3 months (Weeks 39, 52, 65, 78, 91, 104, 117, 130, 143, 156, 169, 182, 195, 208, 221, 234, 247, and 260) (± 2 weeks):~~: • Physical examination o Brief physical examination ~~(~~should be performed at all weeks except Week 52, when a complete physical examination should be ~~done at Week 52, 104, 156, 208, and 260)~~performed • ~~Weight~~ • Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use) • ~~Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)~~ • Vital Signs • ~~Blood chemistry, hematology, and coagulation tests (refer to)~~ • ~~Urine Tests (refer to)~~ • Liver ~~Function~~ Tests (refer to Table 9.8.8.3.1) o ~~LFTs~~LTs may be monitored more or less frequently (and in particular when ALT values are ~~>~~ ≥ 1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but ~~LFTs~~LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN~~.~~. • ~~Samples for hFVIII~~ FVIII Assays o ~~Baseline FVIII activity – chromogenic FVIII assay~~ o ~~Baseline~~ FVIII activity level ~~–~~(chromogenic substrate FVIII assay) o FVIII activity level (one-stage ~~clot~~clotting FVIII assay) o ~~hFVIII~~FVIII coagulation activity exploratory assay ~~(collected but not tested prior to enrollment)~~ o ~~hFVIII inhibitors (~~Bethesda assay (with Nijmegen modification) for FVIII inhibitor level o ~~hFVIII total antibody assay (collected but not tested prior to enrollment)~~ o ~~hFVIII antigen assay (collected but not tested prior to enrollment)~~	5, 11, 28

Section No./Title	Revision	Rationale
	~~o hFVIII total antibody assay~~ ~~o Exploratory biomarker assessments~~ ~~o Blood sample for AAV5 Total Antibody assay~~ ~~o Sample will be tested with a AAV5 TAb post dose immunogenicity monitoring assay~~ ~~o Blood sample for AAV5 TI assay~~ ~~o PBMC collection for CTL baseline~~ ~~o VWF:Ag~~ ~~o Direct Thrombin test~~ ~~o Thrombin Generation Assay~~ o ~~Every 12 months (~~FVIII protein assay	
12.5.3.2/Weeks ~~52, 104, 156, 208~~28, 30, 32, 34, 36, 44, and ~~260)~~52	~~The following procedures will be performed every 12 months (~~At Weeks ~~52, 104, 156, 208~~28, 30, 32, 34, 36, 44, and ~~260) (± 2 weeks):~~ ~~Haemo-QoL-A QoL assessment~~ ~~EQ-5D-5L~~ ~~Hemophilia Activities List (HAL)~~ ~~Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire~~	5, 28
12.5.3.3/Weeks 32, 36, 44, and 52	At Weeks 32, 36, 44, and 52, the following procedures will be performed: • Weight • Blood chemistry, hematology, and coagulation tests (refer to Table 9.8.8.2.1) • FVIII antibody titer • AAV5 TAb Assay • AAV5 TI Assay	5

Section No./Title	Revision	Rationale
	• Cytokine bead array assay	
12.5.3.4/Weeks 32, 36, 40, 44, 48, and 52	At Weeks 32, 36, 40, 44, 48, and 52, the following procedures will be performed: • Exploratory biomarker assessments • TGA Assay • PCR of vector DNA in blood, saliva, urine, semen, and ~~stool~~stools o Sample testing ~~during Weeks 27-260 is not required if~~to occur until at least 3 consecutive ~~samples are clear during the Post-Infusion Follow-Up period in Weeks 1-26~~negative sample results have been obtained. Subjects who have not had 3 consecutive negative semen samples by Week ~~26~~52 should continue to have PCR testing of semen every 4 weeks until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor). o Subjects considered to be treatment failures must continue to provide samples for PCR assessment at these timepoints until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints)	5, 6, 17, 28
12.5.3.5/Week 36 and 52	At Weeks 36 and 52, the following procedures will be performed: • Urine Tests (refer to Table 9.8.8.2.1) • Direct Thrombin test • VWF:Ag	5, 17
12.5.3.6/Week 52	At Week 52, the following procedures will be performed: • Haemo-QoL-A assessment • EQ-5D-5L • HAL • WPAI+CIQ:HS	5
12.6/Years 2-5	Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion	5, 6

Section No./Title	Revision	Rationale
	with the Medical Monitor, follow an abbreviated visit schedule after Week 52 of the study by attending only the Q12W and End of Year visits during Years 2-5. Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every 4 weeks (during Year 2) or every 6 weeks (during Years 3-5) until vector shedding has cleared (but do not have to perform other scheduled assessments at those study visits). During Years 2-5 of Post-Infusion Follow-up, the following procedures will be completed:	

12.6.1/Year 2 – Every 4 Weeks	During Year 2, every 4 weeks (± 2 weeks), the following procedures will be performed:
	
	• Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
	• Liver Tests (refer to Table 9.8.8.3.1)
	  o LTs may be monitored more or less frequently (and in particular when ALT values are ≥ 1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
	• FVIII Assays
	  o FVIII activity level (chromogenic substrate FVIII assay)
	  o FVIII activity level (one-stage clotting FVIII assay)
	  o FVIII coagulation activity exploratory assay
	  o Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
	  o FVIII protein assay
	• PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)
	  o Sample testing during Year 2 is not required if at least 3 consecutive samples are negative during the Post-Infusion Follow-Up period in Weeks 1-52. Subjects who have not had 3 consecutive negative semen samples by Week 52 should continue to have PCR testing of semen every 4 weeks during Years 2 until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).
		5

Section No./Title	Revision	Rationale
	○ Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every 4 weeks during Year 2 until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints).	
12.6.2/Years 3-5 – Every 6 Weeks	During Years 3-5, every 6 weeks (± 2 weeks), the following procedures will be performed: • Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use) • Liver Tests (refer to Table 9.8.8.3.1) ○ LTs may be monitored more or less frequently (and in particular when ALT values are ≥ 1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. • FVIII Assays ○ FVIII activity level (chromogenic substrate FVIII assay) ○ FVIII activity level (one-stage clotting FVIII assay) ○ FVIII coagulation activity exploratory assay ○ Bethesda assay (with Nijmegen modification) for FVIII inhibitor level ○ If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 3-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result. ○ FVIII protein assay • PCR of vector DNA in blood, saliva, urine, semen, and stools (if required) ○ Sample testing during Years 3-5 is not required if at least 3 consecutive samples are clear by the end of Year 2. Subjects who have not had 3 consecutive negative semen samples by the end of Year 2 should continue to have PCR testing of semen every 6 weeks during Years 3-5 until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).	5

Section No./Title	Revision	Rationale
	○ Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every 6 weeks during Years 3-5 until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints).	
12.6.3/Years 2-5 – Every 12 Weeks and End of Year Visits	During Years 2-5, subjects will be asked to return to the study site for visits at the following study weeks (± 2 weeks): • Year 2 – Week 64, Week 76, Week 88, Week 104 • Year 3 – Week 116, Week 128, Week 140, Week 156 • Year 4 – Week 168, Week 180, Week 192, Week 208 • Year 5 – Week 220, Week 232, Week 244, Week 260 For each of these years, the last study visit listed (Week 104, Week 156, Week 208, and Week 260) will serve as an End of Year visit. At the every 12 week and End of Year visits, the following procedures will be performed: • Brief physical examination (at every 12 week visits) or complete physical examination (at End of Year visits) • Weight (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only) • Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use) • Liver Tests (refer to Table 9.8.8.3.1) ○ LTs may be monitored more or less frequently (and in particular when ALT values are ≥ 1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. • FVIII Assays ○ FVIII activity level (chromogenic substrate FVIII assay) ○ FVIII activity level (one-stage clotting FVIII assay) ○ FVIII coagulation activity exploratory assay	5

Section No./Title	Revision	Rationale
	○ Bethesda assay (with Nijmegen modification) for FVIII inhibitor level ○ If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 3-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result. ○ FVIII protein assay • Blood chemistry, hematology, and coagulation tests (refer to Table 9.8.8.2.1) (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only) • Urine Tests (refer to Table 9.8.8.2.1) (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only) • Vital Signs • AAV5 TAb Assay • AAV5 TI Assay • FVIII antibody titer • Haemo-QoL-A assessment (at End of Year visits only) • EQ-5D-5L (at End of Year visits only) • HAL (at End of Year visits only) • WPAI+CIQ:HS (at End of Year visits only) • Exploratory biomarker assessments • PBMC collection • VWF:Ag • Direct Thrombin test • TGA Assay • PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)	

Section No./Title	Revision	Rationale
	○ Sample testing during Years 2-5 is not required if at least 3 consecutive samples are negative during the Post-Infusion Follow-Up period in Weeks 1-52. Subjects who have not had 3 consecutive negative semen samples by Week 52 should continue to have PCR testing of semen every 4 weeks (during Year 2) or every 6 weeks (during Years 3-5) until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).	
15/DRB	The Data Review Board (DRB) will consist of the ~~2~~ Principal Investigators and Sponsor's Medical Monitor, as well as hemophilia expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201.	28
Appendix 1/ Sampson's Criteria	According to the National Institute of Allergy and Infectious Disease/Food Allergy and Anaphylaxis Network (NIAID/FAAN) Second Symposium on the definition and management of anaphylaxis, anaphylaxis is highly likely when any one of the following 3 criteria are fulfilled: 1. Acute onset of an illness (minutes to several hours) with involvement of the skin, mucosal tissue, or both (eg, generalized hives, pruritus or flushing, swollen lips-tongue-uvula) *AND AT LEAST ONE OF THE FOLLOWING* a. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced peak expiratory flow [PEF], hypoxemia) b. Reduced blood pressure (BP) or associated symptoms of end-organ dysfunction (eg, hypotonia [collapse], syncope, incontinence) 2. Two or more of the following that occur rapidly after exposure *to a likely allergen for that patient* (minutes to several hours): a. Involvement of the skin-mucosal tissue (eg, generalized hives, itch-flush, swollen lips-tongue-uvula) b. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced PEF, hypoxemia) c. Reduced BP or associated symptoms (eg, hypotonia [collapse], syncope, incontinence) d. Persistent gastrointestinal symptoms (eg, crampy abdominal pain, vomiting) 3. Reduced BP after exposure to *known allergen for that patient* (minutes to several hours):	1

Section No./Title	Revision	Rationale
	a. Infants and children: low systolic blood pressure (age specific) or greater than 30% decrease is systolic BP b. Adults: systolic BP of less than 90 mmHg or greater than 30% decrease from that person's baseline. Source: Sampson, 2006.	

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CLINICAL STUDY PROTOCOL

Study Title:	A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5
Protocol Number:	270-203
Active Investigational Product:	AAV5-hFVIII-SQ
European Union Drug Regulating Authorities Clinical Trials (EudraCT) Number:	2017-000662-29
Indication:	Hemophilia A
Sponsor:	BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949
Development Phase:	Phase 1/2
Sponsor's Responsible Medical Monitor:	PI MD PhD PI , Clinical Sciences BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949
Study Design:	Single-arm, open-label
Duration of Subject Participation:	Up to 264 weeks
Dose:	6E13 vg/kg as single infusion
Study Population:	Males ≥ 18 years of age with severe hemophilia A and detectable pre-existing antibodies against AAV5 vector capsid
Date of Original Protocol:	29 September 2017
Date of Amendment 1:	5 October 2018
Date of Amendment 2:	4 October 2019

CLINICAL STUDY PROTOCOL AMENDMENT SUMMARY

Amendment 2

Date: 4 October 2019

RATIONALE AND SUMMARY OF CHANGES

A summary of major changes covered by Amendment 2 to the 270-203 protocol is provided below:

1. Prophylactic corticosteroids starting on Day 1 (day of BMN 270 infusion) have been added for all subjects.

Rationale: Preliminary data from the 270-201 and 270-301 studies suggest that earlier initiation of corticosteroid treatment decreases the number and severity of ALT elevations and concomitant decreases in FVIII activity following BMN 270 infusion. Prophylactic corticosteroid treatment, in a regimen similar to what was used in 270-201, is therefore being instituted in this protocol in an effort to increase subject safety in patients with detectable pre-existing immunity to the vector capsid and maximize treatment response.

2. The threshold for considering the additional use of therapeutic oral corticosteroids has been lowered.

Rationale: The previous threshold was ALT ≥ 1.5x ULN or ALT > ULN & > 2x baseline; the new threshold is ALT ≥ ULN or ≥ 2x baseline. This change will encourage earlier treatment with corticosteroids, which could help decrease the extent of increases in ALT and better preserve FVIII expression.

3. Subjects considered to be treatment failures (ie, those who either fail to achieve FVIII activity > 5 IU/dL by the chromogenic substrate assay or are unable to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes) may now start following an abbreviated visit schedule only after Week 52, not after Week 26.

Rationale: Emerging data demonstrate that ALT elevations or increases in FVIII activity may occur between Weeks 26 and 52; therefore, it is important to maintain the standard visit schedule through Week 52.

4. Vector shedding and contraception use language has been updated to change the determination of a “clear” result from negative to below the limit of detection.

Rationale: The change in language better reflects regulatory guidance documents.

5. The Direct Thrombin Assay has been removed.

Rationale: Based on the lack of correlation observed between FVIII activity levels and the Direct Thrombin Assay results in Study 270-201, the test is no longer being collected across the BMN 270 clinical development program.

6. The option to assess an adverse event as related/not related to corticosteroids has been added.

Rationale: With all subjects taking prophylactic corticosteroids, AEs associated with corticosteroids are possible and should be noted as such on the eCRF (and for safety monitoring and risk:benefit assessment purposes).

7. A two-step screening process has been introduced.

Rationale: During step 1 of the screening process, potential subjects may be tested for AAV5 TAb titers using a screening assay developed for that purpose. Given that AAV5 TAb positivity is required for this study, completing this step first allows subjects who are not AAV5 TAb positive to avoid the unnecessary burden of working through additional screening steps before finding out they are not eligible to participate in the study.

8. Language has been added to permit use of mobile nursing (MN) services, provided that the site is able to implement them and the subject consents, for certain designated study visits.

Rationale: Allowing for MN services at designated study visits will help alleviate subject travel burden.

9. The period of recommended abstinence from alcohol after BMN 270 infusion was increased from 26 weeks to 52 weeks, to support subject liver health and subject safety.
10. The period in the exclusion criteria during which subjects should not have planned major surgeries following BMN 270 infusion was increased from 26 to 52 weeks, for purposes of aligning with other BMN 270 studies and for the protection of patient safety.
11. Product characteristics and labeling have been updated to reflect the current manufacturing process.
12. The identity of the Medical Monitor has been updated.
13. The Data Review Board (DRB) has been changed to a independent Data Monitoring Committee (DMC).
14. The vector genome schematic has been updated.
15. Risk-benefit language has been updated to reflect more current clinical results.

16. Inconsistency in the Schedule of Assessments concerning collection of AAV5 TI and TAb assay has been clarified.
17. The proposed number of study sites has been increased.
18. To reduce subject burden and align with other studies in the BMN 270 clinical development program, the every 4 week visits during Year 2 post-infusion have been changed to every 6 week visits.
19. Changes have been made to correct minor errors and for purposes of clarity and consistency.

Refer to Section 25 for a summary of revisions to the Amendment 1 (dated 5 October 2018).

2 SYNOPSIS

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

TITLE OF STUDY:

A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5

PROTOCOL NUMBER:

270-203

STUDY SITES:

Approximately 5-6 sites globally

PHASE OF DEVELOPMENT:

Phase 1/2

STUDY RATIONALE:

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. The clinical phenotype of HA patients generally correlates tightly with the level of residual expression. Severe HA is classified as FVIII activity less than 1% of wild-type (< 1 IU/dL), moderate disease comprises 1-5% of wild-type activity, and the mild form is 5-40% activity. The clinical manifestations of severe HA are frequent spontaneous bleeding episodes, predominantly in joints and soft tissues, with a substantially increased risk of death from hemorrhage when the brain is involved.

Treatment of severe HA presently mainly consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study (Berntorp, 2017) and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with on-demand-only therapy in the same studies, respectively. Continued repeated bleeding events compound

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death.

Extended half-life products created through chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) or bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half-life by approximately 50% (half-life 18-19 hours), and thus, show promise in reduced dosing and maintaining activity levels above a 1% trough for a greater proportion of the dosing interval. However, patients with severe HA who are treated with extended half-life FVIII remain dependent on multiple infusions to maintain critical levels of FVIII activity. More recently, emicizumab has been approved for the treatment of HA. Emicizumab is a humanized, bispecific monoclonal antibody that binds to both activated coagulation factor IX (FIX) and coagulation factor X (FX), thereby mimicking the function of activated FVIII. Emicizumab represents an incremental improvement in therapeutic burden over replacement FVIII products since it can be administered subcutaneously as opposed to intravenously; however, it is still a chronic treatment that requires repeat administration every 1-4 weeks, and sustained FVIII activity levels above 5-10 IU/dL have rarely been attained even with extended half-life products. Therefore, there is a strong unmet need for a treatment that can alter the fundamental problem for patients with HA, namely attainment of sustained, hemostatically effective FVIII activity levels long-term (ie, over multiple years) without the need for treatment.

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of active FVIII following a single intravenous infusion of a vector encoding the appropriate gene sequence. Hemophilia A is well-suited for a gene replacement approach because clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in minute amounts (200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and even modest increases in the level of FVIII (any increase of the plasma level by 2 ng/ml induces an increase in activity of 1%) can ameliorate the severe form of hemophilia A. Thus, relatively small changes in endogenous FVIII activity can result in clinically relevant improvements in disease phenotype. Moreover, the circulating FVIII response to gene transduction can be assessed using validated quantitative endpoints that are easily assayed using established laboratory techniques.

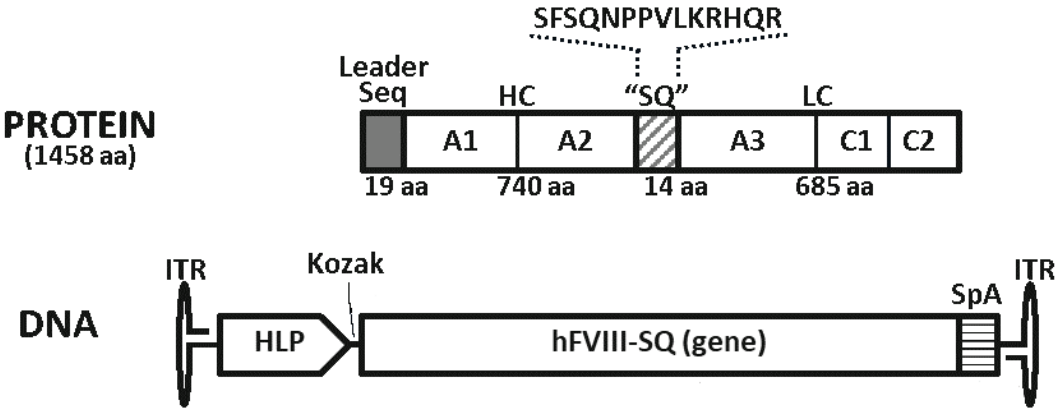
Several different gene transfer strategies for FVIII replacement have been evaluated, with adeno-associated viral (AAV) vectors used in most hemophilia clinical trials. AAV vectors have an excellent and well-defined safety profile, and can direct long-term transgene expression with tropism and promoter specificity for specific tissues, such as the liver (eg serotypes 2, 5, and 8). Indeed, an ongoing gene therapy clinical trial for a related disorder, hemophilia B, has established over 7 years of stable human factor IX (hFIX) expression at levels sufficient for conversion of their bleeding phenotype from severe to moderate or mild, following a single peripheral vein infusion of

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

AAV8-hFIX vector (Nathwani, 2018). Several participants in this trial have been able to discontinue factor prophylaxis without suffering spontaneous hemorrhages, even when they undertook activities that previously resulted in bleeding. Thus, gene therapy treatment has resulted in a substantial improvement in their quality of life.

BMN 270 is an AAV5-based gene therapy vector that expresses the SQ form of hFVIII under the control of a hybrid human liver-specific promoter (Figure 1).

Figure 1: AAV5-hFVIII-SQ Vector Genome and Encoded Protein



Legend –Note that schematic is not to scale; aa = amino acids; ITR = inverted terminal repeat; HLP = human liver promoter; Kozak = Kozak concensus sequence (GCCACC); SpA = Synthetic poly(A) signal

BMN 270 is delivered by a single IV dose and is designed to achieve stable, durable expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.

BMN 270 is being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. Three-year results from 270-201 have demonstrated that following gene transfer, mean and median FVIII activity levels above 15% (15 IU/dL), as measured by a chromogenic substrate assay, are achievable and sustained following a single infusion of 6E13 vg/kg of BMN 270, with administration of prophylactic corticosteroids and an acceptable safety profile (Pasi, 2019). In addition, an interim analysis of clinical study

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

270-301, an ongoing phase 3 study designed to assess the efficacy and safety of BMN 270 at a dose of 6E13 vg/kg and administration of therapeutic corticosteroids (ie, in response to ALT elevations), demonstrated FVIII activity levels that were also well above 15%, albeit lower than what was observed for the 6E13 vg/kg cohort in 270-201 at 26 weeks (Pasi, 2019).

Subjects enrolled and infused in 270-201 screened negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes (Boutin, 2010; Hayes, 2019).

Current data describing the extent of neutralization or inhibited transduction has been inconsistent across studies, each employing one of several varying methodologies to evaluate both vector-specific antibodies and transduction inhibition (TI). Early studies using AAV2 for the treatment of hemophilia B described inhibited transduction in patients with neutralizing antibody titers of 2 or 3 (Manno, 2006), but more recent studies have described successful transduction in patients who, in retrospect, were determined to have pre-existing antibody to the vector capsid (Majowicz, 2017). Importantly, no drug-related adverse events were described in these patients. As the sensitivity of assays used to detect pre-existing antibody has increased greatly in recent years, the biological relevance of any given titer determination, and comparisons across studies using assays with differing titer sensitivities, requires additional study.

Data from a non-clinical study conducted by BioMarin in cynomolgus monkeys further demonstrated that transduction efficiency can vary greatly in the presence of pre-existing anti-AAV5 antibody. In non-human primates, the ability to attain FVIII levels in the face of measurable AAV5 antibody titers without significant safety risks supports the possibility of achieving a similar response in patients with AAV5 antibody positivity. The totality of available data shows the effect of pre-existing immunogenicity to AAV delivery vectors is not completely understood, and indicates that some patients may benefit from AAV5 gene therapy who are currently excluded.

In this study, patients with severe HA and pre-existing antibodies against AAV5 will be enrolled to evaluate the clinical response to BMN 270 gene therapy. The aim is to evaluate the safety, tolerability, and efficacy of BMN 270 in patients with severe HA and pre-existing antibodies against AAV5 vector capsid at various levels of AAV5 antibody titers.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

OBJECTIVES:

The primary objective of the study is to:

- Assess the safety of a single intravenous administration of BMN 270 in severe HA subjects with pre-existing antibody to AAV5 vector capsid, including development of FVIII neutralizing antibody

Secondary objectives of the study are to:

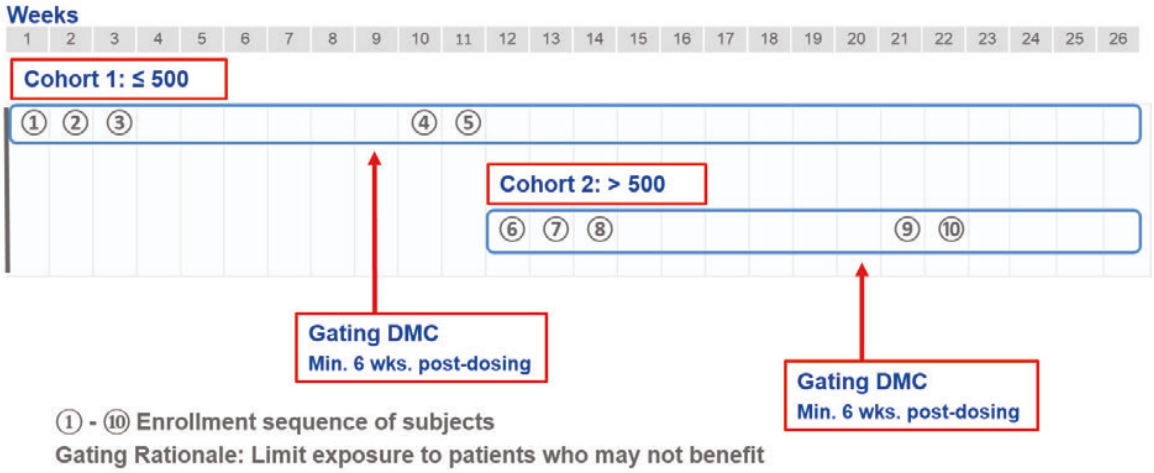
- Assess the efficacy of BMN 270 defined as FVIII activity at or above 5 IU/dL at Week 26
- Assess the impact of BMN 270 on usage of exogenous FVIII replacement therapy
- Assess the impact of BMN 270 on the number of bleeding episodes requiring exogenous FVIII therapy
- Evaluate the pharmacodynamics of FVIII expression following IV infusion of BMN 270
- Assess the impact of BMN 270 on patient-reported outcomes (PROs)

STUDY DESIGN AND PLAN:

This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to the AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects may be enrolled at 5-6 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb titer ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb titer > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date.

An independent Data Monitoring Committee (DMC) will consist of experts in clinical trials, statistics, and hemophilia. The DMC will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 2 represents one dosing schedule scenario):

Figure 2: 270-203 Dosing Schedule (One Possible Scenario)



Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from a minimum of 6 weeks post-infusion) reviewed. FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response, based on data from 270-201. Up to 6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness).

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC evaluation of safety and efficacy in treated subjects, with the following triggers that may potentially pause further enrollment:

- any related SAE;
- any related AE with a severity > CTCAE Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

Following a temporary halt of enrolment, the DMC may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed. Additional blood samples will be collected (tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine bead array (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.

Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DMC based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL. In subjects who experience recurring bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis.

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor or Sponsor-designated Data Monitor, follow an abbreviated visit schedule after Week 52 by attending only the Q12W and End of Year visits during Years 2-5.

The study analysis will be performed after all subjects have been followed for 26 weeks post-BMN 270 infusion, with presentation of safety, efficacy, and FVIII PD assessments. After the safety, efficacy, and FVIII PD analyses at 26 weeks post-BMN 270 infusion, long-term safety and efficacy will be assessed in all subjects for up to a total of 5 years post-infusion. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the DMC.

NUMBER OF SUBJECTS PLANNED:

Approximately 10 subjects

DIAGNOSIS AND ALL CRITERIA FOR INCLUSION AND EXCLUSION:

Individuals eligible to participate in this study must meet all of the following criteria:

1. Males ≥ 18 years of age with hemophilia A and residual FVIII levels ≤ 1 IU/dL as evidenced by medical history, at the time of signing the informed consent.
2. Detectable pre-existing antibodies against the AAV5 vector capsid as measured by AAV5 total antibody ELISA

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

3. Treated/exposed to FVIII concentrates or cryoprecipitate for a minimum of 150 exposure days (EDs)
4. Subject must have been on prophylactic FVIII replacement therapy for at least 12 months prior to study entry.
5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures.
6. No previous documented history of a detectable FVIII inhibitor, and results from a Bethesda assay or Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) (or less than 1.0 BU for laboratories with a historical lower sensitivity cutoff for inhibitor detection of 1.0 BU) on 2 consecutive occasions at least one week apart within the past 12 months (at least one of which should be tested at the central laboratory)
7. Sexually active participants must agree to use an acceptable method of effective contraception, either double-barrier contraception (ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with viral vector DNA below the limit of detection.
8. Willing to abstain from consumption of alcohol for at least the first 52 weeks following BMN 270 infusion.

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Any evidence of active infection or any immunosuppressive disorder, including HIV infection.
2. Significant liver dysfunction with any of the following abnormal laboratory results:
    - ALT (alanine aminotransferase) > 1.25x ULN;
    - AST (aspartate aminotransferase) > 1.25x ULN;
    - GGT (gamma-glutamyltransferase) > 1.25x ULN;
    - Total bilirubin > 1.25x ULN;
    - Alkaline phosphatase > 1.25x ULN; or
    - INR (international normalized ratio) ≥ 1.4

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

Subjects whose liver laboratory assessments fall outside of these ranges may undergo repeat testing of the entire liver test panel within the same Screening window and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor.

3. Prior liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used
4. Evidence of any bleeding disorder not related to hemophilia A
5. Platelet count of $< 100 \times 10^9$/L
6. Creatinine ≥ 1.5 mg/dL
7. Liver cirrhosis of any etiology as assessed by liver ultrasound
8. Chronic or active hepatitis B as evidenced by positive serology testing (hepatitis B surface antigen [HBsAg], hepatitis B surface antibody [HBsAb], and hepatitis B core antibody [HBcAb]) and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results .
9. Active Hepatitis C as evidenced by detectable HCV RNA, or currently on antiviral therapy
10. Active malignancy, except non-melanoma skin cancer
11. History of hepatic malignancy
12. History of arterial or venous thromboembolic events (eg, deep vein thrombosis, non-hemorrhagic stroke, pulmonary embolism, myocardial infarction, arterial embolus), with the exception of catheter-associated thrombosis for which anti-thrombotic treatment is not currently ongoing.
13. Known inherited or acquired thrombophilia, including conditions associated with increased thromboembolic risk, such as atrial fibrillation.
14. A history of known inflammatory, connective tissue, or autoimmune disorders (eg, vasculitis).
15. Treatment with any Investigational Product within 30 days or 5 half-lives of the investigational product (whichever is longer) prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including possible corticosteroid treatment outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.
17. Prior treatment with any vector or gene transfer agent
18. Major surgery planned in the 52-week period following the infusion with BMN 270
19. Use of systemic immunosuppressive agents, not including corticosteroids, or live vaccines within 30 days before the BMN 270 infusion
20. Concurrent enrollment in another clinical study, unless it is an observational (non-interventional) clinical study that does not interfere with the requirements of the current protocol or have the potential to impact the evaluation of safety and efficacy of BMN 270 and with prior consultation with the Medical Monitor
21. Known allergy or hypersensitivity to investigational product formulation
22. Unwilling to receive blood or blood products for treatment of an adverse event and/or a bleed

INVESTIGATIONAL PRODUCT(S), DOSE, ROUTE AND REGIMEN:
Each subject will receive a single IV infusion of BMN 270 at 6E13 vg/kg. The volume of infusion will depend on the subject's weight.

REFERENCE THERAPY(IES), DOSE, ROUTE AND REGIMEN:
No reference therapy will be evaluated in this study.

DURATION OF TREATMENT:
BMN 270 is given as a single dose by IV infusion.

CRITERIA FOR EVALUATION:

Safety:

The following safety outcome measurements will be assessed:

- Incidence of adverse events (AEs), including serious AEs (SAEs)
- Change in clinical laboratory tests (serum chemistry and hematology)
- Change in vital signs
- Change in physical examination

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

- Vector shedding (blood, urine, semen, feces, saliva)
- Liver tests (LTs, including ALT, AST, GGT, total bilirubin, and alkaline phosphatase)
- Immune response to FVIII transgene product and AAV5 vector capsid

No major toxicity is expected based on 270-201 data and non-clinical studies. Each subject will have comprehensive surveillance monitoring of LTs (once per week for Weeks 1-26). During the long-term safety evaluation, LTs will be monitored every three months for up to 5 years post-infusion; the frequency and duration of LT testing may be changed based on discussion between the Medical Monitor and the Investigator and review of subject data.

There will be a detailed assessment of cellular and humoral responses to AAV5 vector capsid and FVIII.

Efficacy:

The efficacy measure will be to assess plasma FVIII activity. The efficacy goal is to achieve FVIII activity ≥ 5 IU/dL at 26 weeks post-BMN 270 administration. Other efficacy measures include assessing the impact of BMN 270 on the use of FVIII replacement therapy and on the number of bleeding episodes during the study. Subjects will be asked to keep a subject diary, provided by the sponsor, to record the relevant details.

Other efficacy endpoints:

- Change from baseline in the total score of HAEMO-QoL-A at Week 26 of the study post-BMN 270 infusion
- Change from baseline in the EQ-5D-5L score at Week 26 of the study post-BMN 270 infusion.
- Change from baseline in the Haemophilia Activities List (HAL) score at Week 26 of the study post-BMN 270 infusion.
- Change from baseline in the Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) score at Week 26 of the study post-BMN 270 infusion.

Pharmacodynamics:

The FVIII antigen and activity level, as measured by a validated immunoassay and a validated FVIII activity assay, respectively, will be used for plasma profiles; FVIII antigen and activity will be used to determine PD parameters.

STATISTICAL METHODS:

Approximately 10 subjects may be dosed in the study. The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:
longitudinal methods, such as a mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at ≥ 5 IU/dL at 26 weeks following study treatment. FVIII activity will be assessed weekly during the study period. Assessment of the true steady state of FVIII will require that FVIII activity is measured after a minimum of 72 hours (or 5x the known half-life of the FVIII replacement therapy administered) has elapsed since the last infusion of FVIII concentrates. Analysis of neutralizing antibody response, other immunological parameters, and vector shedding will be primarily descriptive and involve both inter-subject and intra-subject comparisons across cohorts.		

3 TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

4 LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviations

AAV	adeno-associated virus
ABR	annualized bleeding rate
ADL	activities of daily living
ADR	adverse drug reaction
AE	adverse event
ALT	alanine aminotransferase
APTT	activated partial thromboplastin time
ART	anti-retroviral therapy
AST	aspartate aminotransferase
BPV	BioMarin Pharmacovigilance
BU	Bethesda Unit
CFR	Code of Federal Regulations
CRA	clinical research associate
CRF	case report form
CSR	clinical study report
CTCAE	Common Terminology Criteria for Adverse Events
DMC	Data Monitoring Committee
EC	ethics committee
eCRF	electronic case report form
ED	exposure days
EOSI	events of special interest
ETV	early termination visit
EudraCT	European Union Drug Regulating Authorities Clinical Trials
FDA	Food and Drug Administration
FIH	first-in-human
FVIII	coagulation factor VIII
GCP	Good Clinical Practice
GGT	gamma-glutamyltransferase
HA	Hemophilia A
HAL	Haemophilia Activities List
HBcAb	hepatitis B core antibody
HBsAb	hepatitis B surface antibody
HBsAg	hepatitis B surface antigen

hFIX	human coagulation factor IX
hFVIII	human coagulation factor VIII
HIPAA	Health Insurance Portability and Accountability Act
IB	investigator brochure
ICF	informed consent form
ICH	International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
ICH E6 [R2]	ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6
IND	Investigational New Drug (application)
INR	international normalized ratio
IP	investigational product
IRB	institutional review board
IV	intravenous
LT	liver test
MedDRA	Medical Dictionary for Regulatory Activities
MN	mobile nursing
PBMC	peripheral blood mononuclear cells
PCR	polymerase chain reaction
PD	pharmacodynamics
PEG	polyethylene glycol
PK	Pharmacokinetics
PRO	patient-reported outcome
rhFVIII	recombinant human FVIII protein
SAE	serious adverse event
SAP	statistical analysis plan
SDV	source data verification
SoA	schedule(s) of activities
TGA	thrombin generation assay
ULN	upper limit of normal
vg	vector genomes
VWF:Ag	von Willebrand factor Antigen
WPAI+CIQ:HS	Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific

Definition of Terms:

Investigational Product (IP):

"A pharmaceutical form of an active ingredient or placebo being tested or used as a reference in a clinical trial, including a product with a marketing authorization when used or assembled (formulated or packaged) in a way different from the approved form, or when used for an unapproved indication, or when used to gain further information about an approved use" (from International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6 [ICH E6] (R2)]).

The terms "IP" and "study drug" may be used interchangeably in the protocol.

5 ETHICS

BioMarin Pharmaceutical Inc. (hereafter referred to as BioMarin or the Sponsor) conducts its studies according to the highest ethical and scientific standards. The following Sections articulate standards to which Investigators will be held accountable, as well as matters of compliance to document adherence to such standards.

5.1 Institutional Review Board or Independent Ethics Committee

Investigators are expected to interact with Ethics Committees (ECs) promptly, as required, during the course of the study. This includes, but is not limited to, providing appropriate documentation to support study initiation and maintaining appropriate flow of safety and other information during the course of the study and for study close-out activities. BioMarin (or designee) will assist Investigators with access to timely and accurate information and with assurance of prompt resolution of any queries.

Prior to initiating the study, the Investigator will obtain written confirmation that the institutional review board (IRB) or independent ethics committee (EC) is properly constituted and compliant with International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) and Good Clinical Practice (GCP) requirements, applicable laws, and local regulations. A copy of the confirmation from the IRB/EC will be provided to BioMarin Pharmaceutical Inc. (BioMarin) or its designee. The Investigator will provide the IRB/EC with all appropriate material, including the protocol, Investigator's Brochure (IB), the Informed Consent Form (ICF) including compensation procedures, and any other written information provided to the subjects, including all ICFs translated for patients who do not speak the local language at the clinical site. The study will not be initiated and Investigational Product (IP) supplies will not be shipped to the site until appropriate documents from the IRB/EC confirming unconditional approval of the protocol, the ICF and all subject recruitment materials are obtained in writing by the Investigator and copies are received at BioMarin or its designee. The approval document should refer to the study by protocol title and BioMarin protocol number (if possible), identify the documents reviewed, and include the date of the review and approval. BioMarin will ensure that the appropriate reports on the progress of the study are made to the IRB/EC and BioMarin by the Investigator in accordance with applicable guidance documents and governmental regulations.

5.2 Ethical Conduct of Study

It is expected that Investigators understand and comply with the protocol. This includes, but is not limited to: establishing and meeting enrollment commitments, including providing eligible subjects for study enrollment; adhering to adverse event reporting, diagnostic, or other procedures as specified in the protocol; and assuring appropriate compliance with study treatment administration and accountability.

This study will be conducted in accordance with the following:

- European Clinical Trial Directive 2001/20/EC and Good Clinical Practice Directive 2005/28/EC, for studies conducted within any European country
- US Code of Federal Regulations (CFR) Sections that address clinical research studies, and/or other national and local regulations, as applicable
- ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6(R2) (ICH E6R2)
- The ethical principles established by the Declaration of Helsinki

Specifically, this study is based on adequately performed laboratory and animal experimentation. The study will be conducted under a protocol reviewed and approved by an IRB/EC and will be conducted by scientifically and medically qualified persons. The benefits of the study are in proportion to the risks. The rights and welfare of the subjects will be respected and the investigators conducting the study do not find the hazards to outweigh the potential benefits. Each subject will provide written, informed consent before any study-related tests or evaluations are performed.

5.3 Subject Information and Informed Consent

A properly written and executed informed consent form (ICF), in compliance with ICH E6R2 (Section 4.8), United States Code of Federal Regulations (CFR) 21 CFR §50, European Clinical Trial Directive 2001/20/EC and Good Clinical Practice Directive 2005/28/EC, and other applicable local regulations, will be obtained for each subject prior to entering the subject into the study. The Investigator will prepare the ICF and provide the documents to BioMarin for approval prior to submission to the IRB/EC approval. BioMarin and the IRB/EC must approve the documents before they are implemented. A copy of the approved ICF and if applicable, a copy of the approved subject information sheet and all ICFs translated to a language other than the native language of the clinical site must also be received by BioMarin or designee prior to any study-specific procedures being performed.

6 INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

During administration of informed consent, expectations regarding participation in the study should be made clear to subjects. Subjects who are not willing and/or are not able to comply with all aspects of the study should not be encouraged to participate.

Prior to beginning the study, the Investigator at each site must provide to BioMarin or designee a fully executed and signed Statement of Investigator (SOI) form. A US Food and Drug Administration (FDA) Form FDA 1572 serves as an acceptable SOI form. If Form FDA 1572 may not be used in a particular region, the Investigator must provide a fully executed SOI on the form provided by the Sponsor. All Investigators and Sub-Investigators must be listed on Form FDA 1572 or its equivalent SOI. Financial Disclosure Forms must also be completed for all Investigators and Sub-Investigators listed on the Form FDA 1572 or SOI who will be directly involved in the treatment or evaluation of subjects in this study.

The study will be administered by and monitored by employees or representatives of BioMarin. Clinical Research Associates (CRAs) or trained designees will monitor each site on a periodic basis and perform verification of source documentation for each subject as well as other required review processes. BioMarin's Regulatory Affairs Department (or designee) will be responsible for the timely reporting of serious adverse events (SAEs) to appropriate regulatory authorities as required.

In multicenter studies, a Coordinating Investigator will be identified who will be responsible for study overview. The Coordinating Investigator will read the clinical study report (CSR) and confirm that it accurately describes the conduct and results of the study, to the best of his or her knowledge. The Coordinating Investigator will be chosen on the basis of active participation in the study, ability to interpret data, and willingness to review and sign the report in a specified timeframe. The identity of the Coordinating Investigator and a list of all Investigators participating in the study will be provided in the CSR.

Clinical Laboratory assessments will be performed at a nominated central laboratory. Bioanalytical samples will be sent to the appropriate specialty laboratories for testing. Refer to laboratory manual for more details.

7 INTRODUCTION

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males (Nathwani, 1992). It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. Clinical manifestations of severe FVIII deficiency are frequent unprovoked bleeding episodes in joints and soft tissues causing permanent disability and occasionally death mostly after brain hemorrhage. Treatment in Western countries (Berntorp, 2012) consists of intravenous injection of plasma-derived or recombinant FVIII protein concentrates at the time of a bleed to control it or prophylactically to prevent bleeding episodes. The short half-life for FVIII (12-18 hours) necessitates frequent infusions and makes this treatment prohibitively expensive for the majority of the world's hemophilia A patients. These individuals develop debilitating arthropathy and have a substantially increased risk of death from hemorrhage in life (Stonebraker, 2010). Chemical modification or bioengineering of FVIII may improve half-life to 18-19 hours (Kaufman, 2013). However, these extended half-life FVIII variants do not eliminate the need for lifelong FVIII protein administration (Hay, 2012).

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of human FVIII (hFVIII) following a single administration of vector. Hemophilia A is well-suited for this approach because its clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in low amounts (100-200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and a modest increase in the level of FVIII (a plasma level of 2 ng/ml protein leads to a 1% expression) can ameliorate the severe phenotype (Srivastava, 2013); thus, the therapeutic goal for gene therapy is a modest increase in hFVIII. Finally, the consequences of gene transfer can be assessed using validated quantitative endpoints that can be easily assayed in most clinical laboratories.

BMN 270 contains the cDNA for the B-domain-deleted SQ FVIII with a liver-specific HLP transcription promoter. The expression cassette is inserted between AAV2 ITRs, and this genome is packaged in the AAV5 capsid. A comprehensive review of BMN 270 is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.1 Nonclinical Studies

The nonclinical program supports a single IV infusion of BMN 270, the planned clinical route of administration, for the treatment of hemophilia A in male patients. This nonclinical program took into account the guidelines and reflection papers for gene therapy medicinal products under EMA Advanced Therapies as well as FDA guidance. The primary pharmacodynamics (PD), pharmacokinetics (PK), and toxicity of IV BMN 270 were characterized in a series of single dose studies in species that were vector permissive and responsive to the transgene including normal CD-1 mice, a B- and T-cell deficient mouse model of hemophilia A (B6;129S-*F8*^{tm1Kaz}/J x B6.129S6-*Rag2*^{tm1Fwa} N12; FVIII KO x Rag2), and normal cynomolgus and rhesus monkeys. Some PD studies evaluated additional PK, immunogenicity and toxicity endpoints.

The comparative pharmacodynamics of BMN 270 in cynomolgus monkeys with varying pre-existing AAV5 transduction inhibition (TI) titer and AAV5 TAb status was evaluated in study BMN270-16-021. BMN 270 was administered to 4 groups of monkeys, a control group (Group 1, n=3) that tested negative for both TI and AAV5 TAb, Group 2 (n=4) that was AAV5 TAb negative, and low TI titer (2-5) positive. Group 3 (n=4) was also AAV5 TAb negative, but had higher TI titers (5-10). Group 4 (n=5) tested positive for both AAV5 TAb and TI (TI titers were >5). Administration of BMN 270 by a single intravenous bolus injection was well-tolerated in cynomolgus monkeys regardless of baseline TI titer or TAb status. After dosing, all monkeys showed FVIII-SQ levels above the LLOQ, with the exception of two monkeys in the group that presented with both positive TI and TAb titers at baseline. Though these TAb+ monkeys, regardless of TI titers, showed a significant mean reduction in FVIII expression (68% less) compared to TAb negative monkeys, three of five monkeys showed detectable levels of FVIII-SQ, with one having levels similar to that observed in the TI and TAb negative control group. Monkeys that were TI+ but TAb-at baseline had FVIII expression levels that were similar to those of the TI and TAb negative control group.

Results of the nonclinical program to date are detailed in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.2 Previous Clinical Studies

Ongoing clinical studies for BMN 270 include:

- 270-201, a phase 1/2, dose-escalation study in patients with severe HA

- 270-203, a phase 1/2 study in patients with severe HA who have anti-AAV5 antibody titers
- 270-301, a phase 3 study in patients with severe HA who receive BMN 270 at the 6E13 vg/kg dose level
- 270-302, a phase 3 study in patients with severe HA who receive BMN 270 at the 4E13 vg/kg dose level

A comprehensive review of safety, efficacy, and immunogenicity results from 270-201 and 270-301 as of the latest data cut is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.3 Study Rationale

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. The clinical phenotype of HA patients generally correlates tightly with the level of residual expression. Severe HA is classified as FVIII activity less than 1% of wild-type (< 1 IU/dL), moderate disease comprises 1-5% of wild-type activity, and the mild form is 5-40% activity. The clinical manifestations of severe HA are frequent spontaneous bleeding episodes, predominantly in joints and soft tissues, with a substantially increased risk of death from hemorrhage when the brain is involved.

Treatment of severe HA presently mainly consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study (Berntorp, 2017) and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with on-demand-only therapy in the same studies, respectively. Continued repeated bleeding events compound debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death.

Extended half-life products created through chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) or bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half-life by approximately 50% (half-life 18-19 hours), and thus, show promise in reduced dosing and maintaining activity levels above a 1% trough for a greater proportion of the dosing interval. However, patients with severe HA who are treated with extended half-life FVIII remain dependent on multiple infusions to maintain critical levels of FVIII activity. More recently, emicizumab has been approved for the treatment of HA. Emicizumab is a humanized, bispecific monoclonal antibody that binds to both activated coagulation factor IX (FIX) and coagulation factor X (FX), thereby mimicking the function of activated FVIII. Emicizumab represents an incremental improvement in therapeutic burden over replacement FVIII products since it can be administered subcutaneously as opposed to intravenously; however, it is still a chronic treatment that requires repeat administration every 1-4 weeks, and sustained FVIII activity levels above 5-10 IU/dL have rarely been attained even with extended half-life products. Therefore, there is a strong unmet need for a treatment that can alter the fundamental problem for patients with HA, namely attainment of sustained, hemostatically effective FVIII activity levels long-term (ie, over multiple years) without the need for treatment.

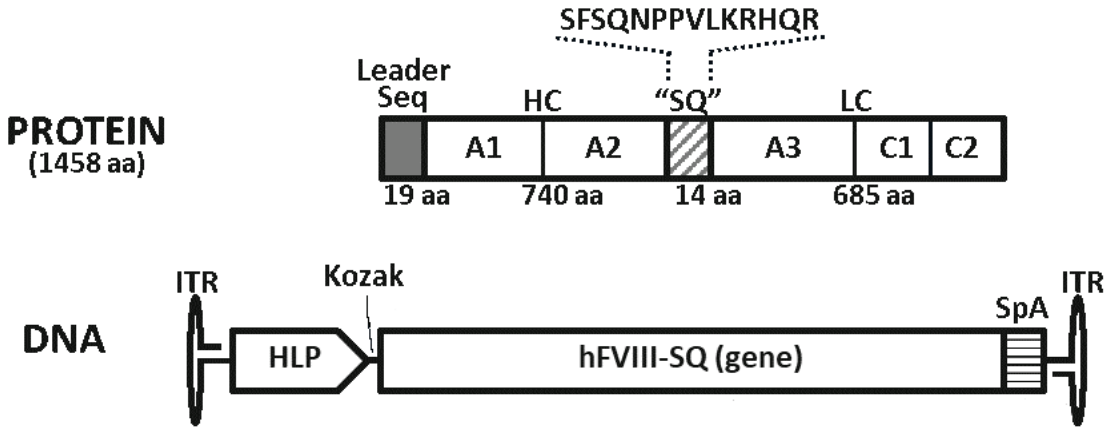
Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of active FVIII following a single intravenous infusion of a vector encoding the appropriate gene sequence. Hemophilia A is well-suited for a gene replacement approach because clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in minute amounts (200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and even modest increases in the level of FVIII (any increase of the plasma level by 2 ng/ml induces an increase in activity of 1%) can ameliorate the severe form of hemophilia A. Thus, relatively small changes in endogenous FVIII activity can result in clinically relevant improvements in disease phenotype. Moreover, the circulating FVIII response to gene transduction can be assessed using validated quantitative endpoints that are easily assayed using established laboratory techniques.

Several different gene transfer strategies for FVIII replacement have been evaluated, with adeno-associated viral (AAV) vectors used in most hemophilia clinical trials. AAV vectors have an excellent and well-defined safety profile, and can direct long-term transgene expression with tropism and promoter specificity for specific tissues, such as the liver (eg serotypes 2, 5, and 8). Indeed, an ongoing gene therapy clinical trial for a related disorder, hemophilia B, has established over 7 years of stable human factor IX (hFIX) expression at levels sufficient for conversion of their bleeding phenotype from severe to moderate or mild,

following a single peripheral vein infusion of AAV8-hFIX vector (Nathwani, 2018). Several participants in this trial have been able to discontinue factor prophylaxis without suffering spontaneous hemorrhages, even when they undertook activities that previously resulted in bleeding. Thus, gene therapy treatment has resulted in a substantial improvement in their quality of life.

BMN 270 is an AAV5-based gene therapy vector that expresses the SQ form of hFVIII under the control of a hybrid human liver-specific promoter (Figure 7.3.1).

Figure 7.3.1: AAV5-hFVIII-SQ Vector Genome and Encoded Protein



Legend –Note that schematic is not to scale; aa = amino acids; ITR = inverted terminal repeat; HLP = human liver promoter; Kozak = Kozak concensus sequence (GCCACC); SpA = Synthetic poly(A) signal

BMN 270 is delivered by a single IV dose and is designed to achieve stable, durable expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.

BMN 270 is being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. Three-year results from 270-201 have demonstrated that following gene transfer, mean and median FVIII activity above 15% (15 IU/dL), as measured by a chromogenic substrate assay, are achievable and sustained following a single infusion of 6E13 vg/kg of BMN 270, with administration of prophylactic corticosteroids and an acceptable safety profile (Pasi, 2019). In addition, an interim analysis of clinical study 270-301, an ongoing phase 3 study designed

to assess the efficacy and safety of BMN 270 at a dose of 6E13 vg/kg and administration of on-demand corticosteroids (ie, in response to ALT elevations), demonstrated FVIII activity levels that were also well above 15% at 26 weeks, albeit lower than what was observed for the 6E13 vg/kg cohort in 270-201 (Pasi, 2019). For additional information on preliminary data in 270-201, refer to the current version of the Investigator's Brochure.

Subjects enrolled and infused in 270-201 were screened and negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes (Hayes, 2019; Boutin, 2010).

Current data describing the extent of neutralization or inhibited transduction has been inconsistent across studies, each employing one of several varying methodologies to evaluate both vector-specific antibodies and transduction inhibition (TI). Early studies using AAV2 for the treatment of hemophilia B described inhibited transduction in patients with neutralizing antibody titers of 2 or 3 (Manno, 2006), but more recent studies have described successful transduction in patients who, in retrospect, were determined to have pre-existing antibody to the vector capsid (Majowicz, 2017). Importantly, no drug-related adverse events were described in these patients. As the sensitivity of assays used to detect pre-existing antibody has increased greatly in recent years, the biological relevance of any given titer determination, and comparisons across studies using assays with differing titer sensitivities, requires additional study.

Data from a non-clinical study conducted by BioMarin in cynomolgus monkeys further demonstrated that transduction efficiency can vary greatly in the presence of pre-existing anti-AAV5 antibody. In non-human primates, the ability to attain FVIII levels in the face of measurable AAV5 antibody titers without significant safety risks supports the possibility of achieving a similar response in patients with AAV5 antibody positivity. The totality of available data shows the effect of pre-existing immunogenicity to AVV delivery vectors is not completely understood, and indicates that some patients may benefit from AAV5 gene therapy who are currently excluded.

In this study, patients with severe HA and pre-existing antibodies against AAV5 will be enrolled to evaluate the clinical response to BMN 270 gene therapy. The aim is to evaluate

the safety, tolerability, and efficacy of BMN 270 in patients with severe HA and pre-existing antibodies against AAV5 vector capsid at various levels of AAV5 antibody titers.

7.4 Summary of Overall Risks and Benefits

The majority of subjects in the ongoing 270-201 clinical study who have received 4E13 or 6E13 vg/kg doses of BMN 270 have had Grade 1 asymptomatic elevations in ALT that have reached only slightly above the upper limit of normal (ULN). Subjects in the 6E13 vg/kg cohort received corticosteroids, predominantly on a prophylactic basis, starting 2-4 weeks following BMN 270 infusion, whereas those in the 4E13 vg/kg cohort received corticosteroids only if they experienced an elevation in their ALT ≥ 1.5x ULN (ie, "therapeutic"). Based on the effectiveness of transient oral corticosteroids used starting 8-10 weeks after vector infusion to suppress a presumed Class 1 (cytotoxic T-cell) response in prior studies with hepatic transduction with AAV vectors (Mingozzi, 2013), subjects were treated with 7-32 weeks of oral corticosteroids prophylactically or in response to the elevations in ALT to limit hepatotoxicity, ensure preservation of the transduced hepatocytes, and minimize any associated impact of such on FVIII levels. Using this approach, no sustained loss of FVIII activity has been observed in subjects with ALT elevations. Moreover, the rise in ALT levels was not accompanied by significant or lasting abnormalities in other liver tests such as AST, bilirubin or albumin, indicating that extent of toxicity is limited. At the highest dose tested in 270-201 (6E13 vg/kg), the majority of subjects achieved chromogenic FVIII activity levels above 50 IU/dL at 52 weeks post-infusion. Subjects in that cohort also reported markedly decreased bleeding compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. Subjects at all dose levels continue to be followed.

In the ongoing 270-301 clinical study, a majority of subjects, all dosed with 6E13 vg/kg BMN 270, have experienced asymptomatic elevations in ALT. Subjects received therapeutic corticosteroids if they experienced an ALT elevation and/or had a FVIII activity level that had declined >35% from peak values. While sporadically observed cytotoxic T-cell responses have not been correlated with ALT elevations, the majority of subjects with transient rises in ALT levels had associated declines in their FVIII activity that subsequently increase to higher than pre-ALT elevation levels following initiation of therapeutic corticosteroids. Similar to 270-201, the rise in ALT levels in 270-301 were not accompanied by significant or lasting aberrations in other liver tests such as AST, bilirubin or albumin, indicating that extent of toxicity is limited. At the time of the 270-301 interim analysis, the modified intent-to-treat population (n=16, who had completed ≥26 weeks on-study) had mean and median chromogenic FVIII activity levels of 36.1 and 33.2 IU/dL at 26 weeks,

respectively, which are lower than the corresponding values observed for the 270-201 6E13 vg/kg cohort.

There has been one HIV-positive subject in the ongoing 270-302 clinical study who experienced Grade 3 asymptomatic elevations in ALT and AST, which has been attributed to an interaction between one or more of his antiretroviral therapy medications and/or unsuspected underlying hepatic disease with BMN 270. In addition, there have been three subjects, including one with Gilbert's syndrome, in the ongoing 270-301 clinical study who have experienced Grade 3 asymptomatic elevations in ALT and AST. These cases have led to the exclusion of subsequent HIV-positive subjects and requirement of liver tests at Screening that are <1.25 times the upper limit of the normal range in the ongoing 270-301 and 270-302 clinical studies. Of note, two HIV-positive subjects in 270-301 and one presumed Gilbert's syndrome subject in 270-201 have received BMN 270 without experiencing any elevations in ALT beyond Grade 1 to date. Overall, the literature and clinical experience with BMN 270 suggests that transient elevations in liver enzymes are expected following AAV-based gene therapy for the treatment for hemophilia A or B without any long-term concerns of hepatic injury (Manno, 2006; Nathwani, 2011; George, 2016; Miesbach, 2016; Pasi, 2017).

As with any infused biological product, there is a potential risk of acute, systemic hypersensitivity reactions (including anaphylaxis) with BMN 270. No hypersensitivity reactions were observed during dosing of BMN 270 in the 270-201 clinical study, although infusion-related hypersensitivity reactions (including anaphylaxis) have been observed during dosing of BMN 270 in the 270-301 clinical study (refer to Investigator's Brochure for full details). All of the infusion-related reactions were effectively managed clinically and resolved without any clinical sequelae.

The current data available for BMN 270 has shown an established positive benefit:risk profile for BMN 270 at the 6E13 vg/kg dosing level, although impact of prophylactic corticosteroids requires further investigation. . Given the monitoring measures in place in the clinical protocol(s) to minimize the risk to subjects participating in the existing studies, the identified risks are justified by the anticipated benefits that may be afforded to subjects. Each subject in 270-203 will have a comprehensive surveillance plan that monitors LTs during the study, and elevations in LT will be addressed according to the guidelines set forth in the protocol. Safety will be assessed by adverse event reporting and clinical laboratory assessments.

For additional information on findings from other BMN 270 clinical studies, refer to the current version of the IB.

8 STUDY OBJECTIVES

The primary objective of the study is to:

- Assess the safety of a single intravenous administration of BMN 270 in severe HA subjects with pre-existing antibody to AAV5 vector capsid, including development of FVIII neutralizing antibody

The secondary objectives of the study are to:

- Assess the efficacy of BMN 270 defined as FVIII activity at or above 5 IU/dL at Week 26
- Assess the impact of BMN 270 on usage of exogenous FVIII replacement therapy
- Assess the impact of BMN 270 on the number of bleeding episodes requiring exogenous FVIII therapy
- Evaluate the pharmacodynamics of FVIII expression following IV infusion of BMN 270
- Assess the impact of BMN 270 on patient-reported outcomes (PROs)

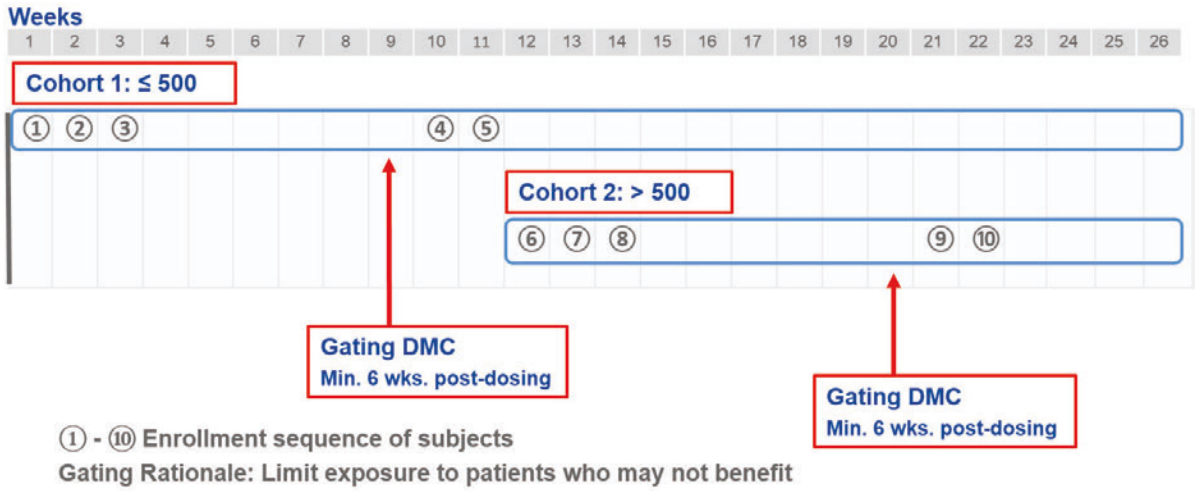
9 INVESTIGATIONAL PLAN

9.1 Overall Study Design and Plan

This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects may be enrolled at 5-6 sites globally in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date.

An independent Data Monitoring Committee (DMC), consisting of experts in clinical trials, statistics, and hemophilia, has been convened. The DMC will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 9.1.1 represents one dosing schedule scenario):

Figure 9.1.1: 270-203 Dosing Schedule (One Scenario)



Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from a minimum of 6 weeks post-infusion) reviewed. Based on data from 270-201, FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response. Up to

6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness).

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC evaluation of safety and efficacy in treated subjects with the following triggers that may potentially pause further enrollment:

- any related SAE;
- any related AE with a severity > CTCAE Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects at 6 weeks post-BMN 270 infusion.

Following a temporary halt of enrolment, the DMC may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed. Additional blood samples will be collected (tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine bead array (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.

Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DMC based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL.

As the relationship between activity assay results of the BMN 270 gene product and bleeding remains to be established, Investigators should strive to minimize bias by avoiding consideration of FVIII activity levels by themselves or subjects in the reporting of bleeding episodes and FVIII usage.

In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis. Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor or Sponsor-designated Data Monitor, follow an abbreviated visit schedule after Week 52 by attending only the Q12W and End of Year visits during Years 2-5.

The study analysis will be performed after all subjects have been followed for 26 weeks post-BMN 270 infusion, with presentation of safety, efficacy, and FVIII PD assessments. After the safety, efficacy, and FVIII PD analyses at 26 weeks post-BMN 270 infusion, long-term safety and efficacy will be assessed in all subjects for up to a total of 5 years post-infusion. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the DMC.

A summary of all assessments is provided in the Schedule of Activities (SoA) in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5.

Table 9.1.1: Schedule of Activities – Screening/Baseline/Day 1

Assessment	Prior to BMN 270 Infusion				BMN 270 Infusion Visit (Day 1)[k]
	Screening		Smart Rescreening[i] (Day -28 to Day -1)	Baseline (Day -7 to Day -1)[j]	
	Screening (Day -42 to Day -29)	Screening (Day -28 to Day -1)			
Informed consent	X	X			
Demographics (age, sex, race, ethnicity)		X			
Medical History (including hemophilia A history, Hepatitis B, Hepatitis C, and HIV)		X			
Physical Examination[a]		X		X	X
Height and Weight[a]		X			
Vital Signs		X	X	X	X
Assessment of Adverse Events and Concomitant Medications		X	X	X	X
Documentation of bleeding episodes and FVIII usage for previous 12 months (by either subject or clinical information)		X	X	X	
Distribution of subject diaries and training in their use[b]		X			
Electrocardiogram		X			
Liver Ultrasound		X			
hFVIII Assays[c]		X	X[l]	X	
AAV5 TAb Assay (ARUP)[d]	X		X		X
AAV5 TAb Assays[e]				X	
AAV5 TI Assay[e]				X	X
Screen for Hepatitis B, Hepatitis C, HIV[f]		X			
Blood chemistry, hematology, and coagulation tests[g]		X	X	X	
Blood fasting lipid panel					X
Urine Tests[g]		X	X	X	

Assessment	Prior to BMN 270 Infusion				BMN 270 Infusion Visit (Day 1)[k]
	Screening		Smart Rescreening[i] (Day -28 to Day -1)	Baseline (Day -7 to Day -1)[j]	
	Screening (Day -42 to Day -29)	Screening (Day -28 to Day -1)			
Liver Tests[g]		X	X	X	
PBMC collection (for baseline determination of AAV5 and FVIII specific cellular immunity)				X	
Von Willebrand Factor Antigen (VWF:Ag)				X	
Thrombin Generation Assay				X	
PCR of vector DNA in blood, saliva, urine, semen, and stools				X	X[k]
Biomarker testing[h]		X			
Exploratory biomarker assessments[h]				X	X
Cytokine bead array assay				X	
Haemo-QOL-A assessment				X	
EQ-5D-5L assessment				X	
HAL assessment				X	
WPAI+CIQ:HS assessment				X	
BMN 270 Infusion					X
Hypersensitivity blood assessments				X	(X)[m]

[a] Complete physical examination should be done at Screening. Brief physical examination may be done at Baseline and at the BMN 270 Infusion Visit.

[b] Diaries should be distributed to subjects who have consented to participate in the study and who have been determined to meet all study eligibility criteria.

[c] Includes baseline FVIII activity (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), coagulation exploratory assay, hFVIII inhibitor level (Bethesda assay with Nijmegen modification), hFVIII total antibody titer, and hFVIII antigen assay. Baseline activity should be assessed at trough (at least >72 hours after last dose of replacement FVIII therapy, or 5x the known half-life of the FVIII concentrates administered).

[d] Screening, Smart Re-screening, and Infusion Day samples will be tested using the ARUP AAV5 TAb assay. During Screening, the ARUP AAV5 TAb assay test may be done first, under a standalone informed consent form, before the main ICF for the study is signed and further screening procedures are performed. Sample collection on the day of the infusion visit must be performed before the BMN 270 infusion is given.

[e] Baseline and all post-dose samples will be tested in a different AAV5 TAb post-dose immunogenicity monitoring assay.

[f] Subjects with documented negative results within the last 30 days do not need to be retested. Hepatitis B screening should include hepatitis B surface antigen (HBsAg), hepatitis B surface antibody (HBsAb), and hepatitis B core antibody (HBcAb).

[g] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. ABO blood typing assessment should be performed at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study.

[g] Includes HLA genotyping and FVIII genotyping.

[h] Blood samples will be collected from subjects to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for those evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[i] Smart rescreening should only be performed if a subject has been determined to be eligible for the study and is unable to complete the Baseline assessments and Infusion prior to the closing of the original Screening window (28 + 14 days). Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once.

[j] Should the screening visit occur within 30 days of the drug infusion, physical examination, blood chemistry, LTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline.

[k] With the exception of the collection of samples for PCR vector DNA analysis, assessments on the day of infusion must be performed prior to the infusion. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit. On the day of the BMN 270 Infusion, vital signs will be monitored prior to the infusion, during the infusion every 15 minutes (± 5 minutes), and following the infusion hourly (± 5 minutes) for a total time of 24 hours during the subject's stay in the clinic. Shedding samples for PCR of vector DNA analysis (blood, saliva, urine, semen, stool) should be collected between 2 and 24 hours after the infusion has been completed.

[l] Only the hFVIII inhibitor level (Bethesda assay with Nijmegen modification) assay must be done at smart rescreening.

[m] In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination, vital signs will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.

Table 9.1.2: Schedule of Activities – Post-Infusion Follow-Up (Week 1-16)

Assessment	Follow-Up After BMN 270 Infusion – Weeks*																	
	Week 1																	
	D2	D4	D8	2	3	4	5[g]	6	7[g]	8	9[g]	10	11[g]	12	13[g]	14	15[g]	16
Study Day*	2	4	8	15	22	29	36	43	50	57	64	71	78	85	92	99	106	113
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight						X				X				X				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X		X		X		X						X		
Urine Tests[b]														X				
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII antibody titer				X		X		X		X		X		X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]	X[d]	X	X	X	X	X		X		X				X				X
Exploratory biomarker assessments[e]				X				X				X				X		
Haemo-QOL-A assessment														X				
EQ-5D-5L assessment														X				
HAL assessment														X				
WPAI+CIQ:HS assessment														X				
AAV5 TAb Assay	X		X	X		X		X		X		X		X		X		X
AAV5 TI Assay	X		X	X		X		X		X		X		X		X		X

Assessment	Follow-Up After BMN 270 Infusion – Weeks*																	
	Week 1																	
	D2	D4	D8	2	3	4	5[g]	6	7[g]	8	9[g]	10	11[g]	12	13[g]	14	15[g]	16
Study Day*	2	4	8	15	22	29	36	43	50	57	64	71	78	85	92	99	106	113
Testing for reactivation of hepatitis B and hepatitis C																		X[f]
PBMC collection (for determination of AAV5 and FVIII specific immunity)	X			X		X		X		X		X		X		X		X
VWF:Ag						X				X				X				X
Cytokine bead array assay	X		X	X				X				X				X		

* Visit windows are ± 24 hours during Week 1 and ± 48 hours starting with the Week 2 visit.

[a] Brief physical examination should be done at all weekly visits.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs may be monitored more or less frequently (and in particular when ALT values are ≥ 1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ≥ 1.5x ULN or > ULN & > 2x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen assay. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Collection for each matrix to occur until at least 3 consecutive results below the limit of detection are obtained. Collection and testing of semen samples must continue through Week 12, even if 3 consecutive results below the limit of detection in that compartment have already been recorded. Separate collection on Day 2 is not required if vector shedding samples were collected on Day 1 post-BMN 270 infusion; if collection on Day 2 is needed, it must occur within 24 hours of the time of the BMN 270 infusion.

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] Testing for reactivation of hepatitis B and hepatitis C at Week 16, for subjects with a past medical history of hepatitis B or hepatitis C prior to study entry, should be performed only in subjects who have not received prophylactic oral corticosteroids; subjects who have received prophylactic oral corticosteroids will have hepatitis B and hepatitis C testing at the time points indicated in Table 9.1.6.

[g] The scheduled visits at Week 5, Week 7, Week 9, Week 11, Week 13, and Week 15 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Events will not be performed at these MN or lab draw-only visits.

Table 9.1.3: Schedule of Activities – Post-Infusion Follow-Up (Week 17-32)

Assessment	Follow-Up After BMN 270 Infusion – Weeks*															
	17[f]	18	19[f]	20	21[f]	22	23[f]	24	25[f]	26	27[f]	28	29[f]	30	31[f]	32
Study Day*	120	127	134	141	148	155	162	169	176	183	190	197	204	211	218	225
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight[a]				X						X						X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X						X						X
Urine Tests[b]										X						
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII antibody titer				X				X		X						X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]				X				X		X						X
Exploratory biomarker assessments[e]		X				X				X						X
Haemo-QOL-A assessment										X						
EQ-5D-5L										X						
HAL										X						
WPAI+CIQ:HS										X						
AAV5 TAb Assay		X		X		X		X		X		X		X		X
AAV5 TI Assay		X		X		X		X		X		X		X		X

Assessment	Follow-Up After BMN 270 Infusion – Weeks*															
	17[f]	**18**	**19[f]**	**20**	**21[f]**	**22**	**23[f]**	**24**	**25[f]**	**26**	**27[f]**	**28**	**29[f]**	**30**	**31[f]**	**32**
Study Day*	120	127	134	141	148	155	162	169	176	183	190	197	204	211	218	225
PBMC collection (for determination of AAV5 and FVIII specific cellular immunity)		X		X		X		X		X		X		X		X
VWF:Ag				X						X						
Thrombin Generation Assay[e]				X				X		X						X
Cytokine bead array assay				X				X		X						X

* Visit windows are ± 48 hours.

[a] Brief physical examination should be done at all weekly visits except Week 26, where a complete physical examination should be performed. Weight should be recorded at Week 20 and Week 26.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs may be monitored more or less frequently (and in particular when ALT values are ≥ 1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ≥ 1.5x ULN or > ULN & > 2x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Collection for each matrix to occur until at least 3 consecutive results below the limit of detection are obtained.

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes

that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] The scheduled visits at Week 17, Week 19, Week 21, Week 23, Week 25, Week 27, Week 29, and Week 31 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Events will not be performed at these MN or lab draw-only visits.

Table 9.1.4: Schedule of Activities – Post-Infusion Follow-Up (Week 33-52)

Assessment	Year 1 – Weeks*											
	33[e]	34	35[e]	36	38[e]	40	42[e]	44	46[e]	48	50[e]	52
Study Day[*]	232	239	246	253	267	281	295	309	323	337	351	365
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X
Weight[a]				X				X				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X				X				X
Urine Tests[b]				X								X
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]	X	X	X	X	X	X	X	X	X	X	X	X
AAV5 TAb Assay		X		X		X		X		X		X
AAV5 TI Assay		X		X		X		X		X		X
FVIII antibody titer				X				X				X
Exploratory biomarker assessments[d]				X		X		X		X		X
PBMC Collection (for determination of FVIII and Capsid specific cellular immunity)		X		X				X				X
VWF:Ag				X								X
Thrombin Generation Assay[d]				X		X		X		X		X
Cytokine bead array assay				X				X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools				X		X		X		X		X
Haemo-QOL-A assessment												X
EQ-5D-5L												X
HAL												X

Assessment	Year 1 – Weeks*											
	33[e]	34	35[e]	36	38[e]	40	42[e]	44	46[e]	48	50[e]	52
WPAI+CIQ:HS												X

* Visit windows are ± 48 hours through Week 36, then ±1 week until Week 52.

[a] Complete physical examination should be performed at Week 52; brief physical exam may be performed at other study visits. Weight should be recorded at Week 36 and every 4 weeks through Week 52.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ≥ 1.5x ULN or > ULN & > 2x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN or increased by > 10 U/L from prior assessment; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII protein assay. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[e] The scheduled visits at Week 33, Week 35, Week 38, Week 42, Week 46, and Week 50 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Events will not be performed at these MN or lab draw-only visits.

Table 9.1.5: Schedule of Events – Long-Term Follow-Up (Year 2 – Year 5)

Assessment	Years 2-5*		End of Year Visit				ETV
			Year 2	Year 3	Year 4	Year 5	
Study Week*	Q12W	Q6W[gh]	W104	W156	W208	W260	
Physical examination[a]	X[a]		X[a]				X
Weight[a]	X[a]		X[a]				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X				X
Vital Signs	X		X				X
Blood chemistry, hematology, and coagulation tests[b]	X[b]		X[b]				X
Urine Tests[b]	X[b]		X[b]				X
Liver Tests[b]	X	X	X				X
FVIII assays[c]	X	X	X				X
AAV5 TAb Assay	X		X				X
AAV5 TI Assay	X		X				X
FVIII antibody titer	X		X				X
Exploratory biomarker assessments[e]	X		X				X
PBMC Collection (for determination of FVIII and Capsid specific cellular immunity)	X		X				X
VWF:Ag	X		X				X
Thrombin Generation Assay[e]	X		X				X
Cytokine bead array assay	X		X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]	(X)[d]	(X)[d]	(X)[d]				(X)[d]
Haemo-QOL-A assessment			X[f]				X
EQ-5D-5L			X[f]				X
HAL			X[f]				X

Assessment	Years 2-5*		End of Year Visit				ETV
			Year 2	Year 3	Year 4	Year 5	
Study Week*	Q12W	Q6W[gh]	W104	W156	W208	W260	
WPAI+CIQ:HS			X[f]				X

* Visit windows are ± 2 weeks for visits in Years 2-5. The Q6W visits during Years 2-5 should restart after each End of Year visit (eg, the first Q6W visit during Year 3 should be ~6 weeks after the End of Year 2 visit).

[a] Brief physical examination should be performed at all visits during Years 2-5. Weight should be recorded at the second Q12W visit each year and at every End of Year visit during Years 2-5.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ≥ 1.5x ULN or > ULN & > 2x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN or increased by > 10 U/L from prior assessment; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. During Years 2-5 of the Post-Infusion Follow-Up period, urine tests and blood, chemistry, and coagulation tests should be performed at the second Q12W visit each year and at every End of Year visit.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII protein assay. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism. If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.

[d] Sample testing during Long-Term Follow-Up is not required if at least 3 consecutive samples were below the limit of detection during the Post-Infusion Follow-Up period. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 52 should continue to have PCR testing of semen every 6 weeks during Years 2-5 until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes

that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] PRO assessments during Years 2-5 of Long-Term Follow-up should be performed at every End of Year visit.

[g] Subjects who meet the definition of treatment failure to BMN 270 therapy after Week 52 (refer to Section 12.6) may omit the Q6W visits during Years 2-5, and must attend only the Q12W and End of Year visits. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in all fluids must still provide samples Q6W during Years 2-5 until vector shedding has been cleared (but do not need to do other scheduled assessments at those visit dates).

[e] The scheduled Q6W visits during Years 2-5 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Events will not be performed at these MN or lab draw-only visits.

Table 9.1.6: Suggested Schedule of Events – Prophylactic Corticosteroids

Assessment	Corticosteroid Treatment Period[b]																Post-Corticosteroid Period[c]				
	Day 1	Week															Week				
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	1	2	3	4	13
Prophylactic corticosteroid dose (mg/day)	40 mg	40 mg	40 mg	40 mg	40 mg	35 mg	35 mg	30 mg	30 mg	25 mg	25 mg	20 mg	20 mg	15 mg	10 mg	5 mg					
FVIII activity testing	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Liver tests	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Hepatitis B testing[d]							X										X				X
HCV Viral Load[d]							X										X				X

[a] This table provides an example of a prophylactic corticosteroid course. Clinical judgment, weighing the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses, and discussions between the Investigator and Medical Monitor are advised for any questions or concerns.

[b] Following initiation or completion of corticosteroid regimen, if a recurrence of ALT values ≥ ULN or ≥ 2x baseline value is reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor. Modification of the corticosteroid regimen may take into consideration possible confounders for the ALT elevation, relationship between increases in ALT and FVIII activity, ALT/FVIII levels post-corticosteroid initiation, and adverse events related to corticosteroid dosing. Guidance for tapering oral corticosteroid dosing can be found in Section 9.4.8.2, although a discussion between the PI and Medical Monitor should take place prior to tapering the corticosteroid dose.

[c] After discontinuation of oral corticosteroids, weekly labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values. If these assessments are already being done as part of normal study follow-up, they do not need to be duplicated.

[d] Should only be performed in subjects with a history of hepatitis B or hepatitis C prior to study entry.

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9.2 Discussion of Study Design, Including Choice of Control Group

Study 270-203 is designed to be a Phase 1/2, single-arm, open-label study in hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid. Hemophilia A patients who provide written informed consent and meet the entry criteria will be eligible to enroll in the study.

Approximately 10 subjects may be enrolled at the 6E13 vg/kg BMN 270 dose. Subjects will be followed for 26 weeks post-BMN 270 infusion during which safety and efficacy assessments will be taken. After the final analysis at 26 weeks post-infusion, safety and efficacy will then continue to be assessed long-term for approximately a total of 5 years.

9.3 Selection of Study Population

Approximately 10 hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid may enroll into the study.

Additional criteria for participation in the study are provided in Section 9.3.1 and Section 9.3.2.

9.3.1 Inclusion Criteria

Individuals eligible to participate in this study must meet all of the following criteria:

1. Males ≥ 18 years of age with hemophilia A and residual FVIII levels ≤ 1 IU/dL as evidenced by medical history, at the time of signing the informed consent
2. Detectable pre-existing antibodies against the AAV5 vector capsid as measured by AAV5 total antibody ELISA
3. Treated/exposed to FVIII concentrates or cryoprecipitate for a minimum of 150 exposure days (EDs)
4. Subject must have been on prophylactic FVIII replacement therapy for at least 12 months prior to study entry.
5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures.
6. No previous documented history of a detectable FVIII inhibitor, and results from a Bethesda assay or Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) (or less than 1.0 BU for laboratories with a historical lower sensitivity cutoff for inhibitor detection of 1.0 BU) on 2 consecutive occasions at least one week apart within the past 12 months (at least one of which should be tested at the central laboratory)

7. Sexually active participants must agree to use an acceptable method of effective contraception, either double barrier contraception (ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with viral vector DNA below the level of detection.
8. Willing to abstain from consumption of alcohol for at least the first 52 weeks following BMN 270 infusion.

9.3.2 Exclusion Criteria

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Any evidence of active infection or any immunosuppressive disorder, including HIV infection.
2. Significant liver dysfunction with any of the following abnormal laboratory results:
    - ALT (alanine aminotransferase) > 1.25x ULN;
    - AST (aspartate aminotransferase) > 1.25x ULN;
    - GGT (gamma-glutamyltransferase) > 1.25x ULN;
    - Total bilirubin > 1.25x ULN;
    - Alkaline phosphatase > 1.25x ULN; or
    - INR (international normalized ratio) ≥ 1.4

    Subjects whose laboratory assessments fall outside of these ranges may undergo repeat testing of the entire liver test panel within the same Screening window and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor.
3. Prior liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used
4. Evidence of any bleeding disorder not related to hemophilia A
5. Platelet count of < 100 x 10^9/L
6. Creatinine ≥ 1.5 mg/dL
7. Liver cirrhosis of any etiology as assessed by liver ultrasound

8. Chronic or active hepatitis B as evidenced by positive serology testing (hepatitis B surface antigen [HBsAg], hepatitis B surface antibody [HBsAb], and hepatitis B core antibody [HBcAb]) and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results in the Laboratory Manual.
9. Active Hepatitis C as evidenced by detectable HCV RNA, or currently on antiviral therapy
10. Active malignancy, except non-melanoma skin cancer
11. History of hepatic malignancy
12. History of arterial or venous thromboembolic events (eg, deep vein thrombosis, non-hemorrhagic stroke, pulmonary embolism, myocardial infarction, arterial embolus), with the exception of catheter-associated thrombosis for which anti-thrombotic treatment is not currently ongoing.
13. Known inherited or acquired thrombophilia, including conditions associated with increased thromboembolic risk, such as atrial fibrillation.
14. A history of known inflammatory, connective tissue, or autoimmune disorders (eg, vasculitis).
15. Treatment with any Investigational Product within 30 days or 5 half-lives of the investigational product (whichever is longer) prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study
16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including possible corticosteroid treatment outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.
17. Prior treatment with any vector or gene transfer agent
18. Major surgery planned in the 52-week period following the infusion with BMN 270
19. Use of systemic immunosuppressive agents, not including corticosteroids, or live vaccines within 30 days before the BMN 270 infusion
20. Concurrent enrollment in another clinical study, unless it is an observational (non-interventional) clinical study that does not interfere with the requirements of the current protocol or have the potential to impact the evaluation of safety and efficacy of BMN 270 and with prior consultation with the Medical Monitor
21. Known allergy or hypersensitivity to investigational product formulation
22. Unwilling to receive blood or blood products for treatment of an adverse event and/or a bleed

9.3.3 Removal of Subjects from Treatment or Assessment

Subjects may withdraw their consent to participate in the study at any time without prejudice. The Investigator must withdraw from the study any subject who requests to be withdrawn. Such subjects will always be asked about the reason(s) for withdrawal. The Investigator will discuss with the subject appropriate procedures for withdrawal from the study. The Investigator should ask the subject's consent to perform the procedures listed under the early termination visit. Should a subject withdraw from the study, all efforts will be made to complete and report the observations as thoroughly as possible up to the date of the withdrawal.

A subject's participation in the study may be discontinued at any time at the discretion of BioMarin or of the Investigator and in accordance with his/her clinical judgment. When possible, the tests and evaluations listed for the termination visit should be carried out and every effort will be made to gather follow-up safety data if possible.

BioMarin must be notified of all subject withdrawals as soon as possible. BioMarin also reserves the right to discontinue the study at any time for either clinical or administrative reasons and to discontinue participation by an individual Investigator or site for poor enrollment or noncompliance.

Reasons for which the Investigator or BioMarin may withdraw a subject from the study include, but are not limited to, the following:

- Subject requires medication or medical procedure prohibited by the protocol
- Subject was erroneously enrolled into the study or does not meet entry criteria and not yet been dosed with BMN 270; subjects who do not meet entry criteria but who erroneously receive BMN 270 should remain in the study for safety monitoring
- Subject is lost to follow-up

If a subject fails to return for scheduled visits, a documented effort must be made to determine the reason. If the subject cannot be reached by telephone, a certified letter should be sent to the subject requesting contact with the Investigator. This information should be recorded in the study records.

The Investigator or designee must explain to each subject, before enrollment into the study, that for evaluation of study results, the subject's protected health information obtained during the study may be shared with the study Sponsor, regulatory agencies, and IRB/EC It is the Investigator's (or designee's) responsibility to obtain written permission to use protected health information per country-specific regulations, such as the Health Insurance Portability

and Accountability Act (HIPAA) in the US, from each subject. If permission to use protected health information is withdrawn, it is the Investigator's responsibility to obtain a written request, to ensure that no further data will be collected from the subject and the subject will be removed from the study.

9.3.3.1 Study Safety Evaluation Criteria

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC evaluation of safety and efficacy in treated subjects with the following triggers for pausing further enrollment:

- any related SAE;
- an related AE with a severity > Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

Additionally, the DMC should consider whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction. Following a temporary halt of enrolment, the DMC may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

If a Grade 4 or Grade 5 adverse event assessed as related to study drug occurs in a study subject, further enrollment into the trial will be halted until a full safety review of the data by the DMC has taken place. Relevant reporting and discussion with the Sponsor and the DMC will take place before resumption of dosing.

If any of the following events occur in a subject in the study who has received BMN 270 infusion, further enrollment into the trial will be temporarily put on hold pending prompt evaluation by the DMC.

1. Liver dysfunction (criteria do not apply to ALT elevations with an extra-hepatic etiology):
    - ALT >5x ULN, for more than 2 weeks
    - ALT >3x ULN and (total bilirubin >2x ULN **or** INR >1.5)
    - ALT >3x ULN with signs and symptoms of liver dysfunction
2. The occurrence of an AE of hepatic failure.
3. The detection of high titer neutralizing antibodies (>5 BU) to hFVIII following BMN 270 infusion in two subjects.

4. The occurrence of any cancer (except non-melanoma skin cancer) at any point after BMN 270 infusion.
5. The occurrence of a thromboembolic event with FVIII activity > 150 IU/dL in one subject.

If any of the following events occurs in a subject in the study who has received BMN 270 infusion, a prompt evaluation by the DMC will be required. Further enrollment into the trial will continue while DMC evaluation is ongoing, unless deemed otherwise by the DMC.

1. Acute hypersensitivity assessed as related to BMN 270
2. The detection of high titer neutralizing antibodies (>5 BU) to hFVIII following BMN 270 infusion in one subject
3. Occurrence of a thromboembolic event

9.3.4 Subject Identification and Replacement of Subjects

Each subject will be assigned a unique subject identifier. This unique identifier will be on all eCRF pages. In legal jurisdictions where use of initials is not permitted, a substitute identifier will be used.

Subjects who withdraw from the study after receiving BMN 270 will not be replaced.

9.3.5 Duration of Subject Participation

The duration of participation for each subject will be approximately 264 weeks. This includes 4 weeks of screening, 1 day of BMN 270 infusion, 26 weeks of Post-Infusion Follow-Up, and 234 weeks of Long-Term Follow-Up.

9.4 Treatments

9.4.1 Treatments Administered

BioMarin and/or its designee will provide the study site with a supply of IP sufficient for the completion of the study. BioMarin is responsible for shipping study drug to clinical sites.

9.4.2 Identity of Investigational Product

9.4.2.1 Product Characteristics and Labeling

BMN 270 is a sterile, clear, colorless-to-pale yellow solution for IV infusion and is supplied in a 10 mL Crystal Zenith® (CZ) vial. Each CZ vial contains 8.5 mL (extractable volume 8 mL) of AAV5-hFVIII-SQ at a concentration of 2E13 vector genomes per mL in a pH 7.4 phosphate buffer.

The study drug is labelled according to the particulars approved by the relevant regulatory agencies.

9.4.3 Storage

At the study site, all IP must be stored under the conditions specified in the Pharmacy Manual in a secure area accessible only to the designated pharmacists and clinical site personnel. All IP must be stored and inventoried and the inventories must be carefully and accurately documented according to applicable state, federal and local regulations, ICH GCP, and study procedures.

9.4.4 Directions for Administration

On the day of infusion, the subject will come to the infusion site, where a physical examination will be performed by the Investigator or designee. If the subject is found to have an active acute illness at the time of planned infusion, then the infusion should be deferred until the illness has resolved; screening procedures may require repetition if outside the specified window. An IV catheter or butterfly needle will be inserted into a suitable peripheral vein (eg, the median cubital vein) and flushed with saline. FVIII replacement therapy will not be given since venipuncture is a minimally invasive procedure in these individuals under ordinary conditions.

BMN 270 will be prepared and infused as a pure solution over a dose-dependent time. Prepared drug will be kept at room temperature prior to administration. An electric syringe pump will be used to infuse through an in-line, low protein binding 0.22 micron filter. BMN 270 will be infused through the catheter using an appropriate infusion pump at an initial rate of 1 mL/min. The infusion rate should be increased every 30 minutes by 1 mL/min up to a maximum of 4 mL/min, provided that the subject's clinical condition permits such an increase. Of note, the IP has been shown to be stable at room temperature for approximately 10 hours following completion of product thaw. Vital signs (pulse, blood pressure, respiration rate and temperature) should be monitored at 15 minute (±5 minutes) intervals throughout the period of the infusion.

As with any infused biological product, there is a potential risk of acute, systemic hypersensitivity reactions (including anaphylaxis) with BMN 270. Dosing will be administered at a qualified infusion site, with appropriate resuscitation equipment and medication available and easily accessible.

Clinical staff administering BMN 270 should be trained appropriately in recognizing and managing the signs and symptoms associated with potential hypersensitivity, anaphylactic,

and anaphylactoid reactions. Additionally, the Investigator should be familiar with Sampson's criteria for defining anaphylaxis (Sampson, 2006; Appendix 1).

Should symptoms of potential hypersensitivity occur, the infusion may be slowed or halted at the Investigator's discretion, with consideration of the subject's clinical condition. If the infusion is halted, it should only be restarted if the Investigator considers it safe and appropriate to do so. Antihistamines, anti-pyretic, and/or corticosteroid administration is permitted prior to restarting an interrupted infusion by an infusion-related reaction. At the restart, the infusion rate may be adjusted (ie, to a slower rate [minimum of 1 mL/min], with the rate increased every 30 minutes by 1 mL/min up to a maximum rate of 4 mL/min, if the subject's clinical condition permits such an increase) with careful monitoring of the subject. In the event of an infusion rate reaction with more than one dosing interruption, the infusion rate would not go beyond 1 mL/min.

In case of hypersensitivity reactions, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.

Following completion of the infusion, vital signs will be monitored hourly (± 5 minutes). If the vital signs are stable the catheter will be removed 8 hours after the infusion. Hemostasis at the puncture site will be established by applying pressure according to standard protocol for infusing FVIII concentrates. Subjects will remain in the clinic for at least 24 hours to observe for any immediate toxicity of the procedure; in-patient observation can be extended beyond 24 hours if needed per Investigator discretion. After 24 hours, subjects will be discharged from the clinic unless toxicity has been observed in which case the stay in the clinic may be extended or the subject may transfer to a separate facility based on the evaluation and judgment of the Principal Investigator after consultation with the Medical Monitor.

Prior to discharging subjects from the clinic, the Investigator or designee should instruct subjects how to recognize signs and symptoms of potential (delayed) hypersensitivity

reactions and anaphylaxis, and to contact a medical practitioner or seek emergency care in case of such an event.

9.4.5 Method of Assigning Subjects to Treatment Groups

Subjects who meet all eligibility criteria (refer to Section 9.3.1 and Section 9.3.2) may be enrolled into the study. Approval by the Medical Monitor will be required prior to enrollment of each study subject. Upon their enrollment into the study, subjects will be assigned a unique subject number.

9.4.6 Selection of Dose Used in the Study

Data from an ongoing first in human study (270-201) indicates that following single escalated doses of BMN 270 (6E12, 2E13, 4E13, 6E13 vg/kg), dose-related increases in FVIII activity were observed, with concurrent improvements in bleeding episodes and exogenous FVIII utilization, particularly at the 4E13 and 6E13 vg/kg dose levels. At all dose levels, BMN 270 is considered to be well-tolerated with mild increases in ALT as the most common adverse event. Please refer to the IB for detailed efficacy and safety data. The 6E13 vg/kg dose has been selected for this study to maximize the likelihood of transduction in the face of pre-existing AAV5 antibodies.

9.4.6.1 Selection of Timing of Dose for Each Subject

9.4.7 Blinding

This is an open-label study.

9.4.8 Prior and Concomitant Medications

All prescription and over-the-counter medications (including dietary and herbal supplements) taken by a subject for 30 days before Screening will be recorded on the designated eCRF. The Investigator may prescribe additional medications, deemed necessary to provide adequate prophylactic or supportive care, during the study, as long as the prescribed medication is not prohibited by the protocol. In the event of an emergency, any needed medications may be prescribed without prior approval, but the Medical Monitor must be notified of the use of any contraindicated medications immediately thereafter. Any concomitant medications added or discontinued during the study should be recorded on the eCRF. Medications should, whenever possible, not be recorded in the electronic database with a frequency of as needed (PRN).

The following medications are prohibited starting 30 days before Screening and through the end of the study, and the Sponsor must be notified if a subject receives any of these during the study:

- Any investigational therapy
- Systemic immunosuppressive agents, except for corticosteroids
- Emicizumab
- Fitusiran
- Concizumab
- Efavirenz
- Lamivudine

Subjects who begin antiretroviral therapy following BMN 270 infusion and during their participation in 270-203 must have their antiretroviral medication regimen approved by the Investigator and Medical Monitor prior to starting antiretroviral treatment.

The following medications should be avoided, starting 30 days prior to and for at least 52 weeks after BMN 270 infusion and minimized throughout the remaining duration of the study.

- Alcohol
- Herbal and natural remedies and dietary supplements
- Medications which may be hepatotoxic
- Medications which may reduce or increase the plasma concentration of corticosteroids

Vaccines should also be avoided during this period, but in particular during the first 26 weeks unless clinically indicated.

The following medications should be avoided during oral corticosteroid therapy:

- Vaccines
- NSAIDs

9.4.8.1 Concomitant Hemophilia Treatments

Subjects on prophylactic FVIII therapy will discontinue their regular treatment regimen starting 4 weeks after the day of infusion and switch to an “on-demand” schedule. FVIII replacement therapy can always be taken as needed by the subject for treatment of an acute bleeding episode; the subject must carefully record his treatment and bleeding episodes in his

diary. Prophylactic FVIII can be used on a case-by-case basis and in consultation with the Medical Monitor to prevent bleeding in extenuating circumstances (eg, peri-operative).

In addition, information on FVIII usage and bleeding episodes by medical history will be collected from subjects for the 12-month period immediately preceding study enrollment.

9.4.8.2 Therapeutic Corticosteroid Treatment of Elevated Hepatic Transaminases

All subjects will be started on prophylactic corticosteroids starting on the day of infusion (Day 1). Table 9.1.6 provides an example of a possible prophylactic corticosteroid course, including taper and post-corticosteroid additional monitoring of FVIII activity, LTs, and hepatitis B/hepatitis C reactivation. Clinical judgment, weighting the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses. Discussions between the Investigator and Medical Monitor are advised for any questions or concerns.

Following initiation or completion of the prophylactic corticosteroid regimen, if ALT levels become increased (eg, ≥ ULN or ≥ 2x baseline value) and alternative etiologies have been ruled out, prompt institution of therapeutic or on-demand oral corticosteroids (prednisone or converted equivalent) should be considered after consultation with the Medical Monitor (refer to Table 9.7.6.3.2).

- Whenever possible, a confirmatory lab draw for ALT should be performed within 72 hours, along with FVIII activity, prior to initiating oral corticosteroids.
- Corticosteroids may be delayed if elevations in ALT are clearly not related to BMN 270 (eg, elevated in ALT with concurrent increase in CPK due to intensive exercise)

Therapeutic corticosteroid treatment should be initiated at a dose of 60 mg/day. At minimum, the recommended duration of therapeutic corticosteroids is 60 mg/day for 3 weeks, 40 mg/day for 4 weeks, and 30 mg/day for 4 weeks, followed by a gradual taper thereafter. Should a scenario arise in which a deviation from the minimum recommended dose and/or duration of therapeutic corticosteroids may be clinically indicated, a discussion should take place between the Investigator and Medical Monitor regarding corticosteroid dose adjustments. Tapering of corticosteroid dosages should be guided by the following:

Table 9.4.8.2.1: Adjustments to Corticosteroid Regimen

Corticosteroids should be tapered on an individual subject basis with the following guiding principles:	Corticosteroids may be tapered **if**: • ALT ≤ 1.5x baseline value; and • FVIII activity levels > 90% of the pre-decline FVIII activity levels; and • There is no concern for adrenal insufficiency post-withdrawal
Increasing Corticosteroid Dose	If ALT level is increasing or FVIII activity level is decreasing while on oral corticosteroids, any increases in oral corticosteroid dosing should be made only upon consultation with the Medical Monitor

For any scenarios that are not accounted for in the above table, a discussion should take place between the Investigator and Medical Monitor regarding corticosteroid dose adjustments.

After discontinuation of oral corticosteroids, labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values.

Following initiation or completion of therapeutic oral corticosteroids, if increased ALT levels (eg, ≥ ULN or ≥ 2x baseline value) are reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor. Modification of the corticosteroid regimen may take into consideration possible confounders for the ALT elevation and impact on FVIII expression.

Management and monitoring of reactions to corticosteroids should be determined by the Investigator's clinical judgment in consultation with the Sponsor's Medical Monitor. This includes the contraindicated use of NSAIDs during corticosteroid treatment and specific monitoring not already covered by the SoA. The use of COX-2 inhibitors, while not contraindicated during corticosteroid treatment, should be limited, if possible. Practical management to prevent complications related to oral corticosteroid therapy may be undertaken at the discretion of the Investigator (eg, evaluation of glucose intolerance, hyperlipidemia etc.). Hepatitis B status and HCV viral load will be rechecked 6 weeks after the start of oral corticosteroid treatment and then 1 week and 13 weeks after the completion of oral corticosteroid treatment in subjects with a history of hepatitis B or hepatitis C. All adverse events (including any adverse events suspected to be caused by or related to corticosteroid use) should be reported as outlined in Section 10 of the protocol.

Subjects on corticosteroids should receive appropriate counselling and support regarding side effects from the Investigator or the treating institution (eg, listings of side effects and when to notify carers, wallet card for emergencies if on steroids, etc.).

9.4.9 Treatment Compliance

Study drug will be administered to subjects at the dosing facility by a qualified health care professional. The quantity dispensed, returned, used, lost, etc. must be recorded on a dispensing log. Sites will be instructed to return or destroy all used and unused study drug containers.

9.5 Investigational Product Accountability

The Investigator or designee is responsible for maintaining accurate records (including dates and quantities) of IP(s) received and IP lost or accidentally or deliberately destroyed. The Investigator or designee must retain all unused or expired study supplies until the study monitor (on-site CRA) has confirmed the accountability data, if allowed by local SOPs.

9.5.1 Return and Disposition of Clinical Supplies

Unused study drug must be kept in a secure location for accountability and reconciliation by the study monitor. The Investigator or designee must provide an explanation for any destroyed or missing study drug or study materials (or must be referenced in their institution SOPs).

Unused study drug may be destroyed on site, per the site's standard operating procedures, but only after BioMarin has granted approval for drug destruction. The monitor must account for all study drug in a formal reconciliation process prior to study drug destruction. All study drug destroyed on site must be documented. Documentation must be provided to BioMarin or designee and retained in the Investigator study files. If a site is unable to destroy study drug appropriately, the site can return unused study drug to BioMarin upon request. The return of study drug or study drug materials must be accounted for on a Study Drug Return Form provided by BioMarin.

All study drug and related materials should be stored, inventoried, reconciled, and destroyed or returned according to applicable state and federal regulations and study procedures. For additional information, please refer to the Study Pharmacy Manual.

9.6 Dietary or Other Protocol Restrictions

There are no dietary or other protocol restrictions for this study. Alcohol should be avoided starting 30 days prior to and for at least the first 52 weeks of the study, and particularly within 48 hours prior to lab work.

Subjects should be advised to abstain from any blood, organ, or sperm donation after BMN 270 infusion, until there is no further evidence of vector shedding from PCR analysis of samples.

9.7 Efficacy and Safety Variables

9.7.1 Efficacy and Safety Measurements Assessed

The SoA (Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5) describe the timing of required evaluations.

9.7.2 Efficacy Variables

9.7.2.1 FVIII Activity

Efficacy (response to treatment) will be defined as FVIII activity ≥ 5 IU/dL at Week 26 following BMN 270 infusion.

Values for FVIII activity will be excluded from analysis if obtained within 72 hours since the last infusion of exogenous FVIII protein concentrates. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; The 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy.

In the event of an FVIII activity level decline during the study:

- If FVIII activity has declined at least 20% from the peak but less than 35% and has declined for at least 2 consecutive assessments, FVIII activity and LTs should be repeated every 7 days until FVIII activity is stable or increasing
- If FVIII activity has declined >35% from the peak and has declined for at least 2 consecutive assessments, FVIII activity and LTs should be repeated every 72 hours until FVIII activity is stable or increasing.

Note that fluctuations in FVIII activity are common, and if no clear trend indicating a decline in FVIII activity is observed, then this additional testing may be deferred (upon consultation between the Investigator and the Medical Monitor) until either a more clear trend of decline has been demonstrated or until the FVIII activity levels stabilize or increase.

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after

discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 52, as applicable and at the discretion of the Investigator.

Details on collecting FVIII activity samples are included in the Laboratory Manual.

9.7.2.2 Factor VIII Replacement Therapy/Bleeding Episodes

Additional efficacy variables are:

- Change of annualized utilization (IU/kg) of exogenous FVIII replacement therapy during Week 5 to Week 26 post BMN 270 infusion from the baseline utilization of exogenous FVIII replacement therapy.
- Change of ABRs requiring exogenous FVIII replacement treatment during Week 5 to Week 26 of the study post BMN 270 infusion from the baseline ABR.

During the study, subjects will be asked at each study visit to report the use of factor replacement therapy and the number of bleeding episodes since the previous visit. This information will be captured on the subject's diary or other subject records.

Subjects are strongly encouraged to immediately consult Investigator for guidance regarding exogenous FVIII administration for suspected bleeds or bleeding episodes within the first 6 weeks post BMN 270 infusion.

In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis.

9.7.2.3 Patient-Reported Outcomes (PRO)

The Haemo-QoL-A questionnaire is a validated hemophilia-specific health-related quality of life questionnaire for adults (Rentz, 2008). It consists of 41 questions covering six domains (Physical Functioning, Role Functioning, Worry, Consequences of Bleeding, Emotional Impact and Treatment Concerns). Items are answered on a 6-point Likert-type scale, ranging from 0 (None of the time) to 5 (All of the time). Higher scores mean better health-related quality of life or less impairment for a particular subscale (Haemo-QoL Study Group, 2017). Details regarding the Haemo-QoL-A assessment will be included in the Study Reference Manual.

The EQ-5D-5L instrument is a self-reported questionnaire designed to measure general health status (The EuroQol Group, 1990) (Brooks, 1996). The EQ-5D-5L is composed of 2-parts: a descriptive system that assesses 5 levels of perceived problems (mobility, self-care, usual activities, pain/discomfort, and anxiety/depression) in 5 dimensions and the EQ visual analogue scale (EQ VAS) assessment for overall health. A sample copy of the EQ-5D-5L and additional information are provided in the Study Reference Manual.

The Haemophilia Activities List (HAL) measures the impact of hemophilia on self-perceived functional abilities in adults (van Genderen, 2006). The instrument consists of multiple domains including lying/sitting/kneeling/standing, leg and arm function, use of transportation, self-care, household tasks, and leisure activities where subjects are asked to rate their level of difficulty with activities of daily living on a 6-point Likert-type scale from 1 (Impossible) to 6 (Never). For some items, subjects are given the choice to answer 'Not applicable'. A sample copy of the HAL and additional information are provided in the Study Reference Manual.

The Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) instrument is designed to measure the effect of disease symptom severity on work productivity and classroom productivity (if applicable) (Recht, 2014). The WPAI+CIQ:HS questionnaire yields scores related to work/classroom absenteeism, reduced on-the-job effectiveness, overall work/classroom impairment, and activity impairment. WPAI+CIQ:HS outcomes are expressed as impairment percentages, with higher numbers indicating greater impairment and less productivity (Reilly, 2002). A sample copy of the WPAI+CIQ:HS and additional information are provided in the Study Reference Manual.

9.7.3 Immunogenicity

Immunogenicity assays will be performed on plasma and PBMCs. The assays will include detection of anti-AAV5 vector capsid and anti-FVIII total antibodies, as well as determination of neutralizing antibodies against FVIII (FVIII inhibitors) and against the AAV5 vector capsid (Transduction Inhibitors, TI). FVIII Inhibitors will be assessed using the Bethesda assay with Nijmegen modification. Any abnormality of the liver parameters will lead to a retrospective immunogenicity assessment to evaluate FVIII-and vector capsid-specific cellular immunogenicity. FVIII- and vector capsid-specific cellular immunity will be assessed by stimulated cytokine secretion using an ELISpot assay performed on collected PBMCs.

9.7.4 Pharmacodynamics

The FVIII protein concentration and activity level as measured by a validated immunoassay and by a validated FVIII activity assay, respectively, will be used for plasma profiles; FVIII protein and activity will be used to determine PD parameters.

9.7.5 Exploratory Assessments

A cytokine bead array assay assessment will be performed at Baseline and then weekly through Week 26.

In addition, blood samples will be collected from subjects at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5 to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV5 gene transfer, and to develop assays used for these evaluations. Subject may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A.

All biomarker samples collected in this study may be used for exploratory biomarker research, including evaluation of additional biomarkers not specifically listed in the protocol. In addition, samples collected for other purposes in this study may be used for exploratory research once testing for the primary purpose has been completed.

9.7.6 Safety Variables

Safety in this study will be determined from evaluation of AEs, clinical laboratory assessments with a particular attention to the liver function, vital signs assessments, physical examinations, and immunogenicity.

9.7.6.1 Adverse Events

The determination, evaluation and reporting of AEs will be performed as outlined in Section 10.

9.7.6.2 Clinical Laboratory Assessments

The scheduled clinical laboratory tests are listed in Table 9.7.6.2.1. Refer to the Study Reference Manual for instructions on obtaining and shipping samples. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample.

Any abnormal test results determined to be clinically significant by the Investigator should be repeated (at the Investigator's discretion) until: (1) the cause of the abnormality is

determined; (2) the value returns to baseline or to within normal limits; or (3) the Investigator determines that the abnormal value is no longer clinically significant.

All abnormal clinical laboratory results should be initialed and dated by an Investigator, along with a comment regarding whether or not the result is clinically significant. Each clinically significant laboratory result should be recorded as an adverse event.

The diagnosis, if known, associated with abnormalities in clinical laboratory tests that are considered clinically significant by the Investigator will be recorded on the AE eCRF.

Table 9.7.6.2.1: Clinical Laboratory Tests

Blood Chemistry	Hematology	Urine Tests	Coagulation Screen including:
Albumin	Hemoglobin	Appearance	APTT
BUN	Hematocrit	Color	PT/INR
Calcium	WBC count	pH	TT
Chloride	RBC count	Specific gravity	
Total cholesterol	Platelet count	Ketones	
CPK	Differential cell count	Protein	
Creatinine	RBC indices (MCV and MCH)	Glucose	
CRP	ABO blood typing*	Bilirubin	
Glucose Phosphorus		Nitrite Urobilinogen	
Potassium		Hemoglobin	
Total protein			
Sodium			
Uric Acid			

BUN, blood urea nitrogen; CPK, creatinine phosphokinase; CRP, C-reactive protein; PT, prothrombin time; APTT, activated partial thromboplastin time; RBC, red blood cell; WBC, white blood cell; TT, thrombin time; INR, international normalized ratio; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin.

* ABO blood typing assessment should be performed at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study.

In addition to scheduled clinical laboratory assessments, a fasting blood lipid panel (including triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) will be assessed at the BMN 270 infusion visit. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit.

In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected

within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.

At applicable sites, certain study assessments may be performed by a mobile nursing (MN) professional at the patient's home or another suitable location, such as their school or office, to improve access and convenience for patients participating in the study. The Sponsor may select a healthcare company that will be responsible for providing MN services for participating sites (the MN vendor). The MN vendor is responsible for ensuring that all MN professionals are licensed, qualified, and in good standing, as per applicable regulations, and that appropriate background checks have been performed. If the investigator at a participating site determines that MN services are appropriate for a patient and the patient gives written informed consent to participate in MN visits, the MN network will communicate with the patient and the patient’s site. Potential MN visits are designated in the Schedules of Events.

9.7.6.3 Liver and Hepatitis Testing

Subjects will be screened for evidence of previous or active hepatitis B or hepatitis C infection at Screening; hepatitis B screening should include HBsAg, HBsAb, and HBcAb. Subjects with documented results showing an absence of active hepatitis B or hepatitis C infection (as measured by negative surface antigen or DNA for hepatitis B or negative RNA testing for hepatitis C) 30 days prior to providing signed informed consent do not need to repeat those tests during the screening period.

Evidence of ongoing hepatitis B or hepatitis C infection is exclusionary. Subjects with history of hepatitis B or hepatitis C infection prior to study entry will be tested for hepatitis B and hepatitis C reactivation at Week 16. Subjects with a history of hepatitis B or hepatitis C will be asked for information about the treatments received as part of their medical history assessment at Screening.

Subjects with a previous history of hepatitis B or hepatitis C who receive therapeutic oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation

assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.6.

A liver ultrasound and liver tests (LTs) during Screening will identify any significant hepatic dysfunction.

Liver tests will be monitored on a regular basis; at each time point specified in the SoA, the following LTs should be assessed:

Table 9.7.6.3.1: Liver Tests

Liver Tests (LTs)			
Alkaline Phosphatase	AST (SGOT)	Total Bilirubin	LDH
ALT (SGPT)	Direct Bilirubin	GGT	

ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma-glutamyltransferase; LDH, lactate dehydrogenase; SGOT, serum glutamic-oxaloacetic transaminase; SGPT, serum glutamic-pyruvic transaminase

Elevated ALT levels should be evaluated according to the following plan:

Table 9.7.6.3.2: Evaluation of ALT Elevations

ALT Level	Work-Up
≥1.5x Baseline - <2x Baseline	• Continue to monitor LTs and FVIII per protocol (repeat within 24-72 hours if next protocol scheduled visit is >24-72 hours from the time of the reported ALT elevation) • Consider evaluation to rule out alternative etiology (eg, concomitant medications, viral or autoimmune hepatitis, alcohol use, recreational drug use, special diets, strenuous exercise, prior and/or concurrent illnesses, exposure to environmental and/or industrial chemicals, etc.) (refer to Table 9.7.6.3.3) • If ALT is > ULN or > 2x baseline in 2 consecutive assessments within 24-72 hours and alternative etiologies have been ruled out, start oral corticosteroids upon consultation with the Medical Monitor (refer to Section 9.4.8.2)
≥2x Baseline or ≥ ULN - <3x ULN	• Repeat LTs and FVIII within 24-72 hours • Continue to monitor LTs weekly until ALT is stable or improving • Evaluate and rule out alternative etiologies (as above) • Consult with Medical Monitor • If ALT is ≥ 2x baseline or ≥ ULN - < 3x ULN in 2 consecutive assessments within 72 hours and alternative etiologies have been ruled out, start oral corticosteroids upon consultation with the Medical Monitor (refer to Section 9.4.8.2) • Obtain other possibly relevant laboratory evaluations (albumin, PT/INR, CRP, etc.) • Obtain complete blood count with differential to assess for eosinophilia • Obtain PBMC to evaluate potential immune response (prior to starting oral corticosteroids) • If no improvement in 14 days, consider gastroenterology and/or hepatology consult, abdominal workup, imaging (including MRI or ultrasound), and/or liver biopsy as appropriate
≥3x ULN	• Consult with Medical Monitor • Evaluate and rule out alternative etiologies (as above) • Repeat LTs and FVIII within 24-48 hours, and continue with monitoring of LTs at least twice weekly for as long as the subject's ALT remains ≥ 3x ULN • If ≥3x ULN in 2 consecutive assessments within 48 hours, start oral corticosteroids (refer to Section 9.4.8.2) • Obtain other possibly relevant laboratory evaluations (albumin, PT/INR, CRP, etc.) • Obtain complete blood count with differential to assess for eosinophilia • Obtain PBMC to evaluate potential immune response (prior to starting oral corticosteroids) • If no improvement in 14 days, consider gastroenterology and/or hepatology consult, abdominal workup, imaging (including MRI or ultrasound), and/or liver biopsy as appropriate

When ruling out alternative viral or autoimmune hepatitis as part of the elevated ALT workup, the following tests should be performed:

Table 9.7.6.3.3: Viral and Autoimmune Hepatitis Testing

Viral Hepatitis Workup Testing	Autoimmune Hepatitis Workup Testing
Hepatitis A	Smooth muscle antibody
Hepatitis B	Mitochondrial antibody
Hepatitis C	Liver/kidney microsomal antibodies
Hepatitis E	Antinuclear antibody (ANA) HEP-2
Cytomegalovirus (CMV)	
Epstein-Barr virus (EBV)	
Herpes simplex virus (HSV) 1 & 2	

9.7.6.4 HIV Testing

HIV testing will be performed at Screening. Subjects with documented negative results within the last 30 days prior to screening do not need to be retested.

9.7.6.5 Vital Signs, Physical Examinations and Other Observations Related to Safety

Vital signs will include seated systolic and diastolic blood pressure, heart rate, respiration rate, and temperature. Any clinically significant change in vital signs will be recorded as an AE.

Systolic blood pressure, diastolic blood pressure, heart rate, respiration rate, and temperature will be assessed at Screening, Baseline, and at the beginning of each visit during the Post-Infusion Follow-Up and Long-Term Follow-Up periods. On the day of the BMN 270 Infusion, vital signs will be monitored prior to infusion, during the infusion every 15 minutes (± 5 minutes), following the infusion hourly (± 5 minutes) for at least 8 hours during the subject's stay in the clinic. Any abnormal vital sign assessments should be repeated, and both values should be recorded in the eCRF.

A complete physical examination should be performed at Screening, Week 26, Week 52, and at the End of Year visits. A complete physical examination will include general appearance (head, eyes, ears, nose, and throat), cardiovascular, dermatologic, lymphatic, respiratory, gastrointestinal, genitourinary, musculoskeletal, and neurologic systems.

At other visits, brief physical examinations may be performed at the discretion of the Investigator based on the subject's clinical condition. A brief physical examination will include general appearance, cardiovascular, dermatologic, respiratory, gastrointestinal, musculoskeletal, and neurologic assessments. Particular attention should be given to signs of bleeding, as well as assessing possible hemarthroses.

Height will be recorded at Screening only. Weight will be recorded at Screening and then every 4 weeks thereafter through Week 20, at Week 26, every 4 weeks through Week 52, and then at the second Q12W visit each year and at every End of Years visit during Years 2-5.

At visits where the MN services are used or shortened lab draw-only visits are conducted at the sites, the physical examination and vital signs assessments indicated in the Schedule of Events will not be performed.

9.7.6.6 Vector Shedding

During the Post-Infusion Follow-Up period, subjects will undergo testing of various bodily samples to look for evidence of vector shedding for possible viral transmission. Bodily fluids will be tested by polymerase chain reaction (PCR). Fluids tested will include:

- Blood
- Saliva
- Semen
- Urine
- Stool

Vector shedding will also be extensively studied in the present clinical trial, at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5. Testing will continue until at least 3 consecutive results below the limit of detection are obtained. Testing of semen will continue at least through Week 12, even if 3 consecutive results below the limit of detection have been recorded in that compartment prior to that time point. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 26 should continue to have PCR testing in semen every 4 weeks until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor). Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule (refer to Section 12.5.3) but who have not cleared vector shedding from all fluids must still provide samples for assessment until vector shedding has cleared: at Weeks 32, 36, 40, 44, 48, and 52 (during Year 1) and every 4 weeks during Years 2-5. Such subjects may provide samples on the designated study visit dates either at the sites or through use of a MN professional.

Samples may be fractionated prior to shedding analysis in order to better characterize the presence, structure, and location of vector DNA and/or vector capsid within each matrix. If needed, the fractionation may be performed with samples collected specifically for shedding analysis (saliva, blood, semen, urine, feces). Alternatively, the vector DNA characterization

during shedding analysis may utilize already fractionated exploratory samples obtained from the above biofluids, such as exploratory plasma samples, exploratory PBMC samples, and red blood cells recovered during PBMC/plasma isolations.

Fractionation of semen to collect purified sperm separately from non-sperm cells may be performed in parallel at any visit where semen samples are collected. The shedding analysis of a fractionated semen sample will only be performed if vector DNA was detected in the whole semen sample for the same visit. Fractionation of semen during shedding analysis may be stopped if purified sperm tested positive for vector DNA on at least three visits, or if purified sperm tested below the limit of detection for vector DNA on at least three consecutive visits.

Contraception use may need to be extended beyond 12 weeks in individual subjects based on observed vector shedding in semen. After 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples below the limit of detection (upon consultation between the Investigator and Medical Monitor).

Details for sample collection and storage are provided in the Laboratory Manual.

10 REPORTING ADVERSE EVENTS

10.1 Safety Parameters and Definitions

Safety assessments will consist of monitoring and recording protocol-defined AEs and SAEs; measurement of protocol-specified hematology, clinical chemistry, and urinalysis variables; measurement of protocol-specified vital signs; and other protocol-defined events of special interest that are deemed critical to the safety evaluation of the study drug.

10.1.1 Adverse Events

For this protocol, an adverse event (AE) is any untoward medical occurrence in a subject, temporally associated with the use of study intervention, whether or not considered related to the study intervention.

An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of study intervention.

Events meeting the AE definition include:

- Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition.
- New conditions detected or diagnosed after study intervention administration even though it may have been present before the start of the study.
- Signs, symptoms, or the clinical sequelae of a suspected drug-drug interaction.

Events not meeting the AE definition include:

- Any clinically significant abnormal laboratory findings or other abnormal safety assessments which are associated with the underlying disease, unless judged by the investigator to be more severe than expected for the subject's condition.
- The disease/disorder being studied or expected progression, signs, or symptoms of the disease/disorder being studied, unless more severe than expected for the subject's condition.
- Medical or surgical procedure (eg, endoscopy, appendectomy): the condition that leads to the procedure is the AE.
- Situations in which an untoward medical occurrence did not occur (social and/or convenience admission to a hospital).
- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

10.1.1.1 Bleeding and Suspected Bleeding Events

All bleeding events and suspected bleeding events, regardless of the need for exogenous FVIII therapy as treatment, should be captured in subject diaries and recorded on the designated bleeding eCRF. Bleeding events and suspected bleeding events should not be reported as adverse events, with the following exception:

- All bleeding events and suspected bleeding events which meet one or more of the criteria for being serious (refer to Section 10.1) should be reported as serious adverse events (whether or not they are bleeding events that are normal sequelae of hemophilia, and whether or not they required exogenous FVIII as treatment).

10.2 Serious Adverse Events

A serious adverse event (SAE) is any untoward medical occurrence that, at any dose:

- Results in death
- Is life-threatening

  Note: Life-threatening refers to an event in which the subject was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.
- Requires inpatient hospitalization or prolongation of existing hospitalization
    - Note: In general, hospitalization signifies that the subject has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization, the event is serious.

  Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE.
- Results in persistent or significant disability or incapacity
- Is a congenital anomaly or birth defect in the child or fetus of a subject exposed to IP prior to conception or during pregnancy
- Is an important medical event or reaction – that, based on medical judgment, may jeopardize the subject or require medical/surgical intervention to prevent one of the other outcomes listed above (eg, anaphylaxis)

10.2.1 Events of Special Interest (EOSI)

The following EOSI need to be reported to the Sponsor within 24 hours of site awareness, irrespective of seriousness, severity or causality:

- Elevation of ALT ≥1.5x ULN or ALT > ULN & >2x baseline value, regardless of whether that elevation triggers an initiation or modification of oral corticosteroid treatment
- Thromboembolic event
- Systemic hypersensitivity, anaphylactic, or anaphylactoid reactions (refer to Appendix 1)

10.3 Methods and Timing for Capturing and Assessing Safety Parameters

10.3.1 Adverse Event Reporting Period

The study AE reporting period is as follows: After informed consent but prior to initiation of study drug, only SAEs associated with any protocol-imposed interventions will be collected. After informed consent is obtained and following infusion of study drug, the reporting period for all non-serious AEs and SAEs begins and continues for approximately 5 years or until study discontinuation/termination, whichever is longer. The criteria for determining, and the reporting of SAEs is provided in Section 10.1.

10.3.2 Eliciting Adverse Events

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and non-leading verbal questioning of the subject is the preferred method to inquire about AE occurrences. The Investigator will record all relevant AE/SAE/EOSI information in the subject's medical record and AE Case Report Form (eCRF).

10.3.3 Assessment of Seriousness, Severity, and Causality

The Investigator responsible for the care of the subject or medically qualified designee will assess AEs for severity, relationship to study drug and/or corticosteroids, and seriousness (refer to Section 10.2 for SAE definitions). These assessments must be made by a study clinician with the training and authority to make a diagnosis (eg, MD/DO, physician's assistant, nurse practitioner, or DDS).

10.3.3.1 Seriousness

The Investigator will assess if an AE should be classified as "serious" based on the seriousness criteria enumerated in Section 10.2. Seriousness serves as a guide for defining regulatory reporting obligations.

10.3.3.2 Severity

Severity (as in mild, moderate, or severe headache) is not equivalent to seriousness, which is based on subject/event outcome or action criteria usually associated with events that pose a threat to a subject's life or functioning. The Investigator will determine the severity of each AE, SAE and EOSI using the NCI CTCAE v4.03. Adverse events that do not have a corresponding CTCAE term will be assessed according to the general guidelines for grading used in the CTCAE v4.03 as stated in Table 10.3.3.2.1.

Table 10.3.3.2.1: Adverse Event Grading (Severity) Scale

Grade	Description	
1	Mild: asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated	
2	Moderate: minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living (ADL)[a]	
3	Severe or medically significant but not immediately life-threatening: hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL[b]	
4	Life threatening consequences; urgent intervention indicated	Grade 4 and 5 AEs should always be reported as SAEs
5	Death related to AE	

[a] Instrumental ADL refer to the following examples: preparing meals, shopping for groceries or clothes, using the telephone, managing money.

[b] Self-care ADL refer to the following examples: bathing, dressing and undressing, feeding oneself, using the toilet, taking medications, not bedridden.

10.3.3.3 Causality

The Investigator will determine the relationship of an AE to the study drug and/or corticosteroids and will record it on the source documents and AE eCRF. To ensure consistency of causality assessments, Investigators should apply the guidance in Table 10.3.3.3.1.

Table 10.3.3.3.1: Causality Attribution Guidance

Relationship	Description
Not Related	• Exposure to the IP and/or corticosteroids has not occurred **OR** • The administration of the IP and/or corticosteroids and the occurrence of the AE are not reasonably related in time **OR** • The AE is considered likely to be related to an etiology other than the use of the IP and/or corticosteroids; that is, there are no facts, evidence, or arguments to suggest a causal relationship to the IP and/or corticosteroids.
Related	• The administration of the IP and/or corticosteroids and the occurrence of the AE are reasonably related in time **AND** • The AE could possibly be explained by factors or causes other than exposure to the IP and/or corticosteroids **OR** • The administration of IP and/or corticosteroids and the occurrence of the AE are reasonably related in time **AND** • The AE is more likely explained by exposure to the IP and/or corticosteroids than by other factors or causes.

Factors suggestive of a causal relationship could include (but are not limited to):

- Plausible temporal relationship
- Absence of alternative explanations
- Rarity of event in a given patient or disease state
- Absence of event prior to study drug and/or corticosteroid exposure
- Consistency with study product pharmacology
- Known relationship to underlying mechanism of study drug and/or corticosteroid action
- Similarity to adverse reactions seen with related drug products

- Abatement of AE with discontinuation of study drug and/or corticosteroids, and/or recurrence of AE with reintroduction of study drug and/or corticosteroids

The Investigator's assessment of causality for individual AE reports is part of the study documentation process. Regardless of the Investigator's assessment of causality for individual AE reports, the Sponsor will promptly evaluate all reported SAEs against cumulative product experience to identify and expeditiously communicate possible new safety findings to Investigators and applicable regulatory authorities.

10.4 Procedures for Recording Adverse Events

10.4.1 Recording Adverse Events on a eCRF

Investigators should use precise medical terminology when recording AEs or SAEs on the AE eCRF. Avoid colloquialisms and abbreviations.

Record only one diagnosis, sign, or symptom per event field on the AE eCRF (eg, nausea and vomiting should not be recorded in the same entry, but as 2 separate entries).

In order to classify AEs and diseases, preferred terms will be assigned by the Sponsor to the original terms entered on the AE eCRF, using MedDRA (Medical Dictionary for Regulatory Activities) terminology.

10.4.1.1 Diagnosis versus Signs and Symptoms

The Investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE. Using accepted medical terminology, enter the diagnosis (if known). If not known, enter sign(s) and/or symptom(s). If a diagnosis subsequently becomes available, then this diagnosis should be entered on the AE (or SAE, as appropriate) eCRF, replacing the original entries where appropriate.

10.4.1.2 Adverse Events Occurring Secondary to Other Events

In general, AEs occurring secondary to other events (eg, cascade events) should be identified by their primary cause. For example, if severe diarrhea is known to have resulted in dehydration, it is sufficient to record only diarrhea as an AE or SAE on the AE eCRF. However, medically important events that may be linked and/or separated in time should be recorded as independent events on the AE eCRF. For example, if severe hemorrhage leads to renal failure, both events should be recorded separately on the AE eCRF.

10.4.1.3 Persistent or Recurrent Adverse Events

A persistent AE (duration of adverse event > 7 days) is one that extends continuously, without resolution, between subject evaluation time points. Events that change in severity necessitate the recording of an additional AE. AEs that do not have a change in severity should be recorded only once on the eCRF.

A recurrent AE is one that occurs and resolves between subject evaluation time points, but then subsequently recurs. All recurrences of the AE should be recorded on the AE eCRF. For example, if a subject has an adverse event of ALT increased that subsequently resolves, but the subject's ALT increases again, that should be reported as two adverse events – the initial ALT increase, and the second ALT increase.

10.4.1.4 Abnormal Laboratory Values

Laboratory test results (including any local FVIII activity or liver test results) will be recorded on the laboratory results pages of the eCRF, or appear on electronically produced laboratory reports submitted directly from the central laboratory, if applicable.

Any laboratory result abnormality fulfilling the criteria for a SAE or EOSI should be reported as such, and recorded in the AE eCRF unless associated with an AE that has already been reported.

Any laboratory result abnormality of CTCAE Grade 4 or 5 should be recorded as an SAE in the AE eCRF.

A clinical laboratory abnormality is considered clinically significant and should be documented as an AE if not refuted by a repeat test to confirm the abnormality and **any** one or more of the following conditions is met:

- Accompanied by clinical symptoms
- Requiring a change in concomitant therapy (eg, addition of, interruption of, discontinuation of, or any other change in a concomitant medication, therapy or treatment).
- The abnormality suggests a disease and/or organ toxicity
- The abnormality is of a degree that requires active management (eg, change of dose, discontinuation of study drug, more frequent follow-up assessments, further diagnostic investigation, etc.)

This applies to any protocol and non-protocol specified safety and efficacy laboratory result from tests performed after the first dose of study medication that falls outside the laboratory reference range and meets the clinical significance criteria.

This does not apply to any abnormal laboratory result that falls outside the laboratory reference range but that does not meet the clinical significance criteria (these will be analyzed and reported as laboratory abnormalities), those that are considered AEs of the type explicitly exempted by the protocol, or those which are a result of an AE that has already been reported.

For purposes of this study, laboratory tests showing a decreased level of FVIII activity should not be reported as adverse events unless there is an impact to clinical outcomes (eg, increased rate of bleeding, worsening of joint disease).

10.4.1.5 Pre-existing Conditions

A pre-existing condition is one that is present prior to administration of BMN 270. Such conditions should be recorded as medical history on the appropriate eCRF.

A pre-existing condition should be recorded as an AE or SAE during the study **only** if the frequency, intensity, or character of the condition worsens during the study period. It is important to convey the concept that a pre-existing condition has changed by including applicable language in the verbatim description of the event (eg, *more frequent* headaches).

10.4.1.6 General Physical Examination Findings

At screening, any clinically significant abnormality should be recorded as a pre-existing condition (refer to Section 10.4.1.5). During the study, any new clinically significant findings and/or abnormalities discovered on physical examination that meet the definition of an AE (or an SAE) must be recorded and documented as an AE or SAE on the AE eCRF.

10.4.1.7 Hospitalization, Prolonged Hospitalization, or Surgery

Any AE that results in hospitalization or prolonged hospitalization should be documented and reported as an SAE unless specifically instructed otherwise in this protocol (refer to Section 10.2).

There are some hospitalization scenarios that do not require reporting as an SAE when there is no occurrence of an AE. These scenarios include planned hospitalizations or prolonged hospitalizations to:

- Perform a protocol-mandated efficacy measurement
- Undergo a diagnostic or elective surgical procedure for a pre-existing medical condition that has not worsened
- Insert an in-dwelling IV catheter (such as a Port-a-Cath or other brand, if applicable) for administration of study drug or FVIII replacement therapy

- Receive scheduled therapy (study drug or otherwise) for the study indication

10.4.1.8 Deaths

All deaths that occur during the AE reporting period (refer to Section 10.3.1), regardless of attribution, will be recorded on the AE eCRF and expeditiously reported to the Sponsor as an SAE.

When recording a death, the event or condition that caused or contributed to the fatal outcome should be recorded as the single medical concept on the eCRF. If the cause of death is unknown and cannot be ascertained at the time of reporting, record "Unexplained Death" or "Death of Unknown Cause" on the AE eCRF.

10.4.1.9 Pregnancy

Although not an AE per se, pregnancy in the partner of a subject taking trial medication should be reported expeditiously to the Sponsor to facilitate outcome monitoring by the Sponsor. Pregnancy in partner should be reported during the period up to 5 years after viral infusion.

Pregnancy in a partner should be reported within 24 hours of the site becoming aware of the pregnancy by entering the information on the Pregnancy eCRF and submitting to BPV within 24 hours of the site becoming aware of the event. The Investigator must make every effort to follow the subject's partner (with that partner's consent) through resolution of the pregnancy (delivery or termination) and to report the resolution on the Pregnancy Follow-up eCRF. In the event of pregnancy in the partner of a study subject, the Investigator should make every reasonable attempt to obtain the woman's consent for release of protected health information.

Abortion, whether therapeutic or spontaneous, should always be classified as an SAE (as the Sponsor considers these to be medically significant), recorded on the AE eCRF, and expeditiously reported to the Sponsor as an SAE.

10.5 Reporting Requirements

10.5.1 Expedited Reporting Requirements

All SAEs and EOSI that occur during the course of the AE Reporting Period (refer to Section 10.3.1), whether or not considered related to study drug, must be reported by entering the information in the AE eCRF and submitting to BPV within 24 hours of the site becoming aware of the event. Investigators should not wait to collect information that fully documents the event before notifying BPV of an SAE. BioMarin may be required to report certain SAEs to regulatory authorities within 7 calendar days of being notified about the event;

therefore, it is important that Investigators submit any information requested by BioMarin as soon as it becomes available. IND safety reports will be submitted within 7 calendar days for unexpected fatal or life-threatening unexpected suspected adverse reactions (SUSARs) and within 15 calendar days for other non-life-threatening SUSARs.

The Sponsor is responsible for identifying, preparing and reporting all SUSARs to the relevant competent authorities, ethics committees and Investigators in accordance with the requirements identified in the Clinical Trials Regulations.

If the EDC is unavailable, all SAEs should be reported to BPV by completing the SAE Report Form and faxing or emailing the completed form to BPV within 24 hours of the site becoming aware of the event. Once the EDC is available, the information should be entered in the AE eCRF.

10.5.2 Institutional Review Board or Independent Ethics Committee Reporting Requirements

Reporting of SAEs to the Independent Ethics Committee (EC) or Institutional Review Board (IRB) will be done in compliance with the standard operating procedures and policies of the IRB/EC and with applicable regulatory requirements. Adequate documentation must be obtained by BioMarin showing that the IRB/EC was properly and promptly notified as required.

10.6 Follow-up of Subjects after Adverse Events

After the initial AE/SAE/EOSI report, the Investigator is required to proactively follow each subject at subsequent visits/contacts. All AEs/SAEs/EOSI will be followed until resolution, stabilization, the event is otherwise explained, or the subject is lost to follow-up. Resolution of AEs/SAEs/EOSI (with dates) should be documented on the AE eCRF and submitted to BioMarin Pharmacovigilance and in the subject's medical record to facilitate source data verification.

For some SAEs and EOSI, the Sponsor may follow up by telephone, fax, electronic mail, and/or a monitoring visit to obtain additional case details (eg, hospital discharge summary, consultant report, or autopsy report) deemed necessary to appropriately evaluate the SAE or EOSI report.

10.7 Post-Study Adverse Events

At the last scheduled visit, the Investigator should instruct each subject to report, to the Investigator and/or to BPV directly, any subsequent SAEs that the subject's personal physician(s) believes might be related to prior study drug.

The Investigator should notify the study Sponsor of any death or SAE occurring at any time after a subject has discontinued or terminated study participation, if the Investigator believes that the death or SAE may have been related to prior study drug. The Sponsor should also be notified if the Investigator should become aware of the development of cancer or of a congenital anomaly in a subsequently conceived offspring of a subject that participated in this study.

10.8 Urgent Safety Measures

The regulations governing clinical trials state that the Sponsor and Investigator are required to take appropriate urgent safety measures to protect subjects against any immediate hazards that may affect the safety of subjects, and that the appropriate regulatory bodies should be notified according to their respective regulations. According to the European Union (EU) Clinical Trial Directive 2001/20/EC, "…in the light of the circumstances, notably the occurrence of any new event relating to the conduct of the trial or the development of the investigational medicinal product where that new event is likely to affect the safety of the patients, the Sponsor and the Investigator shall take appropriate urgent safety measures to protect the patients against any immediate hazard. The Sponsor shall forthwith inform the competent authorities of those new events and the measures taken and shall ensure that the IRB/EC is notified at the same time."

The reporting period for these events which may require the implementation of urgent safety measures is the period from the time of signing of the ICF through the completion of the last study visit or at the Early Termination Visit (ETV). Investigators are required to report any events which may require the implementation of urgent safety measures to BioMarin within 24 hours.

Examples of situations that may require urgent safety measures include discovery of the following:

- Lack of study scientific value, or detrimental study conduct or management
- Discovery that the quality or safety of the IP does not meet established safety requirements

10.9 BioMarin Pharmacovigilance Contact Information

Contact information for BioMarin Pharmacovigilance is as follows:

BioMarin Pharmaceutical Inc.

Address	105 Digital Drive Novato, CA 94949
Phone:	PI
Fax:	PI
E-mail:	drugsafety@bmrn.com

The Investigator is encouraged to discuss with the medical monitor any AEs for which the issue of seriousness is unclear or questioned. Contact information for the medical monitor is as follows:

Name:	PI MD PhD
Address:	Biomarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949
Phone	PI (office) PI (mobile)
E-mail:	PI

11 APPROPRIATENESS OF MEASUREMENTS

The measures of efficacy to be used in this study are standard, ie, widely used and generally recognized as reliable, accurate, and relevant (able to discriminate between effective and ineffective agents). The measures of safety used in this study are routine clinical and laboratory procedures.

The chromogenic substrate FVIII assay and the one-stage clotting FVIII assay are both validated and utilize CE marked reagents.

12 STUDY PROCEDURES

12.1 Prestudy

An ICF must be signed and dated by the subject, the Investigator or designee and witness (if required) before any study-related procedures are performed.

12.2 Screening Visit(s)

Screening assessments should be performed within 42 days of BMN 270 infusion, while baseline assessments will take place within 7 days prior to BMN 270 infusion (Day 1). Should the screening visit occur within 30 days of the drug infusion, physical examination, vital signs, blood chemistry, LTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline.

During the first part of the Screening Period (Day -42 to Day -29), testing for AAV5 TAb titers using the ARUP screening assay may be performed, so that subjects can verify their AAV5 TAB status. Subjects who agree to participate in this activity will be asked to sign a separate ICF documenting this decision.

The following procedures will be performed during the Screening Period:

- Demographics (age, sex, race, ethnicity)
- Full medical history, including hemophilia A history, Hepatitis B, Hepatitis C, and HIV. Subjects with a history of hepatitis B or hepatitis C will be asked for information about the treatments received. Any prior pharmacokinetics information obtained while the subject was receiving prophylactic or on-demand FVIII therapy prior to the study should also be collected.
- Complete Physical Examination
- Height and weight
- Vital Signs (systolic and diastolic blood pressure, heart rate, respiration rate, and temperature)
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information) for the previous 12 months
- Distribution of subject diaries and training in diary completion
- Electrocardiogram
- Liver Ultrasound
- Samples for hFVIII Assays

- Baseline FVIII activity – chromogenic substrate FVIII assay
- Baseline FVIII activity level – one-stage clotting FVIII assay
- hFVIII coagulation activity exploratory assay (collected but not tested prior to enrollment)
- hFVIII inhibitors (Bethesda assay with Nijmegen modification)
- hFVIII total antibody assay (collected but not tested prior to enrollment)
- hFVIII antigen assay (collected but not tested prior to enrollment)

- Screen for Hepatitis B, Hepatitis C, and HIV if required (subjects with documented negative results 30 days prior to informed consent being obtained do not need to be retested)
  - Hepatitis B screening should include HBsAg, HBsAb, and HBcAb
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)
- Blood samples for Biomarker testing (may include HLA genotyping and FVIII genotyping status)

12.2.1 "Smart Rescreening" Visit

Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once.

If a patient has to be screened again because the original assessments have fallen out of the 28 + 14 day period allowed for Screening (refer to Section 12.2), then only the following assessments need to be performed (rather than the full list indicated in Section 12.2) for the patient to be successfully re-screened for the study:

- Vital signs
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- hFVIII Assays (only the hFVIII inhibitor level (Bethesda assay with Nijmegen modification))
- AAV5 TAb assay (ARUP)

- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)

12.3 Baseline Visit

Baseline values will be recorded from 1 to 7 days prior to the infusion visit. The following procedures will be performed during the Baseline Period:

- Brief physical examination
- Vital signs
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay
    - hFVIII coagulation activity exploratory assay
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
    - hFVIII total antibody assay
    - hFVIII antigen assay
- Von Willebrand Factor Antigen (VWF:Ag)
- Blood sample for AAV5 Total Antibody assay
    - Baseline sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
    - ABO blood typing assessment should be performed at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study.
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)
- PBMC collection for CTL baseline
- Blood sample for AAV5 TI assay
- Thrombin Generation Assay

- PCR of vector DNA in blood, saliva, urine, semen, and stools
- Exploratory biomarker assessments
- Cytokine bead array assay
- Hypersensitivity blood assessments
- Haemo-QoL-A assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.4 Treatment Visit/BMN 270 Infusion Visit (Day 1)

There will be one infusion visit for each subject. Subjects will remain in the clinic for at least 24 hours for the BMN 270 Infusion Visit. The following procedures will be performed during the BMN 270 Infusion Visit:

- Brief physical examination
- Start prophylactic corticosteroids (pre-infusion)
- Assessment of Adverse Events and Concomitant Medications
- AAV5 TAb assay (ARUP) (sample collected pre-infusion for analysis)
- Blood sample for AAV5 TI assay (sample collected pre-infusion for analysis)
- Fasting blood sample for future exploratory analysis (sample collected pre-infusion)
- Fasting lipid panel (blood triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) (sample collected pre-infusion)
    - Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit.
- BMN 270 Infusion
- Vital Signs
    - Vital signs will be recorded prior to BMN 270 infusion and then every 15 minutes (± 5 minutes) during BMN 270 infusion. Following infusion, vital signs will be monitored every 1 hour (± 5 minutes) for at least 8 hours during the subject's stay in the clinic.
- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Collection of samples for PCR testing should occur between 2 and 24 hours after the BMN 270 infusion has been completed

In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, C5, C5a, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.

Subjects should also start prophylactic corticosteroids, prior to BMN 270 infusion, on Study Day 1 (refer to Table 9.1.6 for a possible prophylactic corticosteroid regimen, and to Section 9.4.8.2 further discussion). Clinical judgment, weighting the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses. Discussions between the Investigator and Medical Monitor are advised for any questions or concerns.

12.5 BMN 270 Infusion Follow-Up Visits

At applicable sites, certain study assessments may be performed by a mobile nursing (MN) professional at the patient's home or another suitable location, such as their school or office, to improve access and convenience for patients participating in the study. The Sponsor may select a healthcare company that will be responsible for providing MN services for participating sites (the MN vendor). The MN vendor is responsible for ensuring that all MN professionals are licensed, qualified, and in good standing, as per applicable regulations, and that appropriate background checks have been performed. If the investigator at a participating site determines that MN services are appropriate for a patient and the patient gives written informed consent to participate in MN visits, the MN network will communicate with the patient and the patient’s site. Potential MN visits are designated in the Schedules of Events.

12.5.1 Week 1

During Week 1, the subject will be assessed on Study Day 2, Study Day 4, and Study Day 8.

12.5.1.1 Week 1, Study Day 2

On Study Day 2, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Separate collection on Day 2 is not required if vector shedding samples were collected on Day 1 post-BMN 270 infusion; if collection on Day 2 is needed, it must occur within 24 hours of the time of the BMN 270 infusion.
- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection
- Cytokine bead array assay

12.5.1.2 Week 1, Study Day 4

On Study Day 4, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
- PCR of vector DNA in blood, saliva, urine, semen, and stools

12.5.1.3 Week 1, Study Day 8

On Study Day 8, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications

- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- Samples for FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level
    - hFVIII antigen assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools
- Cytokine bead array assay

12.5.2 Weeks 2-26

After Week 1 (Day 8), subjects will return to the study site once a week (± 48 hours) during Weeks 2-26.

12.5.2.1 Once per week (Weeks 2 through 26)

The following procedures will be performed once per week from Weeks 1 through 26:

- Brief physical examination (complete physical examination at Week 26)
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, physical examination will not be performed.
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
    - For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.
- Vital Signs
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, vital signs will not be performed.

- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are >1.5x ULN) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN..
- Samples for FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level
    - hFVIII antigen assay

12.5.2.2 Every Other Week (Weeks 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, and 26)

The following procedures will be performed every other week (at Weeks 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, and 26):

- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection

12.5.2.3 Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26

The following procedures will be performed at Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26:

- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Collection to occur until at least 3 consecutive results below the limit of detection are obtained. Semen samples should continue to be collected and tested through Week 12, even if 3 consecutive results below the limit of detection in that compartment have been recorded prior to that time point.

12.5.2.4 Weeks 2, 4, 6, 8, 10, 12, 16, 20, and 26

The following procedures will be performed at Weeks 2, 4, 6, 8, 10, 12, 16, 20, and 26:

- hFVIII total antibody assay

12.5.2.5 Weeks 2, 4, 6, 8, 14, 20, and 26

The following procedures will be performed at Weeks 2, 4, 6, 8, 14, 20, and 26:

- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)

12.5.2.6 Weeks 2, 6, 10, 14, 18, 22, and 26

The following procedures will be performed at Weeks 2, 6, 10, 14, 18, 22, and 26:

- Exploratory biomarker assessments

12.5.2.7 Weeks 2, 6, 10, 14, 20, 24, and 26

The following procedures will be performed at Weeks 2, 6, 10, 14, 20, 22, and 26:

- Cytokine bead array assay

12.5.2.8 Weeks 4, 8, 12, 16, 20, and 26

The following procedures will be performed at Weeks 4, 8, 12, 16, 20, and 26:

- Weight
- VWF:Ag

12.5.2.9 Weeks 12 and 26

The following procedures will be performed at Weeks 12 and 26:

- Urine tests (to be performed locally)
- Haemo-QoL-A QoL assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.5.2.10 Week 16

The following procedure will be performed at Week 16:

- Testing for reactivation of hepatitis B and hepatitis C (only in subjects with evidence of prior exposure to hepatitis B and/or hepatitis C)
    - Subjects who receive therapeutic oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.6.

12.5.2.11 Weeks 20, 24, and 26

The following procedure will be performed at Weeks 20, 24, and 26:

- Thrombin Generation Assay

12.5.3 Post-Infusion Follow-Up – Weeks 27-52

During Weeks 27-36, subjects will return to the study site weekly (± 48 hours). During Weeks 37-52, subjects will return to the study site every 2 weeks (Week 38, 40, 42, 44, 46, 48, 50, and 52) (± 1 week). At these visits, the following procedures will be completed:

12.5.3.1 Every Visit

At every visit (Weeks 27-36, 38, 40, 42, 44, 46, 48, 50, and 52), the following procedures will be performed:

- Physical examination
    - Brief physical examination should be performed at all weeks except Week 52, when a complete physical examination should be performed
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, physical examination will not be performed.
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
    - For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.
- Vital Signs
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, vital signs will not be performed.
- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)

- FVIII coagulation activity exploratory assay
- Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
- FVIII protein assay

12.5.3.2 Weeks 28, 30, 32, 34, 36, 44, and 52

At Weeks 28, 30, 32, 34, 36, 44, and 52, the following procedure will be performed:

- PBMC collection

12.5.3.3 Weeks 32, 36, 44, and 52

At Weeks 32, 36, 44, and 52, the following procedures will be performed:

- Weight
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.4.8.2.1)
- FVIII antibody titer
- Cytokine bead array assay

12.5.3.4 Weeks 32, 36, 40, 44, 48, and 52

At Weeks 32, 36, 40, 44, 48, and 52, the following procedures will be performed:

- Exploratory biomarker assessments
- TGA Assay
- AAV5 TAb Assay
- AAV5 TI Assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools
  - Sample testing to occur until at least 3 consecutive sample below the limit of detection results have been obtained. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 52 should continue to have PCR testing of semen every 4 weeks until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).
  - Subjects considered to be treatment failures must continue to provide samples for PCR assessment at these timepoints until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints)

12.5.3.5 Week 36 and 52

At Weeks 36 and 52, the following procedures will be performed:

- Urine Tests (refer to Table 9.7.6.2.1)
- VWF:Ag

12.5.3.6 Week 52

At Week 52, the following procedures will be performed:

- Haemo-QoL-A assessment
- EQ-5D-5L
- HAL
- WPAI+CIQ:HS

12.6 Post-Infusion Follow-Up – Years 2-5

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 52 of the study by attending only the Q12W and End of Year visits during Years 2-5.

Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every 6 weeks during Years 2-5 until vector shedding has cleared (but do not have to perform other scheduled assessments at those study visits).

During Years 2-5 of Post-Infusion Follow-up, the following procedures will be completed:

12.6.1 Years 2-5 – Every 6 Weeks (not required for treatment failure)

During Years 2-5, every 6 weeks (± 2 weeks), the following procedures will be performed:

- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
    - For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.

- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
        - If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.
    - FVIII protein assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)
    - Sample testing during Years 2-5 is not required if at least 3 consecutive samples are clear by the end of Year 2. Subjects who have not had 3 consecutive semen samples below the limit of detection by the end of Year 1 should continue to have PCR testing of semen every 6 weeks during Years 2-5 until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).
        - Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every 6 weeks during Years 2-5 until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints).

12.6.2 Years 2-5 – Every 12 Weeks and End of Year Visits (required for all subjects)

During Years 2-5, subjects will be asked to return to the study site for visits at the following study weeks (±2 weeks):

- Year 2 – Week 64, Week 76, Week 88, Week 104
- Year 3 – Week 116, Week 128, Week 140, Week 156
- Year 4 – Week 168, Week 180, Week 192, Week 208
- Year 5 – Week 220, Week 232, Week 244, Week 260

For each of these years, the last study visit listed (Week 104, Week 156, Week 208, and Week 260) will serve as an End of Year visit.

At the every 12 week and End of Year visits, the following procedures will be performed:

- Brief physical examination (at every 12 week visits) or complete physical examination (at End of Year visits)
- Weight (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are ≥1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
        - If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.
    - FVIII protein assay
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1) (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Urine Tests (refer to Table 9.7.6.2.1) (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Vital Signs
- AAV5 TAb Assay
- AAV5 TI Assay
- FVIII antibody titer
- Haemo-QoL-A assessment (at End of Year visits only)
- EQ-5D-5L (at End of Year visits only)

- HAL (at End of Year visits only)
- WPAI+CIQ:HS (at End of Year visits only)
- Exploratory biomarker assessments
- PBMC collection
- VWF:Ag
- TGA Assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)
    - Sample testing during Years 2-5 is not required if at least 3 consecutive samples are below the limit of detection during the Post-Infusion Follow-Up period in Weeks 1-52. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 52 should continue to have PCR testing of semen every 6 weeks during Years 2-5 until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).

12.7 Early Termination Visit

The Early Termination visit will occur on the date the subject withdraws from the study, even if the date does not correspond to a protocol-specific visit.

If a subject leaves the study prior to the Week 260 visit, the subject will be asked to return to the study site and complete an Early Termination visit. At the Early Termination visit, as many of the following assessments as possible should be done:

- Complete physical examination
- Weight
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Vital signs
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay

  - hFVIII coagulation activity exploratory assay
  - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
  - hFVIII antigen assay
- hFVIII total antibody assay
- Exploratory biomarker assessments
- Blood sample for AAV5 Total Antibody assay
  - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection for CTL baseline
- VWF:Ag
- Thrombin Generation Assay
- Haemo-QoL-A QoL assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire
- PCR of vector DNA in blood, saliva, urine, semen, and stool
  - Sample testing at the Early Termination Visit is not required if at least 3 consecutive samples are clear during the period of the subject's participation in the study.

12.8 End of Study

The study will end after the last subject yet to complete the last Long-Term Follow-Up visit (Week 260) does so, has transferred to another BMN 270 study, is withdrawn from the study, or discontinues from the study. BioMarin reserves the right to discontinue the study any time for clinical or administrative reasons and to discontinue participation of an individual Investigator or site for clinical or administrative reasons, including, but not limited to, poor enrollment or noncompliance with procedures of the protocol or GCP. In addition, the study may be terminated if, in the opinion of BioMarin, the safety of the study subjects may be compromised.

13 DATA QUALITY ASSURANCE

BioMarin personnel or designees will visit the study site prior to initiation of the study to review with the site personnel information about the IP, protocol and other regulatory document requirements, any applicable randomization procedures, source document requirements, CRFs, monitoring requirements, and procedures for reporting AEs, including SAEs.

At visits during and after the study, a CRA will monitor the site for compliance with regulatory documentation, with a focus on accurate and complete recording of data on CRFs from source documents, adherence to protocol, randomization (if applicable), SAE reporting and drug accountability records.

Sites will enter study data into eCRFs into the study EDC system. Data Quality Control will be performed by BioMarin Clinical Data Management or designee through implementation of quality control checks specified in the study data management plan and edit check specifications.

14 STATISTICAL METHODS AND DETERMINATION OF SAMPLE SIZE

14.1 Statistical and Analytical Plans

The statistical analysis plan (SAP) will provide additional details on the planned statistical analysis. Unless otherwise stated, all analyses will be performed using SAS.

14.1.1 Interim Analyses

No interim analysis is planned.

14.1.2 Procedures for Accounting for Missing, Unused and Spurious Data

Because the completeness of the data affects the integrity and accuracy of the final study analysis, every effort should be made to ensure complete, accurate, and timely data collection and, therefore, avoid missing data.

Sensitivity analyses will be conducted to assess the impact of missing data on the primary efficacy endpoint analysis. Additional details regarding the handling of missing data will be provided in the SAP.

14.2 Efficacy Analysis

The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using longitudinal methods, such as mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at ≥ 5 IU/dL at 26 weeks following study treatment. FVIII activity will be assessed weekly during the study period. We can only assess the true steady state of FVIII produced from BMN 270 after a minimum of 72 hours (or 5x the known half-life of the FVIII replacement therapy administered) has elapsed since the last infusion of FVIII concentrates.

14.3 Safety Analysis

The Medical Dictionary for Regulatory Activities terminology (MedDRA) will be used by the Sponsor to assign system organ class and preferred term classification to events and diseases, based on the original terms entered on the eCRF.

All AEs will be coded using the current version of MedDRA. The incidence of AEs will be summarized by system organ class, preferred term, relationship to study drug, and severity. A by-subject listing will be provided for those subjects who experience a serious AE (SAE),

including death, or experience an AE associated with early withdrawal from the study or study drug.

Clinical laboratory data will be summarized by the type of laboratory test. For each clinical laboratory test, descriptive statistics will be provided on Baseline as well as all subsequent visits. Descriptive statistics for physical examination results and vital signs will also be provided.

Analysis of neutralizing antibody response and other immunological parameters as well as vector shedding will be primarily descriptive and involve both inter-subject and intra-subject comparisons across doses.

Detailed statistical methods will be provided in the SAP.

14.4 Determination of Sample Size

The sample size is based upon clinical considerations and is sufficient to detect a strong clinical efficacy signal. Approximately 10 subjects may be dosed in the study.

14.5 Analysis Populations

The efficacy analysis set will be comprised of all subjects who have received the BMN 270 infusion.

The safety population will consist of all subjects who receive BMN 270 infusion during the study.

14.6 Changes in the Conduct of the Study or Planned Analyses

Only BioMarin may modify the protocol. Any change in study conduct considered necessary by the Investigator will be made only after consultation with BioMarin, who will then issue a formal protocol amendment to implement the change. The only exception is when an Investigator considers that a subject's safety is compromised without immediate action. In these circumstances, immediate approval of the chairman of the IRB/EC must be sought, and the Investigator should inform BioMarin and the full IRB/EC within 2 working days after the emergency occurs.

With the exception of minor administrative or typographical changes, the IRB/EC must review and approve all protocol amendments. Protocol amendments that have an impact on subject risk or the study objectives, or require revision of the ICF, must receive approval from the IRB/EC prior to their implementation.

When a protocol amendment substantially alters the study design or the potential risks or burden to subjects, the ICF will be amended and approved by BioMarin and the IRB/EC, and all active subjects must again provide informed consent.

15 DATA MONITORING COMMITTEE

The Data Monitoring Committee (DMC) will consist of experts in clinical trials, statistics, and hemophilia. The DMC will review available safety and efficacy data during the study on an ongoing basis to determine, based on emerging data and the benefit/risk profile, whether further enrollment should continue.

Duties of the DMC include:

- Conducting an ongoing review of individual subject safety and efficacy data during the study;
- Recommending whether to proceed with enrollment of subjects at a different gating schedules based on emerging data from 270-203 and the overall risk/benefit analysis of BMN 270;
- If applicable, considering whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction.
- Making other recommendations on the conduct and reporting of the trial based on their evaluation of clinical data including institution of any pause or stopping stages.

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC determination of safety and efficacy in treated subjects with the following triggers for pausing further enrollment:

- any related SAE;
- any related AE with a severity > Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

16 COSTS, COMPENSATION, AND SUBJECT INJURY

There will be no charge to study subjects to be in this study. BioMarin will pay all costs of tests, procedures, and treatments that are part of this study. In addition, after IRB/EC approval, BioMarin may reimburse the reasonable cost of travel for study-related visits in accordance with BioMarin's travel and reimbursement policy. BioMarin will not pay for any hospitalizations, tests, or treatments for medical problems of any sort related solely to the study subject's disease. Costs associated with such hospitalizations, tests, and treatments should be billed and collected in the way that such costs are usually billed and collected outside the study.

The Investigator should contact BioMarin immediately upon notification that a study subject has been injured by the study drug or by procedures performed as part of the study. Any subject who experiences a study-related injury should be instructed by the Investigator to seek immediate medical treatment at a pre-specified medical institution if possible, or at the closest medical treatment facility if necessary. The subject should be given the name of a person to contact to seek further information about, and assistance with, treatment for study-related injuries. The treating physician should bill the subject's health insurance company or other third party payer for the cost of this medical treatment. If the cost of the medical treatment is not covered by health insurance or another third party that usually pays these costs, then either BioMarin or the institution may pay for reasonable and necessary medical services to treat the injuries caused by the study drug or study procedures. In some jurisdictions, BioMarin is obligated by law to pay for study-related injuries without prior recourse to third party payer billing and/or regardless of fault. If this is the case, BioMarin will comply with the law.

17 CASE REPORT FORMS AND SOURCE DOCUMENTS

Electronic case report forms will be provided for each subject. The Investigator must review and electronically sign the completed eCRF casebook to verify its accuracy.

eCRFs must be completed using a web–based application developed and validated. Study site personnel will be trained on the application and will enter the clinical data from source documentation. Unless explicitly allowed in the eCRF instructions, blank data fields are not acceptable.

In the event of an entry error, or if new information becomes available, the value will be corrected by deselecting the erroneous response and then selecting or entering the factual response. In compliance with ICH GCP Guidelines and 21 CFR Part 11, the system will require the personnel making the correction to enter a reason for changing the value. The documented audit trail will include the reason for the change, the original value, the new value, the time of the correction and the identity of the operator.

BioMarin's policy is that study data on the eCRFs must be verifiable to the source data, which necessitates access to all original recordings, laboratory reports, and subject records. In addition, all source data should be attributable (signed and dated). The Investigator must therefore agree to allow direct access to all source data. Subjects must also allow access to their medical records, and subjects will be informed of this and will confirm their agreement when giving informed consent. If direct source document verification of study data by the site monitor is prohibited by institutional policy or local law, then the Investigator must make available facilities and/or personnel to allow GCP-compliant source verification to occur. Examples of such methods include certified copies of records which have study data visible but sensitive information redacted, or other GCP-compliant means agreed between the Investigator and the Sponsor.

A site monitor designated by BioMarin will compare the eCRFs with the original source documents at the study site and evaluate them for completeness and accuracy before designating them as "Source Data Verified" (SDV). If an error is discovered at any time or a clarification is needed, the site monitor, or designee, will create an electronic query on the associated field. Site personnel will then answer the query by either correcting the data or responding to the query. The site monitor will then review the response and determine either to close the query or re–query the site if the response does not fully address the question. This process is repeated until all open queries have been answered and closed.

Before a subject's eCRF casebook can be locked, data fields must be source data verified and all queries closed. Refer to the Study Monitoring Plan for details on which fields must be source data verified. The Data Manager, or designee, will set the status of the forms, visits, and the entire casebook to Locked. The Investigator will then electronically sign the casebook, specifying that the information on the eCRFs is accurate and complete. Upon completion of the CSR, an electronic copy of each site's casebooks will be copied to a compact disk (CD) and sent to each site for retention with other study documents.

18 STUDY MONITORING AND AUDITING

Qualified individuals designated by BioMarin will monitor all aspects of the study according to GCP and standard operating procedures for compliance with applicable government regulations. The Investigator agrees to allow these monitors direct access to the clinical supplies, dispensing, and storage areas and to the clinical files, including original medical records, of the study subjects, and, if requested, agrees to assist the monitors. The Investigator and staff are responsible for being present or available for consultation during routinely scheduled site visits conducted by BioMarin or its designees.

Members of BioMarin's GCP Compliance Department or designees may conduct an audit of a clinical site at any time before, during, or after completion of the study. The Investigator will be informed if an audit is to take place and advised as to the scope of the audit. Representatives of the FDA or other Regulatory Agencies may also conduct an audit of the study. If informed of such an inspection, the Investigator should notify BioMarin immediately. The Investigator will ensure that the auditors have access to the clinical supplies, study site facilities, original source documentation, and all study files.

19 RETENTION OF RECORDS

The Investigator must retain all study records required by BioMarin and by the applicable regulations in a secure and safe facility. The Investigator must consult a BioMarin representative before disposal of any study records, and must notify BioMarin of any change in the location, disposition or custody of the study files. The Investigator /institution must take measures to prevent accidental or premature destruction of essential documents, that is, documents that individually and collectively permit evaluation of the conduct of a study and the quality of the data produced, including paper copies of study records (eg, subject charts) as well as any original source documents that are electronic as required by applicable regulatory requirements.

All study records must be retained for at least 2 years after the last approval of a marketing application in the US or an ICH region and until (1) there are no pending or contemplated marketing applications in the U.S. or an ICH region or (2) at least 2 years have elapsed since the formal discontinuation of clinical development of the IP. The Investigator /institution should retain subject identifiers for at least 15 years after the completion or discontinuation of the study. Subject files and other source data must be kept for the maximum period of time permitted by the hospital, institution or private practice, but not less than 15 years. These documents should be retained for a longer period, however, if required by the applicable regulatory requirements or by a BioMarin agreement. BioMarin must be notified and will assist with retention should Investigator /institution be unable to continue maintenance of subject files for the full 15 years. It is the responsibility of BioMarin to inform the Investigator /institution as to when these documents no longer need to be retained.

20 USE OF INFORMATION AND PUBLICATION

BioMarin recognizes the importance of communicating medical study data and therefore encourages the publication of these data in reputable, peer-reviewed scientific journals and at seminars or conferences. The details of the processes of producing and reviewing reports, manuscripts, and presentations based on the data from this study will be described in the Clinical Trial Agreement between BioMarin and the Investigator/Institution. Consideration for authorship of all publications will be based on compliance with the Uniform Requirements for Manuscripts Submitted to Biomedical Journals ("Uniform Requirements") of the International Committee of Medical Journal Editors (ICMJE) (http://www.icmje.org/ethical_1author.html) and good publication practices (GPP).

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22 INVESTIGATOR RESPONSIBILITIES

22.1 Conduct of Study and Protection of Human Subjects

In accordance with FDA Form 1572 and/or principles of ICH E6 GCP, the Investigator will ensure that:

- He or she will conduct the study in accordance with the relevant, current protocol and will only make changes in a protocol after notifying the sponsor, except when necessary to protect the safety, rights, or welfare of subjects.
- He or she will personally conduct or supervise the study.
- He or she will inform any potential subjects, or any persons used as controls, that the drugs are being used for investigational purposes, and he or she will ensure that the requirements relating to obtaining informed consent in 21 CFR Part 50 and/or ICH E6 Sections 2.9 and 4.8 are met. As well, he or she will ensure that IRB/EC review and approval in 21 CFR Part 56 and/or ICH E6 Section 2.6 are met.
- He or she will report to the sponsor adverse experiences that occur in the course of the investigation in accordance with 21 CFR 312.64 and/or ICH E6 Section 4.11.
- He or she has read and understands the information in the Investigator's Brochure, including potential risks and side effects of the drug.
- His or her staff and all persons who assist in the conduct of the study are informed about their obligations in meeting the above commitments
- Adequate and accurate records in accordance with 21 CFR 312.62 and/or ICH E6 Section 4.9 are kept, and those records are available for inspection in accordance with 21 CFR 312.68 and/or ICH E6 Section 4.9.7.
- The IRB/EC complies with the requirements of 21 CFR Part 56, ICH E6 Section 3.0, and other applicable regulations, and conducts initial and ongoing reviews and approvals of the study. He or she will also ensure that any change in research activity and all problems involving risks to human subjects or others are reported to the IRB/EC. Additionally, he or she will not make any changes in the research without IRB/EC approval, except where necessary to eliminate apparent immediate hazards to human subjects.
- He or she agrees to comply with all other requirements regarding the obligations of clinical Investigators and all other pertinent requirements in 21 CFR Part 312 and/or ICH E6.

23 SIGNATURE PAGE

Protocol Title: A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5

Protocol Number: 270-203 Amendment 2

I have read the foregoing protocol and agree to conduct this study as outlined. I agree to conduct the study in compliance with all applicable regulations and guidelines, including E6R2 ICH, as stated in the protocol, and other information supplied to me.

Investigator Signature Date

Printed name: _____

Accepted for the Sponsor:

DocuSigned by:
PI
Signer Name: PI
Signing Reason: I approve this document
Signing Time: 04-Oct-2019 | 8:58 AM PDT
2CCBE97A041140AFB8B0B5194FC717C0

Medical Monitor Signature Date

Printed name: _PI MD PhD, PI , Clinical Sciences___

24 APPENDIX 1: SAMPSON'S ANAPHYLAXIS CRITERIA

According to the National Institute of Allergy and Infectious Disease/Food Allergy and Anaphylaxis Network (NIAID/FAAN) Second Symposium on the definition and management of anaphylaxis, anaphylaxis is highly likely when any one of the following 3 criteria are fulfilled:

1. Acute onset of an illness (minutes to several hours) with involvement of the skin, mucosal tissue, or both (eg, generalized hives, pruritus or flushing, swollen lips-tongue-uvula)
2. AND AT LEAST ONE OF THE FOLLOWING

a. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced peak expiratory flow [PEF], hypoxemia)

b. Reduced blood pressure (BP) or associated symptoms of end-organ dysfunction (eg, hypotonia [collapse], syncope, incontinence)

3. Two or more of the following that occur rapidly after exposure *to a likely allergen for that patient* (minutes to several hours):

a. Involvement of the skin-mucosal tissue (eg, generalized hives, itch-flush, swollen lips-tongue-uvula)

b. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced PEF, hypoxemia)

c. Reduced BP or associated symptoms (eg, hypotonia [collapse], syncope, incontinence)

d. Persistent gastrointestinal symptoms (eg, crampy abdominal pain, vomiting)

4. Reduced BP after exposure to *known allergen for that patient* (minutes to several hours):

a. Infants and children: low systolic blood pressure (age specific) or greater than 30% decrease is systolic BP

b. Adults: systolic BP of less than 90 mmHg or greater than 30% decrease from that person's baseline.

Source: Sampson, 2006.

25 PROTOCOL AMENDMENT TEXT REVISIONS

The following table summarizes the revisions made to Protocol Amendment 1 and relates the changes to the appropriate rationale (see pages 2-4). Added text is indicated by underlined font and deleted text is indicated by ~~strikethrough~~ font.

Section No./Title	Revision	Rationale
Synopsis/Study Sites	Approximately ~~2-3~~5-6 sites ~~in the United Kingdom~~globally	17
Synopsis/Study Rationale	Treatment of severe HA presently mainly consists of intravenous injection….	19
Synopsis/Study Rationale	Extended half-life products created through chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) ~~and~~or bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half-life ~~to maximum of~~ by approximately 50% (half-life 18-19 hours~~, leaving critical periods when FVIII~~), and thus, show promise in reduced dosing and maintaining activity levels ~~are below the therapeutic range and leaving~~above a 1% trough for a greater proportion of the dosing interval. However, patients ~~vulnerable to bleeding and concomitant sequelae. As such, despite currently available~~ with severe HA who are treated with extended half-life FVIII remain dependent on multiple infusions to maintain critical levels of FVIII activity. More recently, emicizumab has been approved for the treatment of HA. Emicizumab is a humanized, bispecific monoclonal antibody that binds to both activated coagulation factor IX (FIX) and coagulation factor X (FX), thereby mimicking the function of activated FVIII. Emicizumab represents an incremental improvement in therapeutic burden over replacement ~~therapies, a high~~FVIII products since it can be administered subcutaneously as opposed to intravenously; however, it is still a chronic treatment that requires repeat administration every 1-4 weeks, and sustained FVIII activity levels above 5-10 IU/dL have rarely been attained even with extended half-life products. Therefore, there is a strong unmet need ~~remains for maintenance of a sustained, consistent FVIII level compatible with a normal and hemorrhage-free life~~for a treatment that can alter the fundamental problem for patients with HA, namely attainment of sustained, hemostatically effective FVIII activity levels long-term (ie, over multiple years) without the need for treatment.	19
Synopsis/Study Rationale	…. bleeding phenotype from severe to moderate or mild, following a single peripheral vein infusion of AAV8-hFIX vector (Nathwani, ~~2014~~2018).	19
Synopsis/Study Rationale	The vector schematic figure and notes have been updated. BMN 270 is delivered by a single IV dose and is designed to achieve stable, ~~potentially life-long~~durable expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.	14, 19
Synopsis/Study Rationale	~~Preliminary~~Three-year results from 270-201 have demonstrated that following gene transfer, ~~sustained~~ mean and median FVIII activity levels above 15% (15 IU/dL~~) up to~~), as measured by a ~~year's observation is~~chromogenic substrate assay, are achievable ~~with~~and sustained following a ~~dose~~single infusion of ~~4-~~6E13 vg/kg ~~with~~of BMN	15

Section No./Title	Revision	Rationale
	270, with administration of prophylactic corticosteroids and an acceptable safety profile (Pasi, ~~2017).~~ 2019). In addition, an interim analysis of clinical study 270-301, an ongoing phase 3 study designed to assess the efficacy and safety of BMN 270 at a dose of 6E13 vg/kg and administration of therapeutic corticosteroids (ie, in response to ALT elevations), demonstrated FVIII activity levels that were also well above 15%, albeit lower than what was observed for the 6E13 vg/kg cohort in 270-201 at 26 weeks (Pasi, 2019).	
Synopsis/Study Rationale	AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes~~.~~ (Boutin, 2010; Hayes, 2019).	19
Synopsis/Study Design and Plan	Approximately 10 subjects may be enrolled at ~~2-3~~5-6 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion.	17
Synopsis/Study Design and Plan	~~The~~ An independent Data ~~Review Board (DRB~~Monitoring Committee (DMC) will consist of ~~the Principal Investigators~~experts in clinical trials, statistics, and ~~Sponsor's Medical Monitor, as well as~~ hemophilia ~~expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201. The DRB~~. The DMC will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 2 represents one dosing schedule scenario):	13
Synopsis/Study Design and Plan	Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on ~~DRB~~DMC evaluation of safety and efficacy in treated subjects	13
Synopsis/Study Design and Plan	Following a temporary halt of enrolment, the ~~DRB~~DMC may approve resumption of enrolment in….	13
Synopsis/Study Design and Plan	… prior FVIII prophylaxis for each subject will be continued at the discretion of the ~~DRB~~DMC based on individual subject status…	13
Synopsis/Study Design and Plan	Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by ~~either Week 26 or~~ Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor or Sponsor-designated Data Monitor, follow an abbreviated visit schedule after ~~either Week 26 or~~ Week 52~~, as applicable~~ by attending only the Q12W and ~~at the discretion~~End of ~~the Investigator~~Year visits during Years 2-5.	3
Synopsis/Study Design and Plan	… additional subjects may be recruited into each cohort at any time, if deemed necessary by the ~~DRB~~DMC.	13
Synopsis/Diagnosis and all criteria for inclusion and exclusion	… after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with ~~no detectable~~ viral vector DNA below the limit of detection Willing to abstain from consumption of alcohol for at least the first ~~26~~52 weeks following BMN 270 infusion	4, 9, 10

Section No./Title	Revision	Rationale
	Major surgery planned in the ~~26~~52-week period following the infusion with BMN 270	
7.2/Previous Clinical Studies	~~Study~~ Ongoing clinical studies for BMN 270 include: 270-201 ~~is an ongoing,~~ a phase 1/2, dose-escalation study ~~to assess the safety, tolerability, and efficacy of BMN 270~~ in patients with severe ~~hemophilia A (FVIII <1 IU/dL). Subjects received a single BMN~~ HA 270 ~~infusion and are to be followed for safety and efficacy for up to 5 years. A total of 15 subjects have been enrolled and dosed~~-203, a phase 1/2 study in patients with ~~a single IV infusion of~~severe HA who have anti-AAV5 antibody titers 270-301, a phase 3 study in patients with severe HA who receive BMN 270 at the 6E13 vg/kg dose level 270-302, a phase 3 study in patients with severe HA who receive BMN 270 at ~~one of 4 dose levels (6E12, 2E13,~~ the 4E13 ~~, and 6E13~~ vg/kg~~).~~ dose level A comprehensive review of safety, efficacy, and immunogenicity results from 270-201 and 270-301 as of the latest data cut….	19
7.3/Study Rationale	Treatment of severe HA presently mainly consists of intravenous injection….	19
7.3/Study Rationale	Extended half-life products created through chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) ~~and~~or bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half~~-~~-life ~~to maximum of~~by approximately 50% (half-life 18-19 hours~~, leaving critical periods when FVIII~~), and thus, show promise in reduced dosing and maintaining activity levels ~~are below the therapeutic range and leaving~~above a 1% trough for a greater proportion of the dosing interval. However, patients ~~vulnerable to bleeding and concomitant sequelae. As such, despite currently available~~ with severe HA who are treated with extended half-life FVIII remain dependent on multiple infusions to maintain critical levels of FVIII activity. More recently, emicizumab has been approved for the treatment of HA. Emicizumab is a humanized, bispecific monoclonal antibody that binds to both activated coagulation factor IX (FIX) and coagulation factor X (FX), thereby mimicking the function of activated FVIII. Emicizumab represents an incremental improvement in therapeutic burden over replacement ~~therapies, a high~~FVIII products since it can be administered subcutaneously as opposed to intravenously; however, it is still a chronic treatment that requires repeat administration every 1-4 weeks, and sustained FVIII activity levels above 5-10 IU/dL have rarely been attained even with extended half-life products. Therefore, there is a strong unmet need ~~remains for maintenance of a sustained, consistent FVIII level compatible with a normal and hemorrhage-free life~~for a treatment that can alter the fundamental problem for patients with HA, namely attainment of sustained, hemostatically effective FVIII activity levels long-term (ie, over multiple years) without the need for treatment.	19
7.3/Study Rationale	… following a single peripheral vein infusion of AAV8-hFIX vector (Nathwani, ~~2014~~2018).	19
7.3/Study Rationale	The vector schematic figure and notes have been updated.	14

Section No./Title	Revision	Rationale
7.3/Study Rationale	BMN 270 is delivered by a single IV dose and is designed to achieve stable, ~~potentially life long~~durable expression….	19
7.3/Study Rationale	FVIII activity and whether these levels are sufficient to alter the clinical phenotype. ~~Preliminary~~Three-year results from 270-201 have demonstrated that following gene transfer, ~~sustained~~ mean and median FVIII activity above 15% (15 IU/dL~~) up to~~), as measured by a ~~year's observation is~~chromogenic substrate assay, are achievable ~~with~~and sustained following a ~~dose~~single infusion of ~~4~~ 6E13 vg/kg ~~with~~of BMN 270, with administration of prophylactic corticosteroids and an acceptable safety profile (Pasi, ~~2017~~ 2019). In addition, an interim analysis of clinical study 270-301, an ongoing phase 3 study designed to assess the efficacy and safety of BMN 270 at a dose of 6E13 vg/kg and administration of on-demand corticosteroids (ie, in response to ALT elevations), demonstrated FVIII activity levels that were also well above 15% at 26 weeks, albeit lower than what was observed for the 6E13 vg/kg cohort in 270-201 (Pasi, 2019).	19
7.3/Study Rationale	AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes~~.~~ (Hayes, 2019; Boutin, 2010).	19
7.4/Summary of Overall Risks and Benefits	The majority of subjects in the ongoing 270-201 clinical study who have received 4E13 or 6E13 vg/kg doses of BMN 270 have had Grade 1 asymptomatic elevations in ALT~~. For most subjects, the elevations~~ that have reached only slightly above the upper limit of normal (ULN). Subjects in the 6E13 vg/kg cohort received corticosteroids, predominantly on a prophylactic basis, starting 2-4 weeks following BMN 270 infusion, whereas those in the 4E13 vg/kg cohort received corticosteroids only if they experienced an elevation in their ALT ≥ 1.5x ULN (ie, "therapeutic"). Based on the effectiveness of transient oral ~~corticosteroid~~corticosteroids used starting 8-10 weeks after vector infusion to suppress a presumed Class 1 (cytotoxic T-cell) response in prior studies with hepatic transduction with AAV vectors (Mingozzi, 2013), subjects were treated with 7-32 weeks of oral corticosteroids prophylactically or in response to the elevations in ALT to limit hepatotoxicity, ensure preservation of the transduced hepatocytes, and minimize any associated impact of such on FVIII levels. Using this approach, no sustained loss of FVIII activity has been observed in subjects with ALT elevations~~, consistent with maintaining a high level of hepatocyte function. Moreover, the rise in ALT levels~~. Moreover, the rise in ALT levels was not accompanied by significant or lasting abnormalities in other liver tests such as AST, bilirubin or albumin, indicating that extent of toxicity is limited. At the highest dose tested in 270-201 (6E13 vg/kg), the majority of subjects achieved chromogenic FVIII activity levels above 50 IU/dL at 52 weeks post-infusion. Subjects in that cohort also reported markedly decreased bleeding compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. Subjects at all dose levels continue to be followed. In the ongoing 270-301 clinical study, a majority of subjects, all dosed with 6E13 vg/kg BMN 270, have experienced asymptomatic elevations in ALT. Subjects received therapeutic corticosteroids if they experienced an ALT elevation and/or had a FVIII activity level that had declined >35% from peak values. While sporadically observed cytotoxic T-cell responses have not been correlated with ALT elevations, the majority of subjects with transient rises in ALT levels had associated declines in their FVIII activity that subsequently increase to higher	15

Section No./Title	Revision	Rationale
	than pre-ALT elevation levels following initiation of therapeutic corticosteroids. Similar to 270-201, the rise in ALT levels in 270-301 were not accompanied by significant or lasting aberrations in other liver tests such as AST, bilirubin or albumin, indicating that extent of toxicity is limited. At the time of the 270-301 interim analysis, the modified intent-to-treat population (n=16, who had completed ≥26 weeks on-study) had mean and median chromogenic FVIII activity levels of 36.1 and 33.2 IU/dL at 26 weeks, respectively, which are lower than the corresponding values observed for the 270-201 6E13 vg/kg cohort. There has been one HIV-positive subject in the ongoing 270-302 clinical study who experienced Grade 3 asymptomatic elevations in ALT and AST, which has been attributed to an interaction between one or more of his antiretroviral therapy medications and/or unsuspected underlying hepatic disease with BMN 270. In addition, there ~~has~~have been three subjects, including one ~~subject~~ with Gilbert's syndrome, in the ongoing 270-301 clinical study who ~~has~~have experienced Grade 3 asymptomatic elevations in ALT and AST. These cases have led to the exclusion of subsequent HIV-positive subjects and requirement of liver tests at Screening that are <1.25 times the upper limit of the normal range in the ongoing 270-301 and 270-302 clinical studies. Of note, two HIV-positive subjects in 270-301 and one presumed Gilbert's syndrome subject in 270-201 have received BMN 270 without experiencing any elevations in ALT beyond Grade 1 to date. Overall, the literature and clinical experience with BMN 270 suggests that transient elevations in liver enzymes are expected following AAV-based gene therapy for the treatment for hemophilia A or B without any long-term concerns of hepatic injury (Manno, 2006; Nathwani, 2011; George, 2016; Miesbach, 2016; Pasi, 2017). ~~At the highest dose evaluated in 270-201 (6E13 vg/kg), the majority of subjects achieved FVIII levels above 50 IU/dL at 52 weeks post-infusion. Subjects in that cohort also reported markedly decreased bleeding episodes compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. Subjects at all dose levels continue to be followed. Subjects in 270-203, who have pre-existing immunity to the AAV5 vector capsid, may get no benefit from the study (in terms of increased FVIII activity) while possibly creating cross-reactive antibodies that may potentially preclude dosing with other serotypes.~~ As with any infused biological product, there is a potential risk of acute, systemic hypersensitivity reactions (including anaphylaxis) with BMN 270. No hypersensitivity reactions were observed during dosing of BMN 270 in the 270-201 clinical study, although ~~one SAE of pyrexia was reported approximately 16 hours after the infusion in a subject in the 4E13 vg/kg cohort. The subject was treated with acetaminophen, and the fever resolved within 48 hours (see Investigator's Brochure for full details).~~ infusion-related hypersensitivity reactions~~,~~ (including ~~allergic reaction, maculopapular rash, and presyncope,~~anaphylaxis) have been ~~reported from ongoing, actively~~observed during dosing ~~clinical studies~~ of BMN 270~~, including this~~ in the 270-301 clinical study~~.~~ (refer to Investigator's Brochure for full details). All of the infusion-related reactions were effectively managed clinically and resolved without any clinical sequelae. ~~Refer to the Investigator's Brochure for additional details.~~ The current data available for BMN 270 ~~does not yet permit comprehensive assessment of the~~ has shown an established positive benefit:risk profile ~~of this investigational drug~~for BMN 270 at the 6E13 vg/kg dosing level, although impact of prophylactic corticosteroids requires further investigation. . Given the monitoring measures in	

Section No./Title	Revision	Rationale
	place in the clinical protocol(s) to minimize the risk to subjects participating in the existing studies, the identified risks are justified by the anticipated benefits that may be afforded to subjects. Each subject in 270-203 will have a comprehensive surveillance plan that monitors LTs during the study, and elevations in LT will be addressed according to the guidelines set forth in the protocol. Safety will be assessed by adverse event reporting and clinical laboratory assessments. For additional information on findings ~~in 270-201~~from other BMN 270 clinical studies, refer to the current version of the IB.	
9.1/Overall Study Design and Plan	Approximately 10 subjects may be enrolled at ~~2-3~~5-6 sites globally in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date. ~~The Data Review Board (DRB) will consist of the Principal Investigators and Sponsor's Medical Monitor, as well as hemophilia expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201. The DRB~~An independent Data Monitoring Committee (DMC), consisting of experts in clinical trials, statistics, and hemophilia, has been convened. The DMC will review available safety and efficacy data… Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on ~~DRB~~DMC evaluation of safety and efficacy…. Following a temporary halt of enrolment, the ~~DRB~~DMC may approve resumption of enrolment in either cohort…. Monitor will discuss whether to resume prior FVIII prophylaxis. Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by ~~either Week 26 or~~ Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor or Sponsor-designated Data Monitor, follow an abbreviated visit schedule after ~~either Week 26 or~~ Week 52~~, as applicable~~ by attending only the Q12W and ~~at the discretion~~End of ~~the Investigator~~Year visits during Years 2-5. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the ~~DRB~~DMC.	3, 13, 17, 19
Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, Table 9.1.5, Table 9.1.6	Updates were made to the tables and associated footnotes to be consistent with the changes in the protocol	1, 2, 3, 4, 5, 7, 8, 16, 18, 19
9.3.1/Inclusion Criteria	Participants must agree to contraception use for at least 12 weeks post-infusion; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with ~~no detectable~~ viral vector DNA below the level of detection.	4, 9

Section No./Title	Revision	Rationale
	Willing to abstain from consumption of alcohol for at least the first ~~26~~52 weeks following BMN 270 infusion.	
9.3.2/Exclusion Criteria	Major surgery planned in the ~~26~~52-week period following the infusion with BMN 270	10
9.3.3.1/Study Safety Evaluation Criteria	Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on ~~DRB~~DMC evaluation of safety and efficacy…. Additionally, the ~~DRB~~DMC should consider whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction. Following a temporary halt of enrolment, the ~~DRB~~DMC may approve… If a Grade 4 or Grade 5 adverse event assessed as related to study drug occurs in a study subject, further enrollment into the trial will be halted until a full safety review of the data by the ~~DRB~~DMC has taken place. Relevant reporting and discussion with the Sponsor and the ~~DRB~~DMC will take place before resumption of dosing. If any of the following events occur in a subject in the study who has received BMN 270 infusion, further enrollment into the trial will be temporarily put on hold pending prompt evaluation by the ~~DRB~~DMC. If any of the following events occurs in a subject in the study who has received BMN 270 infusion, a prompt evaluation by the ~~DRB~~DMC will be required. Further enrollment into the trial will continue while ~~DRB~~DMC evaluation is ongoing, unless deemed otherwise by the ~~DRB~~DMC.	13
9.4.2.1/Product Characteristics and Labeling	BMN 270 is a sterile, clear, colorless-to-pale yellow solution for IV infusion and is supplied in a ~~2~~10 mL ~~polypropylene cryovial.~~Crystal Zenith® (CZ) vial. Each CZ vial contains ~~1.1~~ 8.5 mL (extractable volume 8 mL) of AAV5-hFVIII-SQ at a concentration of 2E13 vector genomes per mL in a pH 7.4 phosphate buffer.	11
9.4.4/Directions for Administration	Of note, the IP has been shown to be stable at room temperature for ~~7.5~~approximately 10 hours following completion of product thaw. In the event of an infusion rate reaction with more than one dosing interruption, the infusion rate would not go beyond 1 mL/min. In case of hypersensitivity ~~or adverse drug reaction~~reactions, safety assessment, including physical examination and vital signs, will be done and additional blood samples….	19
9.4.8/Prior and Concomitant Medications	The following medications should be avoided, starting 30 days prior to and for at least ~~26~~52 weeks after BMN 270 infusion and minimized throughout the remaining duration of the study. • Medications which may reduce or increase the plasma concentration of corticosteroids	1
9.4.8.2/ Therapeutic ~~Glucocorticoid~~Corticosteroid	All subjects will be started on prophylactic corticosteroids starting on the day of infusion (Day 1). Table 9.1.6~~Therapeutic~~ provides an example of a possible prophylactic corticosteroid course, including taper and post-corticosteroid additional monitoring of FVIII activity, LTs, and hepatitis B/hepatitis C reactivation. Clinical	1

Section No./Title	Revision	Rationale
Treatment ~~for~~of Elevated Hepatic Transaminases	judgment, weighting the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses. Discussions between the Investigator and Medical Monitor are advised for any questions or concerns. Following initiation or completion of the prophylactic corticosteroid regimen, if ALT levels become increased (eg, ≥ ULN or ≥ 2x baseline value) and alternative etiologies have been ruled out, prompt institution of therapeutic or on-demand oral corticosteroids (prednisone or converted equivalent) should be ~~initiated when either of the following occurs post BMN 270 infusion in any subject and~~ considered after consultation with the Medical Monitor~~:~~ ~~ALT ≥ 1.5x ULN or ALT > ULN & > 2x baseline value in 2 consecutive assessments within 72 hours and alternative etiologies have been ruled out, or ALT ≥ 3x ULN in 2 consecutive assessments within 48 hours~~ (refer to ~~)~~Table 9.7.6.3.2). • Whenever possible, a confirmatory lab draw for ALT should be performed within 72 hours, along with FVIII activity, prior to initiating oral corticosteroids. • Corticosteroids may be delayed if elevations in ALT are clearly not related to BMN 270 (eg, elevated in ALT with concurrent increase in CPK due to intensive exercise) ~~The prescribed regimen for therapeutic oral corticosteroids is detailed in . Changes to the corticosteroid regimen should be made as follows:~~ Therapeutic corticosteroid treatment should be initiated at a dose of 60 mg/day. At minimum, the recommended duration of therapeutic corticosteroids is 60 mg/day for 3 weeks, 40 mg/day for 4 weeks, and 30 mg/day for 4 weeks, followed by a gradual taper thereafter. Should a scenario arise in which a deviation from the minimum recommended dose and/or duration of therapeutic corticosteroids may be clinically indicated, a discussion should take place between the Investigator and Medical Monitor regarding corticosteroid dose adjustments. Tapering of corticosteroid dosages should be guided by the following: Following initiation or completion of therapeutic oral corticosteroids, if increased ALT ~~elevation ≥1.5x~~ levels (eg, ≥ ULN or ~~ALT ≥ ULN & ≥~~ ≥ 2x baseline value ~~is~~) are reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor. Subjects on corticosteroids should receive appropriate counselling and support regarding side effects from the Investigator or the treating institution (eg, listings of side effects and when to notify carers, wallet card for emergencies if on steroids, etc.).	
Table 9.4.8.2.1/Adjustments to Corticosteroid Regimens	The table has been updated to reflect changes made elsewhere in the body of the protocol.	1, 2
9.6/Dietary or Other Protocol Restrictions	There are no dietary or other protocol restrictions for this study. Alcohol should be avoided starting 30 days prior to and for at least the first ~~26~~52 weeks of the study, and particularly within 48 hours prior to lab work.	9, 19

Section No./Title	Revision	Rationale
	Subjects should be advised to abstain from any blood, organ, or sperm donation after BMN 270 infusion, until there is no further evidence of vector shedding from PCR analysis of samples.	
9.7.2.1/FVIII Activity	… discussion with the Medical Monitor, follow an abbreviated visit schedule after ~~either Week 26 or~~ Week 52, as applicable and at the discretion of the Investigator.	3
Table 9.7.6.2.1	* ABO blood typing assessment should be performed ~~as part of the hematology assessment (~~at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study~~).~~	19
9.7.6.2/Clinical Laboratory Assessments	At applicable sites, certain study assessments may be performed by a mobile nursing (MN) professional at the patient's home or another suitable location, such as their school or office, to improve access and convenience for patients participating in the study. The Sponsor may select a healthcare company that will be responsible for providing MN services for participating sites (the MN vendor). The MN vendor is responsible for ensuring that all MN professionals are licensed, qualified, and in good standing, as per applicable regulations, and that appropriate background checks have been performed. If the investigator at a participating site determines that MN services are appropriate for a patient and the patient gives written informed consent to participate in MN visits, the MN network will communicate with the patient and the patient's site. Potential MN visits are designated in the Schedules of Events.	8
9.7.6.3/Liver and Hepatitis Testing	Subjects with documented results showing an absence of active hepatitis B or hepatitis C infection (as measured by ~~positive~~negative surface antigen or DNA for hepatitis B or ~~positive~~negative RNA testing for hepatitis C)….	19
Table 9.7.6.3.2/Evaluation of ALT Elevations	The table has been updated to reflect changes made elsewhere in the body of the protocol.	2, 19
9.7.6.5/Vital Signs	At visits where the MN services are used or shortened lab draw-only visits are conducted at the sites, the physical examination and vital signs assessments indicated in the Schedule of Events will not be performed. Vector shedding will also be extensively studied in the present clinical trial, at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5. Testing will continue until at least 3 ~~negative~~consecutive results below the limit of detection are obtained. Testing of semen will continue at least through Week 12, even if 3 consecutive ~~negative~~ results below the limit of detection have been recorded in that compartment prior to that time point. Subjects who have not had 3 consecutive ~~negative~~ semen samples below the limit of detection by Week 26 should continue to have PCR testing in semen every 4 weeks until 3 consecutive ~~negative~~ samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor). Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule (refer to Section 12.5.3) but who have not cleared vector shedding from all fluids must still provide samples for assessment until vector shedding has cleared: at Weeks 32, 36, 40, 44, 48, and 52 (during Year 1~~),~~) and every 4 weeks ~~(during Year 2), or every 6 weeks (~~during Years ~~3~~2-5~~).~~. Such subjects may provide samples on the designated study visit dates either at the sites or through use of a MN professional.	3, 4, 8, 18, 19

Section No./Title	Revision	Rationale
	… sperm tested positive for vector DNA on at least three visits, or if purified sperm tested ~~negative~~below the limit of detection for vector DNA on at least three consecutive visits. Contraception use may need to be extended beyond ~~26~~12 weeks in individual subjects based on observed vector shedding in semen. After ~~26~~12 weeks, subjects may stop contraception use only if they have had 3 consecutive ~~negative~~ semen samples below the limit of detection (upon consultation between the Investigator and Medical Monitor).	
10.3.3/Assessment of Seriousness, Severity, and Causality	The Investigator responsible for the care of the subject or medically qualified designee will assess AEs for severity, relationship to study drug and/or corticosteroids, and seriousness (refer to Section 10.2 for SAE definitions).	6
10.3.3.3/Causality	The Investigator will determine the relationship of an AE to the study drug and/or corticosteroids and will record it on the source documents and AE eCRF. Factors suggestive of a causal relationship could include (but are not limited to): • Plausible temporal relationship • Absence of alternative explanations • Rarity of event in a given patient or disease state • Absence of event prior to study drug and/or corticosteroid exposure • Consistency with study product pharmacology • Known relationship to underlying mechanism of study drug and/or corticosteroid action • Similarity to adverse reactions seen with related drug products • Abatement of AE with discontinuation of study drug and/or corticosteroids, and/or recurrence of AE with reintroduction of study drug and/or corticosteroids	6
Table 10.3.3.3.1/Causality Attribution Guidance	The table has been updated to reflect changes made elsewhere in the body of the protocol.	6
10.4.1.4/Abnormal Laboratory Values	Any laboratory result abnormality fulfilling the criteria for a SAE or EOSI should be reported as such, and recorded in the AE eCRF unless associated with an AE that has already been reported.	19
10.9/BioMarin Pharmacovigilance Contact Information	PI , ~~MA MB BChir~~ PI ch MD PhD Biomarin ~~(UK) Ltd~~Pharmaceutical Inc. ~~10 Bloomsbury Way~~ ~~London WC1A 2SL~~	12

Section No./Title	Revision	Rationale
	105 Digital Drive Novato, CA 94949 PI (office) PI (mobile) PI	
11/Appropriateness of Measurements	The chromogenic substrate FVIII assay and the one-stage clotting FVIII assay are both validated and utilize CE marked reagents. ~~The exploratory FVIII activity assay will be used for exploratory purposes only.~~	19
12.2/Screening Visit(s)	Screening assessments should be performed within ~~28~~42 days of BMN 270 infusion ~~(and must be performed within 42 days prior to BMN 270 infusion).~~, while baseline assessments will take place within 7 days prior to BMN 270 infusion (Day 1). Should the screening visit occur within 30 days of the drug infusion, physical examination, vital signs, blood chemistry, LTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline. During the first part of the Screening Period (Day -42 to Day -29), testing for AAV5 TAb titers using the ARUP screening assay may be performed, so that subjects can verify their AAV5 TAB status. Subjects who agree to participate in this activity will be asked to sign a separate ICF documenting this decision. ~~Blood sample for AAV5 total antibody (TAb) assay~~ ~~Screening sample will be tested in an AAV5 Tab pre-screening assay specifically developed for enrolment purposes.~~	7, 16
12.2.1/"Smart Rescreening" Visit	• AAV5 ~~Total Antibody~~TAb assay (ARUP) ○ ~~Smart Re-screening sample will be tested in an AAV5 Tab pre-screening assay specifically developed for enrolment purposes.~~	7, 19
12.3/Baseline Visit	• Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1) ○ ABO blood typing assessment should be performed ~~as part of the hematology assessment (~~at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study~~).~~. ~~• Direct Thrombin test~~	5, 19
12.4/Treatment Visit	• ~~Blood sample for~~ AAV5 TAb assay (ARUP) (sample collected pre-infusion for analysis) ○ ~~Infusion Day sample will be tested in an AAV5 Tab pre-screening assay specifically developed for enrolment purposes.~~ Subjects should also start prophylactic corticosteroids, prior to BMN 270 infusion, on Study Day 1 (refer to Table 9.1.6 for a possible prophylactic corticosteroid regimen, and to Section 9.4.8.2 further discussion). Clinical judgment, weighting the potential risks and benefits of corticosteroid treatment, should always be exercised when	1, 7

Section No./Title	Revision	Rationale
	considering adjustment of corticosteroid doses. Discussions between the Investigator and Medical Monitor are advised for any questions or concerns.	
12.5/BMN 270 Infusion Follow-Up Visits	At applicable sites, certain study assessments may be performed by a mobile nursing (MN) professional at the patient's home or another suitable location, such as their school or office, to improve access and convenience for patients participating in the study. The Sponsor may select a healthcare company that will be responsible for providing MN services for participating sites (the MN vendor). The MN vendor is responsible for ensuring that all MN professionals are licensed, qualified, and in good standing, as per applicable regulations, and that appropriate background checks have been performed. If the investigator at a participating site determines that MN services are appropriate for a patient and the patient gives written informed consent to participate in MN visits, the MN network will communicate with the patient and the patient's site. Potential MN visits are designated in the Schedules of Events.	8

12.5.2.1/Once per week	• Brief physical examination (complete physical examination at Week 26)
	  ○ For visits where a MN service is being used or a lab draw-only site visit is conducted, physical examination will not be performed.
	• Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
	  ○ For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.
	• Vital Signs
	  ○ For visits where a MN service is being used or a lab draw-only site visit is conducted, vital signs will not be performed.
		8

12.5.2.3/ Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26	Collection to occur until at least 3 consecutive ~~negative~~ results below the limit of detection are obtained. Semen samples should continue to be collected and tested through Week 12, even if 3 consecutive ~~negative~~ results below the limit of detection in that compartment have been recorded prior to that time point.	4
	~~Weeks 13 and 26~~ ~~The following procedure will be performed at Weeks 13 and 26:~~ • ~~Direct thrombin activity test~~	5
12.5.3/Post-Infusion Follow-Up	~~Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 26 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 26 of the study by~~	3

Section No./Title	Revision	Rationale
	~~attending only the Week 36, 44, and 52 visits during the remainder of Year 1. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in all fluids must still provide samples at Week 32, Week 40, and Week 48, if necessary (but do not need to do other scheduled assessments on that date).~~	

12.5.3.1/Every Visit	• Physical examination
	  ○ Brief physical examination should be performed at all weeks except Week 52, when a complete physical examination should be performed
	  ○ For visits where a MN service is being used or a lab draw-only site visit is conducted, physical examination will not be performed.
	• Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
	  ○ For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.
	• Vital Signs
	  ○ For visits where a MN service is being used or a lab draw-only site visit is conducted, vital signs will not be performed.
		3

12.5.3.3/ Weeks 32, 36, 44, and 52	• Weight • Blood chemistry, hematology, and coagulation tests (refer to Table 9.4.8.2.1) • FVIII antibody titer • ~~AAV5 TAb Assay~~ • ~~AAV5 TI Assay~~	16
12.5.3.4/ Weeks 32, 36, 40, 44, 48, and 52	At Weeks 32, 36, 40, 44, 48, and 52, the following procedures will be performed: • AAV5 TAb Assay • AAV5 TI Assay • PCR of vector DNA in blood, saliva, urine, semen, and stools ○ Sample testing to occur until at least 3 consecutive ~~negative~~ sample below the limit of detection results have been obtained. Subjects who have not had 3 consecutive ~~negative~~ semen samples below the limit of detection by Week 52 should continue to have PCR testing of semen every 4 weeks until 3 consecutive ~~negative~~ samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).	4, 16
12.5.3.5/ Week 36 and 52	At Weeks 36 and 52, the following procedures will be performed:	5

Section No./Title	Revision	Rationale
	• ~~Direct Thrombin test~~	
12.6/ Post-Infusion Follow-Up – Years 2-5	Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every ~~4 weeks (during Year 2) or every~~ 6 weeks ~~(~~during Years ~~3~~2-5~~)~~ until vector shedding has cleared (but do not have to perform other scheduled assessments at those study visits).	18
12.6/ Post-Infusion Follow-Up – Years 2-5	~~Year 2 – Every 4 Weeks (not required for treatment failure)~~ ~~During Year 2, every 4 weeks (± 2 weeks), the following procedures will be performed:~~ • ~~Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)~~ • ~~Liver Tests (refer to)~~ o ~~LTs may be monitored more or less frequently (and in particular when ALT values are ≥ 1.5x ULN or > ULN & > 2x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.~~ • ~~FVIII Assays~~ o ~~FVIII activity level (chromogenic substrate FVIII assay)~~ o ~~FVIII activity level (one-stage clotting FVIII assay)~~ o ~~FVIII coagulation activity exploratory assay~~ o ~~Bethesda assay (with Nijmegen modification) for FVIII inhibitor level~~ o ~~FVIII protein assay~~ • ~~PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)~~ o ~~Sample testing during Year 2 is not required if at least 3 consecutive samples are negative during the Post-Infusion Follow-Up period in Weeks 1-52. Subjects who have not had 3 consecutive negative semen samples by Week 52 should continue to have PCR testing of semen every 4 weeks during Years 2 until 3 consecutive negative samples are documented (or upon consultation between the Investigator and Medical Monitor).~~ o ~~Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every 4 weeks during Year 2 until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints).~~ ~~Years 3-5 – Every 6 Weeks (not required for treatment failure)~~	3, 4, 5, 8, 18, 19

Section No./Title	Revision	Rationale

	Years 2-5 – Every 6 Weeks (not required for treatment failure)
	During Years ~~3~~2-5, every 6 weeks (± 2 weeks), the following procedures will be performed:
	• Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
	  ○ For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.
	• PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)
	  ○ Sample testing during Years ~~3~~2-5 is not required if at least 3 consecutive samples are clear by the end of Year 2. Subjects who have not had 3 consecutive ~~negative~~ semen samples below the limit of detection by the end of Year ~~2~~1 should continue to have PCR testing of semen every 6 weeks during Years ~~3~~2-5 until 3 consecutive ~~negative~~ samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).
	  ○ Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from all fluids must still provide samples for assessment every 6 weeks during Years ~~3~~2-5 until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints).
	• FVIII Assays
	  ○ FVIII activity level (chromogenic substrate FVIII assay)
	  ○ FVIII activity level (one-stage clotting FVIII assay)
	  ○ FVIII coagulation activity exploratory assay
	  ○ Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
	  ▪ If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years ~~3~~2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.
	• VWF:Ag
	• ~~Direct Thrombin test~~
	• TGA Assay
	• PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)
	  ○ Sample testing during Years 2-5 is not required if at least 3 consecutive samples are ~~negative~~below the limit of detection during the Post-Infusion Follow-Up period in Weeks 1-52. Subjects who have not had 3 consecutive ~~negative~~ semen samples below the limit of detection	

Section No./Title	Revision	Rationale
	by Week 52 should continue to have PCR testing of semen every ~~4 weeks (during Year 2) or every~~ 6 weeks ~~(~~during Years ~~3~~2-5~~)~~ until 3 consecutive ~~negative~~ samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).	
12.7/Early Termination Visit	~~• Direct Thrombin test~~	5
Section 15	Data ~~REVIEW BOARD~~monitoring committee The Data ~~Review Board (DRB~~Monitoring Committee (DMC) will consist of ~~the Principal Investigators~~experts in clinical trials, statistics, and ~~Sponsor's Medical Monitor, as well as~~ hemophilia ~~expert advisors experienced in the conduct and evaluation of safety and efficacy parameters from 270-201.~~. The ~~DRB~~DMC will review available safety and efficacy data during the study on an ongoing basis to determine, based on emerging data and the benefit/risk profile, whether further enrollment should continue. ~~The DRB will meet weekly until all subjects have completed Week 26, and then monthly thereafter.~~ Duties of the ~~DRB~~DMC include:… Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on ~~DRB~~DMC determination of safety and efficacy….	13
Section 17/Case Report Forms and Source Documents	Before a subject's eCRF casebook can be locked, data fields must be source data verified and all queries closed. Refer to the Study Monitoring Plan for details on which fields must be source data verified. The Data Manager, or designee, will set the status of the forms, visits, and the entire casebook to Locked. The Investigator will then electronically sign the casebook, specifying that the information on the eCRFs is accurate and complete. ~~The Data Manager, or designee, will then set the status of the forms, visits, and the entire casebook to Locked.~~ Upon completion of the CSR, an electronic copy of each site's casebooks will be copied to a compact disk (CD) and sent to each site for retention with other study documents.	19
21/References	Boutin S, Monteilhet V, Veron P et al. Prevalence of serum IgG and neutralizing factors against adeno-associated virus (AAV) types 1, 2, 5, 6, 8, and 9 in the healthy population: implications for gene therapy using AAV vectors. Hum Gen Ther. 2010;21:704-712. Hayes G, Andreeva T, Gregg K, Klamroth R et al. Global seroprevalence of pre-existing immunity against various AAV serotypes in the hemophilia A population. International Society on Thrombosis and Haemostasis (ISTH) 2019 XXVII Congress. Presentation. Nathwani AC, Reiss U, Tuddenham E, et al. Adeno-Associated Mediated Gene Transfer for Hemophilia B:8 Year Follow up and Impact of Removing "Empty Viral Particles" on Safety and Efficacy of Gene Transfer. Blood. 2018;132:491.	19

BIOMARIN®

CLINICAL STUDY PROTOCOL

Study Title:	A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5
Protocol Number:	270-203
Active Investigational Product:	AAV5-hFVIII-SQ
European Union Drug Regulating Authorities Clinical Trials (EudraCT) Number:	2017-000662-29
Indication:	Hemophilia A
Sponsor:	BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949
Development Phase:	Phase 1/2
Sponsor's Responsible Medical Monitor:	PI , MA MB BChir MSc PI Clinical Sciences BioMarin (UK) Ltd. 10 Bloomsbury Way London, WC1A 2SK
Study Design:	Single-arm, open-label
Duration of Subject Participation:	Up to 264 weeks
Dose:	6E13 vg/kg as single infusion
Study Population:	Males ≥ 18 years of age with severe hemophilia A and detectable pre-existing antibodies against AAV5 vector capsid
Date of Original Protocol:	29 September 2017
Date of Amendment 1:	5 October 2018
Date of Amendment 2:	4 October 2019
Date of Amendment 3:	24 August 2020

CLINICAL STUDY PROTOCOL AMENDMENT SUMMARY

Amendment 3

Date: 24 August 2020

RATIONALE AND SUMMARY OF CHANGES

A summary of major changes covered by Amendment 3 to the 270-203 protocol is provided below:

1. Screening testing for COVID-19 has been added.

Rationale: Exclusion criteria for the study already included a provision excluding subjects with an active infection. In light of the emergence of the novel coronavirus SARS-CoV-2, the criterion has been modified to specify that this includes testing for active COVID-19 infection, which has been added to the Screening visit in the Schedule of Activities.

2. Complement panel details have been updated.

Rationale: Current protocol language says that blood samples will be tested for C3, C3a, C4, C5, and C5a complement levels in case of suspected hypersensitivity or adverse drug reaction. The revised language adds Bb and substitutes sC5b-9 for C5 and C5a, which brings the 270-203 protocol into harmony with more recent BMN 270 study protocols. The breadth of additional complement markers allows delineation of the classical and alternative/lectin mediated pathways of activation.

3. Additional complement panel assessments have been added during the first 12 weeks post-BMN 270 infusion.

Rationale: This change both aligns with more recent BMN 270 study protocols and is of particular importance in the seropositive subjects being treated in this study. Complement panel assessments will be done on the infusion day (within 2 hours after infusion), Day 2, 4, and 8, and then every other week between Week 2 and Week 12. This change permits assessment of potential innate immune responses at earlier timepoints post-BMN 270 infusion, which is of particular relevance to the AAV5+ population being evaluated in this study. The breadth of markers is the revised panel is designed to delineate between the classical and alternative or lectin-based pathways of complement activation.

4. The occurrence of events of Hy's law has been added as an event of special interest (EOSI) for purposes of expedited safety reporting, and additional safety monitoring in the event of a case potentially meeting Hy's law criteria has been added.

Rationale: Events potentially meeting the criteria for Hy's law involve combined assessment of elevations in aminotransferases and total bilirubin levels, while the current list of EOSI

focuses on elevations in aminotransferases. To date, no events meeting the criteria for Hy's law have been reported in any BMN 270 study. While monitoring for events of Hy's law has been ongoing as part of routine pharmacovigilance in all BMN 270 studies, this change ensures that the occurrence of any events in the future will be reported in an expedited manner. In addition, expanded laboratory monitoring (to include albumin and PT/INR) has been added to the guidelines for evaluating potential Hy's law cases.

5. Language has been added concerning the use of liver biopsy sample information for biopsies performed for safety-related reasons.

Rationale: Where a biopsy has been taken for safety-related reasons or was available from a past procedure, the Sponsor may want to see that biopsy information to help evaluate the impact of BMN 270 on the liver. The Sponsor may request that slides from a liver biopsy be made available for additional histopathological review.

6. FibroScan has been added as an alternative to liver ultrasound at Screening, and a fasting FibroTest has been added to the Day 1 (infusion day) assessments.

Rationale: This change aligns baseline liver health assessments with more recent BMN 270 study protocols.

7. Clarifying language has been provided for circumstances where a positive vector shedding sample occurs after 3 consecutive tests below the limit of detection have already been obtained.

Rationale: The protocol did not previously specify whether testing should be restarted after a positive result occurs after 3 consecutive results below the limit of detection in a matrix have been obtained. While this situation would be expected to be rare, and usually subjects remain below the limit of detection after having achieved 3 results below the limit of detection in a row, in the instance where a positive test occurs then testing should restart and continue until an additional 3 consecutive results below the limit of detection have been obtained. The purpose of the testing is to declare vector clearance, and in the instance where a positive test occurs even after 3 tests below the limit of detection, clearance cannot be confirmed without further testing.

8. The prohibition on the use of non-corticosteroid systemic immunosuppressive agents following BMN 270 dosing has been removed.

Rationale: The intention of the language was to prohibit use of non-steroidal systemic immunosuppressive agents within 30 days before the BMN 270 infusion, in line with the study exclusion criteria. Following dosing with BMN 270, non-steroidal systemic immunosuppressive agents may be used, following a discussion between the Investigator and the Medical Monitor, if corticosteroid use for the treatment of elevated hepatic transaminases

has been clinically deemed to be ineffective, not tolerated, and/or contraindicated by the Investigator.

9. Guidance for the monitoring and management of elevated hepatic transaminases has been modified.

Rationale: To reflect observations and data gathered from this study and other BMN 270 studies, the threshold for suggested increase in monitoring of hepatic transaminases and for reporting as an EOSI has been changed from ≥ 1.5x ULN or > ULN & > 2x baseline value to > ULN or ≥ 1.5x baseline value. A similar change has been made to the definition of an event of special interest related to elevated transaminases. Guidance has also been added for investigators to provide counselling and support to study subjects regarding the side effects of corticosteroids. This change should also encourage earlier treatment with corticosteroids, which could help decrease the extent of increases in ALT and better preserve FVIII expression.

10. An optional monthly phone check-in has been added during Years 2-5 for subjects who are returning to the site only every 12 weeks due to poor FVIII response following BMN 270 infusion.

Rationale: To ensure timely safety monitoring and promote subject retention, subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.

11. The option to assess an adverse event as related/not related to corticosteroids or other systemic immunosuppressive agents has been added.

Rationale: AEs associated with corticosteroids or other systemic immunosuppressive agents (if used) are possible and should be noted as such on the eCRF (and for safety monitoring and risk:benefit assessment purposes).

12. Lamivudine has been removed as a prohibited medication.

Rationale: Lamivudine was added as a prohibited medication after an HIV-positive subject in a BMN 270 study developed severe ALT elevations while receiving anti-retroviral therapy that included lamivudine as one of its components (and out of concern that lamivudine might be interacting with BMN 270 to exacerbate ALT elevations). However, after discussion with a liver health advisory board, lamivudine is not viewed as a likely medication that would interact with BMN 270 and, as such, should no longer be listed as a prohibited medication.

13. The requirements around the use of mobile nursing (MN) services to conduct unscheduled visits for assessment of FVIII levels or liver tests (LTs) have been clarified.

Rationale: In instances where a subject's LTs have been elevated, assessment and workup of those elevations may require additional laboratory work (FVIII and LT levels) to be collected at unscheduled visits. At sites where MN services have been approved, these unscheduled laboratory tests may be performed by a MN professional, rather than requiring a site visit.

14. Updates have been made to incorporate changes previously communicated to sites through protocol clarification letters. These changes include:
    - Corrections to the timing of weight assessment footnotes in the Schedules of Activities
    - Clarification of the visit window for AAV5 TAb testing during Screening
    - Update to Section 9.7.6.6 to clarify that vector shedding should be performed every 6 weeks during Years 2-5
    - Update Section 12 to include FVIII antibody titer at Week 24 (as is reflected in the Schedule of Activities)
    - Update Section 12.5.2.7 to state that the cytokine bead array assay should be performed at Week 24 (rather than Week 22)
    - Add the cytokine bead array assay to the Early Termination Visit in Section 12 (as is reflected in the Schedule of Activities)
    - Add AAV5 TAb and TI assays to Weeks 28, 30, and 34 in the Schedule of Activities and in Section 12
15. Guidance concerning how to determine whether a subject has been lost to follow-up has been added.
16. The costs and compensation language has been updated to clarify which study-related costs will be covered by the Sponsor.
17. The summary of risks and benefits has been updated.
18. The required timepoints for vector shedding sample collection during Years 2-5 (if required) have been clarified.
19. The identity of the medical monitor has been updated.
20. Additional minor changes have been made for consistency and clarity.

Refer to Section 25 for a summary of revisions to Amendment 2 (dated 4 October 2019).

2 SYNOPSIS

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:
TITLE OF STUDY: A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5		
PROTOCOL NUMBER: 270-203		
STUDY SITES: Approximately 5-6 sites globally		
PHASE OF DEVELOPMENT: Phase 1/2		
STUDY RATIONALE: Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. The clinical phenotype of HA patients generally correlates tightly with the level of residual expression. Severe HA is classified as FVIII activity less than 1% of wild-type (< 1 IU/dL), moderate disease comprises 1-5% of wild-type activity, and the mild form is 5-40% activity. The clinical manifestations of severe HA are frequent spontaneous bleeding episodes, predominantly in joints and soft tissues, with a substantially increased risk of death from hemorrhage when the brain is involved. Treatment of severe HA presently mainly consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study (Berntorp, 2017) and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with on-demand-only therapy in the same studies, respectively. Continued repeated bleeding events compound		

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debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death.

Extended half-life products created through chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) or bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half-life by approximately 50% (half-life 18-19 hours), and thus, show promise in reduced dosing and maintaining activity levels above a 1% trough for a greater proportion of the dosing interval. However, patients with severe HA who are treated with extended half-life FVIII remain dependent on multiple infusions to maintain critical levels of FVIII activity. More recently, emicizumab has been approved for the treatment of HA. Emicizumab is a humanized, bispecific monoclonal antibody that binds to both activated coagulation factor IX (FIX) and coagulation factor X (FX), thereby mimicking the function of activated FVIII. Emicizumab represents an incremental improvement in therapeutic burden over replacement FVIII products since it can be administered subcutaneously as opposed to intravenously; however, it is still a chronic treatment that requires repeat administration every 1-4 weeks, and sustained FVIII activity levels above 5-10 IU/dL have rarely been attained even with extended half-life products. Therefore, there is a strong unmet need for a treatment that can alter the fundamental problem for patients with HA, namely attainment of sustained, hemostatically effective FVIII activity levels long-term (ie, over multiple years) without the need for treatment.

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of active FVIII following a single intravenous infusion of a vector encoding the appropriate gene sequence. Hemophilia A is well-suited for a gene replacement approach because clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in minute amounts (200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and even modest increases in the level of FVIII (any increase of the plasma level by 2 ng/ml induces an increase in activity of 1%) can ameliorate the severe form of hemophilia A. Thus, relatively small changes in endogenous FVIII activity can result in clinically relevant improvements in disease phenotype. Moreover, the circulating FVIII response to gene transduction can be assessed using validated quantitative endpoints that are easily assayed using established laboratory techniques.

Several different gene transfer strategies for FVIII replacement have been evaluated, with adeno-associated viral (AAV) vectors used in most hemophilia clinical trials. AAV vectors have an excellent and well-defined safety profile, and can direct long-term transgene expression with tropism and promoter specificity for specific tissues, such as the liver (eg serotypes 2, 5, and 8). Indeed, an ongoing gene therapy clinical trial for a related disorder, hemophilia B, has established over 7 years of stable human factor IX (hFIX) expression at levels sufficient for conversion of their bleeding phenotype from severe to moderate or mild, following a single peripheral vein infusion of AAV8-hFIX vector (Nathwani, 2018). Several participants in this trial have been able to

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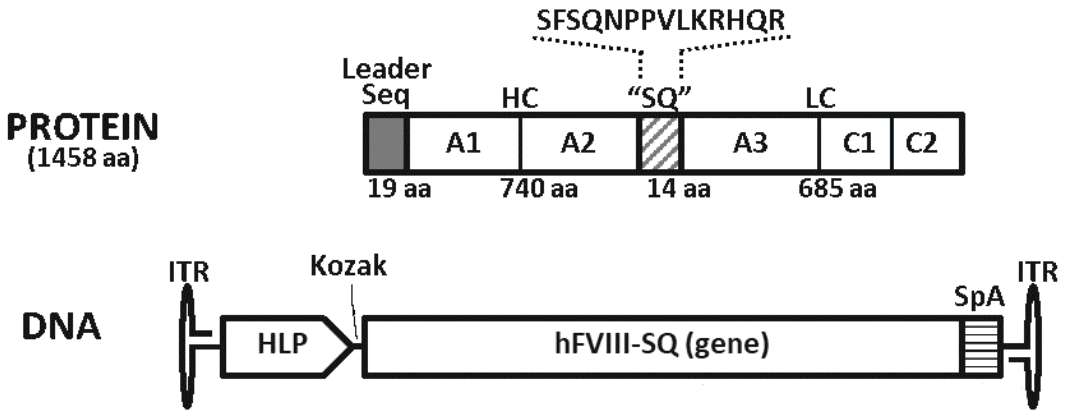
discontinue factor prophylaxis without suffering spontaneous hemorrhages, even when they undertook activities that previously resulted in bleeding. Thus, gene therapy treatment has resulted in a substantial improvement in their quality of life.

BMN 270 is an AAV5-based gene therapy vector that expresses the SQ form of hFVIII under the control of a hybrid human liver-specific promoter (Figure 1).

Figure 1: AAV5-hFVIII-SQ Vector Genome and Encoded Protein



Legend –Note that schematic is not to scale; aa = amino acids; ITR = inverted terminal repeat; HLP = human liver promoter; Kozak = Kozak concensus sequence (GCCACC); SpA = Synthetic poly(A) signal

BMN 270 is delivered by a single IV dose and is designed to achieve stable, durable expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.

BMN 270 is being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. Three-year results from 270-201 have demonstrated that following gene transfer, mean and median FVIII activity levels above 15% (15 IU/dL), as measured by a chromogenic substrate assay, are achievable and sustained following a single infusion of 6E13 vg/kg of BMN 270, with administration of prophylactic corticosteroids and an acceptable safety profile (Pasi, 2019). In addition, an interim analysis of clinical study 270-301, an ongoing phase 3 study designed to assess the efficacy and safety of BMN 270 at a dose of 6E13 vg/kg and administration of therapeutic corticosteroids (ie, in response to ALT

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elevations), demonstrated FVIII activity levels that were also well above 15%, albeit lower than what was observed for the 6E13 vg/kg cohort in 270-201 at 26 weeks (Pasi, 2019).

Subjects enrolled and infused in 270-201 screened negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes (Boutin, 2010; Hayes, 2019).

Current data describing the extent of neutralization or inhibited transduction has been inconsistent across studies, each employing one of several varying methodologies to evaluate both vector-specific antibodies and transduction inhibition (TI). Early studies using AAV2 for the treatment of hemophilia B described inhibited transduction in patients with neutralizing antibody titers of 2 or 3 (Manno, 2006), but more recent studies have described successful transduction in patients who, in retrospect, were determined to have pre-existing antibody to the vector capsid (Majowicz, 2017). Importantly, no drug-related adverse events were described in these patients. As the sensitivity of assays used to detect pre-existing antibody has increased greatly in recent years, the biological relevance of any given titer determination, and comparisons across studies using assays with differing titer sensitivities, requires additional study.

Data from a non-clinical study conducted by BioMarin in cynomolgus monkeys further demonstrated that transduction efficiency can vary greatly in the presence of pre-existing anti-AAV5 antibody. In non-human primates, the ability to attain FVIII levels in the face of measurable AAV5 antibody titers without significant safety risks supports the possibility of achieving a similar response in patients with AAV5 antibody positivity. The totality of available data shows the effect of pre-existing immunogenicity to AAV delivery vectors is not completely understood, and indicates that some patients may benefit from AAV5 gene therapy who are currently excluded.

In this study, patients with severe HA and pre-existing antibodies against AAV5 will be enrolled to evaluate the clinical response to BMN 270 gene therapy. The aim is to evaluate the safety, tolerability, and efficacy of BMN 270 in patients with severe HA and pre-existing antibodies against AAV5 vector capsid at various levels of AAV5 antibody titers.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

OBJECTIVES:

The primary objective of the study is to:

- Assess the safety of a single intravenous administration of BMN 270 in severe HA subjects with pre-existing antibody to AAV5 vector capsid, including development of FVIII neutralizing antibody

Secondary objectives of the study are to:

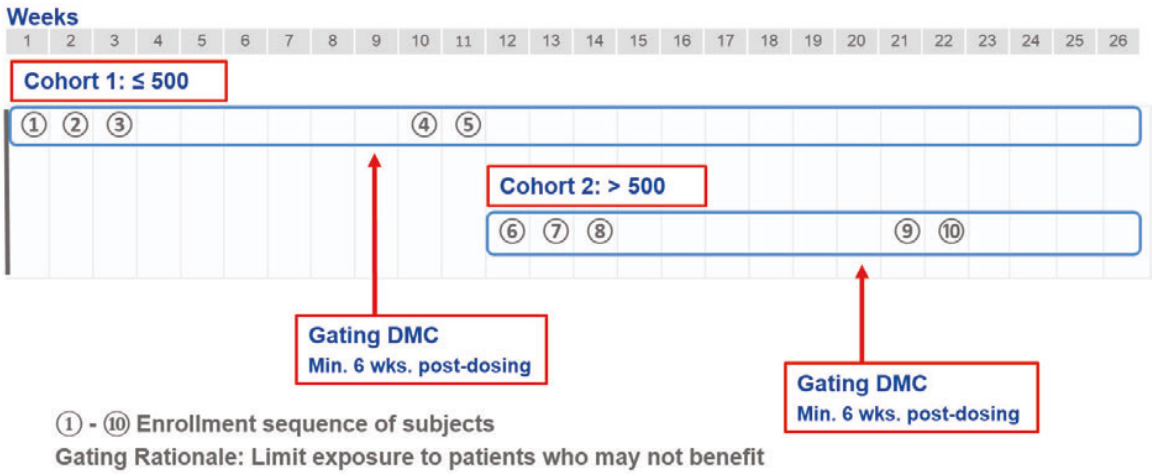
- Assess the efficacy of BMN 270 defined as FVIII activity at or above 5 IU/dL at Week 26
- Assess the impact of BMN 270 on usage of exogenous FVIII replacement therapy
- Assess the impact of BMN 270 on the number of bleeding episodes requiring exogenous FVIII therapy
- Evaluate the pharmacodynamics of FVIII expression following IV infusion of BMN 270
- Assess the impact of BMN 270 on patient-reported outcomes (PROs)

STUDY DESIGN AND PLAN:

This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to the AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects may be enrolled at 5-6 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb titer ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb titer > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date.

An independent Data Monitoring Committee (DMC) will consist of experts in clinical trials, statistics, and hemophilia. The DMC will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 2 represents one dosing schedule scenario):

Figure 2: 270-203 Dosing Schedule (One Possible Scenario)



Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from a minimum of 6 weeks post-infusion) reviewed. FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response, based on data from 270-201. Up to 6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness).

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC evaluation of safety and efficacy in treated subjects, with the following triggers that may potentially pause further enrollment:

- any related SAE;
- any related AE with a severity > CTCAE Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

Following a temporary halt of enrolment, the DMC may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed. Additional blood samples will be collected (tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine bead array (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.

Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DMC based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL. In subjects who experience recurring bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis.

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor or Sponsor-designated Data Monitor, follow an abbreviated visit schedule after Week 52 by attending only the Q12W and End of Year visits during Years 2-5. Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.

The study analysis will be performed after all subjects have been followed for 26 weeks post-BMN 270 infusion, with presentation of safety, efficacy, and FVIII PD assessments. After the safety, efficacy, and FVIII PD analyses at 26 weeks post-BMN 270 infusion, long-term safety and efficacy will be assessed in all subjects for up to a total of 5 years post-infusion. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the DMC.

NUMBER OF SUBJECTS PLANNED:

Approximately 10 subjects

DIAGNOSIS AND ALL CRITERIA FOR INCLUSION AND EXCLUSION:

Individuals eligible to participate in this study must meet all of the following criteria:

1. Males ≥ 18 years of age with hemophilia A and residual FVIII levels ≤ 1 IU/dL as evidenced by medical history, at the time of signing the informed consent.
2. Detectable pre-existing antibodies against the AAV5 vector capsid as measured by AAV5 total antibody ELISA

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

3. Treated/exposed to FVIII concentrates or cryoprecipitate for a minimum of 150 exposure days (EDs)
4. Subject must have been on prophylactic FVIII replacement therapy for at least 12 months prior to study entry.
5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures.
6. No previous documented history of a detectable FVIII inhibitor, and results from a Bethesda assay or Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) (or less than 1.0 BU for laboratories with a historical lower sensitivity cutoff for inhibitor detection of 1.0 BU) on 2 consecutive occasions at least one week apart within the past 12 months (at least one of which should be tested at the central laboratory)
7. Sexually active participants must agree to use an acceptable method of effective contraception, either double-barrier contraception (ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with viral vector DNA below the limit of detection.
8. Willing to abstain from consumption of alcohol for at least the first 52 weeks following BMN 270 infusion.

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Any evidence of active infection, including COVID-19, or any immunosuppressive disorder, including HIV infection.
2. Significant liver dysfunction with any of the following abnormal laboratory results:
    - ALT (alanine aminotransferase) > 1.25x ULN;
    - AST (aspartate aminotransferase) > 1.25x ULN;
    - GGT (gamma-glutamyltransferase) > 1.25x ULN;
    - Total bilirubin > 1.25x ULN;
    - Alkaline phosphatase > 1.25x ULN; or
    - INR (international normalized ratio) ≥ 1.4

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

Subjects whose liver laboratory assessments fall outside of these ranges may undergo repeat testing of the entire liver test panel within the same Screening window and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor.

3. Most recent, prior FibroScan or liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used.
4. Evidence of any bleeding disorder not related to hemophilia A
5. Platelet count of $< 100 \times 10^9$/L
6. Creatinine ≥ 1.5 mg/dL
7. Liver cirrhosis of any etiology as assessed by liver ultrasound/FibroScan.
8. Chronic or active hepatitis B as evidenced by positive serology testing (hepatitis B surface antigen [HBsAg], hepatitis B surface antibody [HBsAb], and hepatitis B core antibody [HBcAb]) and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results .
9. Active Hepatitis C as evidenced by detectable HCV RNA, or currently on antiviral therapy
10. Active malignancy, except non-melanoma skin cancer
11. History of hepatic malignancy
12. History of arterial or venous thromboembolic events (eg, deep vein thrombosis, non-hemorrhagic stroke, pulmonary embolism, myocardial infarction, arterial embolus), with the exception of catheter-associated thrombosis for which anti-thrombotic treatment is not currently ongoing.
13. Known inherited or acquired thrombophilia, including conditions associated with increased thromboembolic risk, such as atrial fibrillation.
14. A history of known inflammatory, connective tissue, or autoimmune disorders (eg, vasculitis).
15. Treatment with any Investigational Product within 30 days or 5 half-lives of the investigational product (whichever is longer) prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	**SUMMARY TABLE** Referring to Part of the Dossier: Volume: Page: Reference:	**FOR NATIONAL AUTHORITY USE ONLY:**

16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including corticosteroid treatment and/or the use of alternative immunosuppressive agents outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.
17. Prior treatment with any vector or gene transfer agent
18. Major surgery planned in the 52-week period following the infusion with BMN 270
19. Use of systemic immunosuppressive agents, not including corticosteroids, or live vaccines within 30 days before the BMN 270 infusion
20. Concurrent enrollment in another clinical study, unless it is an observational (non-interventional) clinical study that does not interfere with the requirements of the current protocol or have the potential to impact the evaluation of safety and efficacy of BMN 270 and with prior consultation with the Medical Monitor
21. Known allergy or hypersensitivity to investigational product formulation
22. Unwilling to receive blood or blood products for treatment of an adverse event and/or a bleed

INVESTIGATIONAL PRODUCT(S), DOSE, ROUTE AND REGIMEN:
Each subject will receive a single IV infusion of BMN 270 at 6E13 vg/kg. The volume of infusion will depend on the subject's weight.

REFERENCE THERAPY(IES), DOSE, ROUTE AND REGIMEN:
No reference therapy will be evaluated in this study.

DURATION OF TREATMENT:
BMN 270 is given as a single dose by IV infusion.

CRITERIA FOR EVALUATION:

Safety:

The following safety outcome measurements will be assessed:

- Incidence of adverse events (AEs), including serious AEs (SAEs)
- Change in clinical laboratory tests (serum chemistry and hematology)
- Change in vital signs
- Change in physical examination
- Vector shedding (blood, urine, semen, feces, saliva)

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

- Liver tests (LTs, including ALT, AST, GGT, LDH, total bilirubin, and alkaline phosphatase)
- Immune response to FVIII transgene product and AAV5 vector capsid

No major toxicity is expected based on 270-201 data and non-clinical studies. Each subject will have comprehensive surveillance monitoring of LTs (once per week for Weeks 1-26). During the long-term safety evaluation, LTs will be monitored every three months for up to 5 years post-infusion; the frequency and duration of LT testing may be changed based on discussion between the Medical Monitor and the Investigator and review of subject data.

There will be a detailed assessment of cellular and humoral responses to AAV5 vector capsid and FVIII.

Efficacy:

The efficacy measure will be to assess plasma FVIII activity. The efficacy goal is to achieve FVIII activity ≥ 5 IU/dL at 26 weeks post-BMN 270 administration. Other efficacy measures include assessing the impact of BMN 270 on the use of FVIII replacement therapy and on the number of bleeding episodes during the study. Subjects will be asked to keep a subject diary, provided by the sponsor, to record the relevant details.

Other efficacy endpoints:

- Change from baseline in the total score of HAEMO-QoL-A at Week 26 of the study post-BMN 270 infusion
- Change from baseline in the EQ-5D-5L score at Week 26 of the study post-BMN 270 infusion.
- Change from baseline in the Haemophilia Activities List (HAL) score at Week 26 of the study post-BMN 270 infusion.
- Change from baseline in the Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) score at Week 26 of the study post-BMN 270 infusion.

Pharmacodynamics:

The FVIII antigen and activity level, as measured by a validated immunoassay and a validated FVIII activity assay, respectively, will be used for plasma profiles; FVIII antigen and activity will be used to determine PD parameters.

STATISTICAL METHODS:

Approximately 10 subjects may be dosed in the study. The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:
longitudinal methods, such as a mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at ≥ 5 IU/dL at 26 weeks following study treatment. FVIII activity will be assessed weekly during the study period. Assessment of the true steady state of FVIII will require that FVIII activity is measured after a minimum of 72 hours (or 5x the known half-life of the FVIII replacement therapy administered) has elapsed since the last infusion of FVIII concentrates. Analysis of neutralizing antibody response, other immunological parameters, and vector shedding will be primarily descriptive and involve both inter-subject and intra-subject comparisons across cohorts.		

3 TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

4 LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviations

AAV	adeno-associated virus
ABR	annualized bleeding rate
ADL	activities of daily living
ADR	adverse drug reaction
AE	adverse event
ALT	alanine aminotransferase
APTT	activated partial thromboplastin time
ART	anti-retroviral therapy
AST	aspartate aminotransferase
BPV	BioMarin Pharmacovigilance
BU	Bethesda Unit
CFR	Code of Federal Regulations
CRA	clinical research associate
CRF	case report form
CSR	clinical study report
CTCAE	Common Terminology Criteria for Adverse Events
DMC	Data Monitoring Committee
EC	ethics committee
eCRF	electronic case report form
ED	exposure days
EOSI	events of special interest
ETV	early termination visit
EudraCT	European Union Drug Regulating Authorities Clinical Trials
FDA	Food and Drug Administration
FIH	first-in-human
FVIII	coagulation factor VIII
GCP	Good Clinical Practice
GGT	gamma-glutamyltransferase
HA	Hemophilia A
HAL	Haemophilia Activities List
HBcAb	hepatitis B core antibody
HBsAb	hepatitis B surface antibody
HBsAg	hepatitis B surface antigen

hFIX	human coagulation factor IX
hFVIII	human coagulation factor VIII
HIPAA	Health Insurance Portability and Accountability Act
IB	investigator brochure
ICF	informed consent form
ICH	International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
ICH E6 [R2]	ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6
IND	Investigational New Drug (application)
INR	international normalized ratio
IP	investigational product
IRB	institutional review board
IV	intravenous
LT	liver test
MedDRA	Medical Dictionary for Regulatory Activities
MN	mobile nursing
PBMC	peripheral blood mononuclear cells
PCR	polymerase chain reaction
PD	pharmacodynamics
PEG	polyethylene glycol
PK	Pharmacokinetics
PRO	patient-reported outcome
rhFVIII	recombinant human FVIII protein
SAE	serious adverse event
SAP	statistical analysis plan
SDV	source data verification
SoA	schedule(s) of activities
TGA	thrombin generation assay
ULN	upper limit of normal
vg	vector genomes
VWF:Ag	von Willebrand factor Antigen
WPAI+CIQ:HS	Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific

Definition of Terms:

Investigational Product (IP):

"A pharmaceutical form of an active ingredient or placebo being tested or used as a reference in a clinical trial, including a product with a marketing authorization when used or assembled (formulated or packaged) in a way different from the approved form, or when used for an unapproved indication, or when used to gain further information about an approved use" (from International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6 [ICH E6] (R2)]).

The terms "IP" and "study drug" may be used interchangeably in the protocol.

5 ETHICS

BioMarin Pharmaceutical Inc. (hereafter referred to as BioMarin or the Sponsor) conducts its studies according to the highest ethical and scientific standards. The following Sections articulate standards to which Investigators will be held accountable, as well as matters of compliance to document adherence to such standards.

5.1 Institutional Review Board or Independent Ethics Committee

Investigators are expected to interact with Ethics Committees (ECs) promptly, as required, during the course of the study. This includes, but is not limited to, providing appropriate documentation to support study initiation and maintaining appropriate flow of safety and other information during the course of the study and for study close-out activities. BioMarin (or designee) will assist Investigators with access to timely and accurate information and with assurance of prompt resolution of any queries.

Prior to initiating the study, the Investigator will obtain written confirmation that the institutional review board (IRB) or independent ethics committee (EC) is properly constituted and compliant with International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) and Good Clinical Practice (GCP) requirements, applicable laws, and local regulations. A copy of the confirmation from the IRB/EC will be provided to BioMarin Pharmaceutical Inc. (BioMarin) or its designee. The Investigator will provide the IRB/EC with all appropriate material, including the protocol, Investigator's Brochure (IB), the Informed Consent Form (ICF) including compensation procedures, and any other written information provided to the subjects, including all ICFs translated for patients who do not speak the local language at the clinical site. The study will not be initiated and Investigational Product (IP) supplies will not be shipped to the site until appropriate documents from the IRB/EC confirming unconditional approval of the protocol, the ICF and all subject recruitment materials are obtained in writing by the Investigator and copies are received at BioMarin or its designee. The approval document should refer to the study by protocol title and BioMarin protocol number (if possible), identify the documents reviewed, and include the date of the review and approval. BioMarin will ensure that the appropriate reports on the progress of the study are made to the IRB/EC and BioMarin by the Investigator in accordance with applicable guidance documents and governmental regulations.

5.2 Ethical Conduct of Study

It is expected that Investigators understand and comply with the protocol. This includes, but is not limited to: establishing and meeting enrollment commitments, including providing

eligible subjects for study enrollment; adhering to adverse event reporting, diagnostic, or other procedures as specified in the protocol; and assuring appropriate compliance with study treatment administration and accountability.

This study will be conducted in accordance with the following:

- European Clinical Trial Directive 2001/20/EC and Good Clinical Practice Directive 2005/28/EC, for studies conducted within any European country
- US Code of Federal Regulations (CFR) Sections that address clinical research studies, and/or other national and local regulations, as applicable
- ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6(R2) (ICH E6R2)
- The ethical principles established by the Declaration of Helsinki

Specifically, this study is based on adequately performed laboratory and animal experimentation. The study will be conducted under a protocol reviewed and approved by an IRB/EC and will be conducted by scientifically and medically qualified persons. The benefits of the study are in proportion to the risks. The rights and welfare of the subjects will be respected and the investigators conducting the study do not find the hazards to outweigh the potential benefits. Each subject will provide written, informed consent before any study-related tests or evaluations are performed.

5.3 Subject Information and Informed Consent

A properly written and executed informed consent form (ICF), in compliance with ICH E6R2 (Section 4.8), United States Code of Federal Regulations (CFR) 21 CFR §50, European Clinical Trial Directive 2001/20/EC and Good Clinical Practice Directive 2005/28/EC, and other applicable local regulations, will be obtained for each subject prior to entering the subject into the study. The Investigator will prepare the ICF and provide the documents to BioMarin for approval prior to submission to the IRB/EC approval. BioMarin and the IRB/EC must approve the documents before they are implemented. A copy of the approved ICF and if applicable, a copy of the approved subject information sheet and all ICFs translated to a language other than the native language of the clinical site must also be received by BioMarin or designee prior to any study-specific procedures being performed.

6 INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

During administration of informed consent, expectations regarding participation in the study should be made clear to subjects. Subjects who are not willing and/or are not able to comply with all aspects of the study should not be encouraged to participate.

Prior to beginning the study, the Investigator at each site must provide to BioMarin or designee a fully executed and signed Statement of Investigator (SOI) form. A US Food and Drug Administration (FDA) Form FDA 1572 serves as an acceptable SOI form. If Form FDA 1572 may not be used in a particular region, the Investigator must provide a fully executed SOI on the form provided by the Sponsor. All Investigators and Sub-Investigators must be listed on Form FDA 1572 or its equivalent SOI. Financial Disclosure Forms must also be completed for all Investigators and Sub-Investigators listed on the Form FDA 1572 or SOI who will be directly involved in the treatment or evaluation of subjects in this study.

The study will be administered by and monitored by employees or representatives of BioMarin. Clinical Research Associates (CRAs) or trained designees will monitor each site on a periodic basis and perform verification of source documentation for each subject as well as other required review processes. BioMarin's Regulatory Affairs Department (or designee) will be responsible for the timely reporting of serious adverse events (SAEs) to appropriate regulatory authorities as required.

In multicenter studies, a Coordinating Investigator will be identified who will be responsible for study overview. The Coordinating Investigator will read the clinical study report (CSR) and confirm that it accurately describes the conduct and results of the study, to the best of his or her knowledge. The Coordinating Investigator will be chosen on the basis of active participation in the study, ability to interpret data, and willingness to review and sign the report in a specified timeframe. The identity of the Coordinating Investigator and a list of all Investigators participating in the study will be provided in the CSR.

Clinical Laboratory assessments will be performed at a nominated central laboratory. Bioanalytical samples will be sent to the appropriate specialty laboratories for testing. Refer to laboratory manual for more details.

7 INTRODUCTION

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males (Nathwani, 1992). It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. Clinical manifestations of severe FVIII deficiency are frequent unprovoked bleeding episodes in joints and soft tissues causing permanent disability and occasionally death mostly after brain hemorrhage. Treatment in Western countries (Berntorp, 2012) consists of intravenous injection of plasma-derived or recombinant FVIII protein concentrates at the time of a bleed to control it or prophylactically to prevent bleeding episodes. The short half-life for FVIII (12-18 hours) necessitates frequent infusions and makes this treatment prohibitively expensive for the majority of the world's hemophilia A patients. These individuals develop debilitating arthropathy and have a substantially increased risk of death from hemorrhage in life (Stonebraker, 2010). Chemical modification or bioengineering of FVIII may improve half-life to 18-19 hours (Kaufman, 2013). However, these extended half-life FVIII variants do not eliminate the need for lifelong FVIII protein administration (Hay, 2012).

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of human FVIII (hFVIII) following a single administration of vector. Hemophilia A is well-suited for this approach because its clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in low amounts (100-200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and a modest increase in the level of FVIII (a plasma level of 2 ng/ml protein leads to a 1% expression) can ameliorate the severe phenotype (Srivastava, 2013); thus, the therapeutic goal for gene therapy is a modest increase in hFVIII. Finally, the consequences of gene transfer can be assessed using validated quantitative endpoints that can be easily assayed in most clinical laboratories.

BMN 270 contains the cDNA for the B-domain-deleted SQ FVIII with a liver-specific HLP transcription promoter. The expression cassette is inserted between AAV2 ITRs, and this genome is packaged in the AAV5 capsid. A comprehensive review of BMN 270 is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.1 Nonclinical Studies

The nonclinical program supports a single IV infusion of BMN 270, the planned clinical route of administration, for the treatment of hemophilia A in male patients. This nonclinical program took into account the guidelines and reflection papers for gene therapy medicinal products under EMA Advanced Therapies as well as FDA guidance. The primary pharmacodynamics (PD), pharmacokinetics (PK), and toxicity of IV BMN 270 were characterized in a series of single dose studies in species that were vector permissive and responsive to the transgene including normal CD-1 mice, a B- and T-cell deficient mouse model of hemophilia A (B6;129S-*F8*^{tm1Kaz}/J x B6.129S6-*Rag2*^{tm1Fwa} N12; FVIII KO x Rag2), and normal cynomolgus and rhesus monkeys. Some PD studies evaluated additional PK, immunogenicity and toxicity endpoints.

The comparative pharmacodynamics of BMN 270 in cynomolgus monkeys with varying pre-existing AAV5 transduction inhibition (TI) titer and AAV5 TAb status was evaluated in study BMN270-16-021. BMN 270 was administered to 4 groups of monkeys, a control group (Group 1, n=3) that tested negative for both TI and AAV5 TAb, Group 2 (n=4) that was AAV5 TAb negative, and low TI titer (2-5) positive. Group 3 (n=4) was also AAV5 TAb negative, but had higher TI titers (5-10). Group 4 (n=5) tested positive for both AAV5 TAb and TI (TI titers were >5). Administration of BMN 270 by a single intravenous bolus injection was well-tolerated in cynomolgus monkeys regardless of baseline TI titer or TAb status. After dosing, all monkeys showed FVIII-SQ levels above the LLOQ, with the exception of two monkeys in the group that presented with both positive TI and TAb titers at baseline. Though these TAb+ monkeys, regardless of TI titers, showed a significant mean reduction in FVIII expression (68% less) compared to TAb negative monkeys, three of five monkeys showed detectable levels of FVIII-SQ, with one having levels similar to that observed in the TI and TAb negative control group. Monkeys that were TI+ but TAb-at baseline had FVIII expression levels that were similar to those of the TI and TAb negative control group.

Results of the nonclinical program to date are detailed in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.2 Previous Clinical Studies

Ongoing clinical studies for BMN 270 include:

- 270-201, a phase 1/2, dose-escalation study in patients with severe HA
- 270-203, a phase 1/2 study in patients with severe HA who have anti-AAV5 antibody titers

- 270-301, a phase 3 study in patients with severe HA who receive BMN 270 at the 6E13 vg/kg dose level
- 270-302, a phase 3 study in patients with severe HA who receive BMN 270 at the 4E13 vg/kg dose level

A comprehensive review of safety, efficacy, and immunogenicity results from 270-201 and 270-301 as of the latest data cut is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.3 Study Rationale

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. The clinical phenotype of HA patients generally correlates tightly with the level of residual expression. Severe HA is classified as FVIII activity less than 1% of wild-type (< 1 IU/dL), moderate disease comprises 1-5% of wild-type activity, and the mild form is 5-40% activity. The clinical manifestations of severe HA are frequent spontaneous bleeding episodes, predominantly in joints and soft tissues, with a substantially increased risk of death from hemorrhage when the brain is involved.

Treatment of severe HA presently mainly consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study (Berntorp, 2017) and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with on-demand-only therapy in the same studies, respectively. Continued repeated bleeding events compound debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death.

Extended half-life products created through chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) or bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half-life by approximately 50% (half-life 18-19 hours), and thus,

show promise in reduced dosing and maintaining activity levels above a 1% trough for a greater proportion of the dosing interval. However, patients with severe HA who are treated with extended half-life FVIII remain dependent on multiple infusions to maintain critical levels of FVIII activity. More recently, emicizumab has been approved for the treatment of HA. Emicizumab is a humanized, bispecific monoclonal antibody that binds to both activated coagulation factor IX (FIX) and coagulation factor X (FX), thereby mimicking the function of activated FVIII. Emicizumab represents an incremental improvement in therapeutic burden over replacement FVIII products since it can be administered subcutaneously as opposed to intravenously; however, it is still a chronic treatment that requires repeat administration every 1-4 weeks, and sustained FVIII activity levels above 5-10 IU/dL have rarely been attained even with extended half-life products. Therefore, there is a strong unmet need for a treatment that can alter the fundamental problem for patients with HA, namely attainment of sustained, hemostatically effective FVIII activity levels long-term (ie, over multiple years) without the need for treatment.

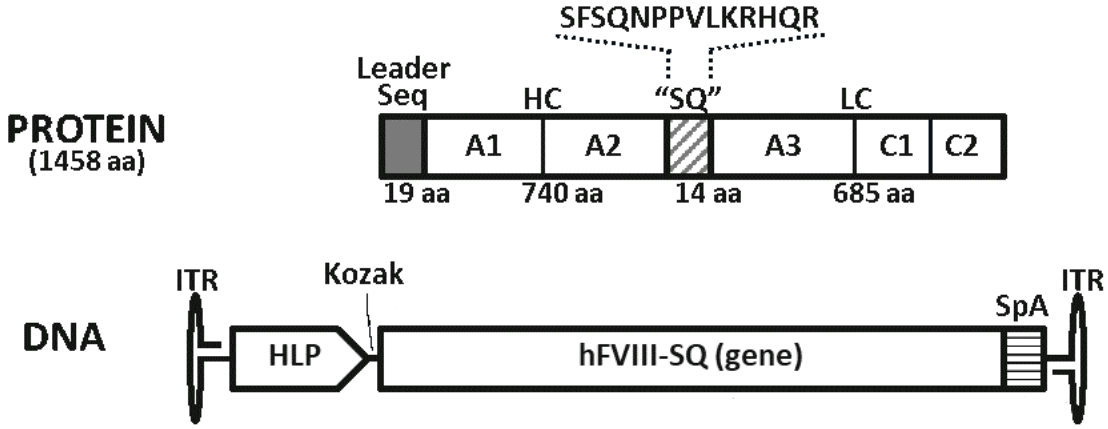
Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of active FVIII following a single intravenous infusion of a vector encoding the appropriate gene sequence. Hemophilia A is well-suited for a gene replacement approach because clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in minute amounts (200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and even modest increases in the level of FVIII (any increase of the plasma level by 2 ng/ml induces an increase in activity of 1%) can ameliorate the severe form of hemophilia A. Thus, relatively small changes in endogenous FVIII activity can result in clinically relevant improvements in disease phenotype. Moreover, the circulating FVIII response to gene transduction can be assessed using validated quantitative endpoints that are easily assayed using established laboratory techniques.

Several different gene transfer strategies for FVIII replacement have been evaluated, with adeno-associated viral (AAV) vectors used in most hemophilia clinical trials. AAV vectors have an excellent and well-defined safety profile, and can direct long-term transgene expression with tropism and promoter specificity for specific tissues, such as the liver (eg serotypes 2, 5, and 8). Indeed, an ongoing gene therapy clinical trial for a related disorder, hemophilia B, has established over 7 years of stable human factor IX (hFIX) expression at levels sufficient for conversion of their bleeding phenotype from severe to moderate or mild, following a single peripheral vein infusion of AAV8-hFIX vector (Nathwani, 2018). Several participants in this trial have been able to discontinue factor prophylaxis without suffering spontaneous hemorrhages, even when they undertook activities

that previously resulted in bleeding. Thus, gene therapy treatment has resulted in a substantial improvement in their quality of life.

BMN 270 is an AAV5-based gene therapy vector that expresses the SQ form of hFVIII under the control of a hybrid human liver-specific promoter (Figure 7.3.1).

Figure 7.3.1: AAV5-hFVIII-SQ Vector Genome and Encoded Protein



Legend –Note that schematic is not to scale; aa = amino acids; ITR = inverted terminal repeat; HLP = human liver promoter; Kozak = Kozak concensus sequence (GCCACC); SpA = Synthetic poly(A) signal

BMN 270 is delivered by a single IV dose and is designed to achieve stable, durable expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.

BMN 270 is being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. Three-year results from 270-201 have demonstrated that following gene transfer, mean and median FVIII activity above 15% (15 IU/dL), as measured by a chromogenic substrate assay, are achievable and sustained following a single infusion of 6E13 vg/kg of BMN 270, with administration of prophylactic corticosteroids and an acceptable safety profile (Pasi, 2019). In addition, an interim analysis of clinical study 270-301, an ongoing phase 3 study designed to assess the efficacy and safety of BMN 270 at a dose of 6E13 vg/kg and administration of on-demand corticosteroids (ie, in response to ALT elevations), demonstrated FVIII activity levels that were also well above 15% at 26 weeks, albeit lower than what was observed for

the 6E13 vg/kg cohort in 270-201 (Pasi, 2019). For additional information on preliminary data in 270-201, refer to the current version of the Investigator's Brochure.

Subjects enrolled and infused in 270-201 were screened and negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes (Hayes, 2019; Boutin, 2010).

Current data describing the extent of neutralization or inhibited transduction has been inconsistent across studies, each employing one of several varying methodologies to evaluate both vector-specific antibodies and transduction inhibition (TI). Early studies using AAV2 for the treatment of hemophilia B described inhibited transduction in patients with neutralizing antibody titers of 2 or 3 (Manno, 2006), but more recent studies have described successful transduction in patients who, in retrospect, were determined to have pre-existing antibody to the vector capsid (Majowicz, 2017). Importantly, no drug-related adverse events were described in these patients. As the sensitivity of assays used to detect pre-existing antibody has increased greatly in recent years, the biological relevance of any given titer determination, and comparisons across studies using assays with differing titer sensitivities, requires additional study.

Data from a non-clinical study conducted by BioMarin in cynomolgus monkeys further demonstrated that transduction efficiency can vary greatly in the presence of pre-existing anti-AAV5 antibody. In non-human primates, the ability to attain FVIII levels in the face of measurable AAV5 antibody titers without significant safety risks supports the possibility of achieving a similar response in patients with AAV5 antibody positivity. The totality of available data shows the effect of pre-existing immunogenicity to AVV delivery vectors is not completely understood, and indicates that some patients may benefit from AAV5 gene therapy who are currently excluded.

In this study, patients with severe HA and pre-existing antibodies against AAV5 will be enrolled to evaluate the clinical response to BMN 270 gene therapy. The aim is to evaluate the safety, tolerability, and efficacy of BMN 270 in patients with severe HA and pre-existing antibodies against AAV5 vector capsid at various levels of AAV5 antibody titers.

7.4 Summary of Overall Risks and Benefits

Overall, 151 subjects have received a BMN 270 infusion at one of 4 dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, or 6E13 vg/kg) in one of the four ongoing BMN 270 clinical studies (270-201, 270-301, 270-302, 270-203). Single infusions have been generally well-tolerated across all investigated doses. All subjects have successfully completed their full-dose infusion of BMN 270, with no discontinuation of dosing due to adverse events observed during the infusion. No deaths have been reported in any of the BMN 270 studies, and no participants have discontinued from studies as a result of an adverse event.

Transient ALT elevation (Grade 1 to 3 in severity) has been observed in most subjects administered BMN 270 shortly after dosing, with no evidence for major impacts upon liver function; no events meeting the Hy's Law criteria have been identified. Liver function has remained stable over time. ALT elevations have been reported as events of interest in 13 subjects in 270-201, 1 subject in 270-302, and 91 subjects in 270-301. Although the majority of events have been Grade 1 or Grade 2 in severity, 11 subjects (1 in 270-302 and 10 in 270-301) had a reported Grade 3 ALT elevation. Only one serious event of ALT increased has been reported by investigators (in addition to one event that BioMarin conservatively assessed as serious based on the details of the case). While the majority of ALT elevations responded rapidly to corticosteroids, given current interest in the field of AAV gene therapy for the use of non-steroidal approaches to managing or preventing ALT elevations, alternate non-steroidal systemic immunosuppressive agents have also been used to manage hepatic reactions where corticosteroids have proven to be ineffective or where high doses/and or prolonged exposure to corticosteroids have led to unwanted side effects. Overall, the literature and clinical experience with BMN 270 suggests that transient elevations in liver enzymes are expected following AAV-based gene therapy for the treatment for hemophilia A or B without any long-term concerns of hepatic injury (Manno, 2006; Nathwani, 2011; George, 2016; Miesbach, 2016; Pasi, 2017).

Short-lived infusion reactions associated with one-time BMN 270 administration have included symptoms such as nausea, maculopapular rash, urticaria, diarrhea, watery eyes, rigors, chills, myalgia, fever, tachycardia and hypotension emerging within 24 hours of receiving BMN 270. Most infusion-related reactions were Grade 1 or Grade 2 in severity, and all events resolved, typically within 48 hours following medical management. Three of these cases required temporary interruption of the infusion, followed by re-initiation at a slower rate. All subjects completed their infusions. The reactions with onset during or within approximately 5 hours after the end of infusion responded to treatment with systemic

antihistamines and/or corticosteroids, where administered. Infusion-related reactions were effectively mitigated by managing infusion rate and medications.

No subjects have experienced thromboembolic events or developed inhibitors to FVIII following BMN 270 infusion.

At the highest dose tested in 270-201 (6E13 vg/kg), the majority of subjects achieved FVIII levels above 50 IU/dL at 52 weeks post-infusion. Subjects in that cohort also reported markedly decreased bleeding compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. Subjects at all dose levels continue to be followed.

In 270-301, an interim analysis has shown increased FVIII activity in the majority of subjects to mild HA or normal levels at 26 weeks post-infusion, also with markedly decreased bleeding compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. All subjects who will be included in the final analysis have been dosed with 6E13 vg/kg and continue to be followed.

The current data available for BMN 270 has shown an established positive benefit:risk profile for BMN 270 at the 6E13 vg/kg dosing level, although impact of prophylactic corticosteroids requires further investigation. Given the monitoring measures in place in the clinical protocol(s) to minimize the risk to subjects participating in the existing studies, the identified risks are justified by the anticipated benefits that may be afforded to subjects. Each subject in 270-203 will have a comprehensive surveillance plan that monitors LTs during the study, and elevations in LT will be addressed according to the guidelines set forth in the protocol. Safety will be assessed by adverse event reporting and clinical laboratory assessments.

For additional information on findings from other BMN 270 clinical studies, refer to the current version of the IB.

8 STUDY OBJECTIVES

The primary objective of the study is to:

- Assess the safety of a single intravenous administration of BMN 270 in severe HA subjects with pre-existing antibody to AAV5 vector capsid, including development of FVIII neutralizing antibody

The secondary objectives of the study are to:

- Assess the efficacy of BMN 270 defined as FVIII activity at or above 5 IU/dL at Week 26
- Assess the impact of BMN 270 on usage of exogenous FVIII replacement therapy
- Assess the impact of BMN 270 on the number of bleeding episodes requiring exogenous FVIII therapy
- Evaluate the pharmacodynamics of FVIII expression following IV infusion of BMN 270
- Assess the impact of BMN 270 on patient-reported outcomes (PROs)

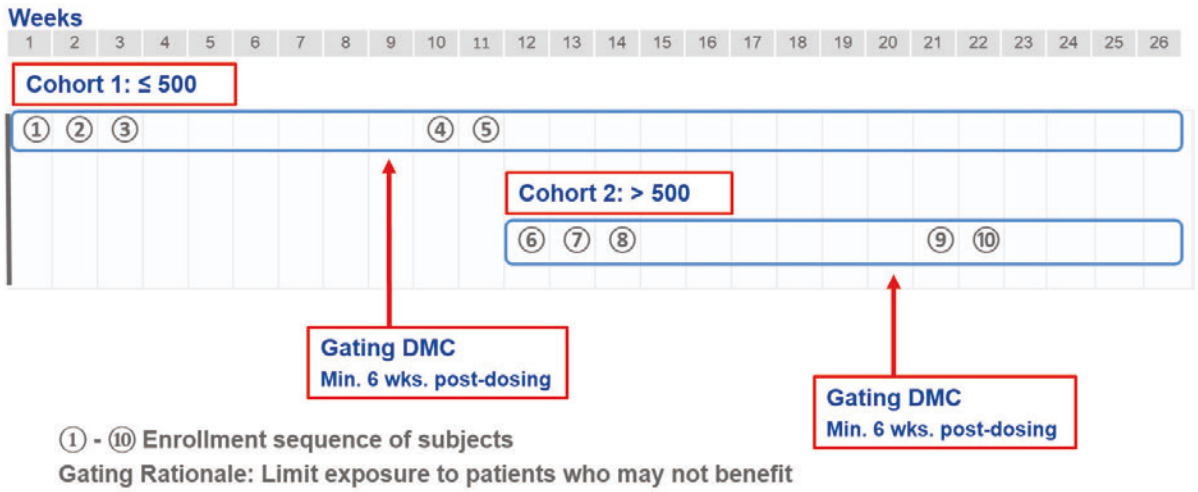
9 INVESTIGATIONAL PLAN

9.1 Overall Study Design and Plan

This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects may be enrolled at 5-6 sites globally in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date.

An independent Data Monitoring Committee (DMC), consisting of experts in clinical trials, statistics, and hemophilia, has been convened. The DMC will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 9.1.1 represents one dosing schedule scenario):

Figure 9.1.1: 270-203 Dosing Schedule (One Scenario)



Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from a minimum of 6 weeks post-infusion) reviewed. Based on data from 270-201, FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII

activity levels, compared with the one subject who had a slightly lower response. Up to 6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness).

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC evaluation of safety and efficacy in treated subjects with the following triggers that may potentially pause further enrollment:

- any related SAE;
- any related AE with a severity > CTCAE Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects at 6 weeks post-BMN 270 infusion.

Following a temporary halt of enrolment, the DMC may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed. Additional blood samples will be collected (tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine bead array (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.

Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DMC based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL.

As the relationship between activity assay results of the BMN 270 gene product and bleeding remains to be established, Investigators should strive to minimize bias by avoiding consideration of FVIII activity levels by themselves or subjects in the reporting of bleeding episodes and FVIII usage.

In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis. Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor or Sponsor-designated Data Monitor, follow an abbreviated visit schedule after Week 52 by attending only the Q12W and End of Year visits during Years 2-5. Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.

The study analysis will be performed after all subjects have been followed for 26 weeks post-BMN 270 infusion, with presentation of safety, efficacy, and FVIII PD assessments. After the safety, efficacy, and FVIII PD analyses at 26 weeks post-BMN 270 infusion, long-term safety and efficacy will be assessed in all subjects for up to a total of 5 years post-infusion. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the DMC.

A summary of all assessments is provided in the Schedule of Activities (SoA) in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5.

Table 9.1.1: Schedule of Activities – Screening/Baseline/Day 1

Assessment	Prior to BMN 270 Infusion				BMN 270 Infusion Visit (Day 1)[m]
	Screening		Smart Rescreening[k] (Day -28 to Day -1)	Baseline (Day -7 to Day -1)[l]	
	Screening (Day -42 to Day -29)	Screening (Day -28 to Day -1)			
Informed consent	X	X			
Demographics (age, sex, race, ethnicity)		X			
Medical History (including hemophilia A history, Hepatitis B, Hepatitis C, and HIV)		X			
Physical Examination[a]		X		X	X
Height and Weight[a]		X			
Vital Signs		X	X	X	X
Assessment of Adverse Events and Concomitant Medications		X	X	X	X
Documentation of bleeding episodes and FVIII usage for previous 12 months (by either subject or clinical information)		X	X	X	
Distribution of subject diaries and training in their use[b]		X			
Electrocardiogram		X			
Liver Ultrasound/FibroScan		X			
hFVIII Assays[c]		X	X[n]	X	
AAV5 TAb Assay (ARUP)[d]	X	X[d]	X		X
AAV5 TAb Assays[e]				X	
AAV5 TI Assay[e]				X	X
Screen for Hepatitis B, Hepatitis C, HIV[f]		X			
Screen for COVID-19 (local or central)[g]		X	X		
Blood chemistry, hematology, and coagulation tests[h]		X	X	X	
Blood fasting lipid panel					X
Fasting FibroTest					X
Urine Tests[h]		X	X	X	
Liver Tests[h]		X	X	X	

Assessment	Prior to BMN 270 Infusion				BMN 270 Infusion Visit (Day 1)[m]
	Screening		Smart Rescreening[k] (Day -28 to Day -1)	Baseline (Day -7 to Day -1)[l]	
	Screening (Day -42 to Day -29)	Screening (Day -28 to Day -1)			
PBMC collection (for baseline determination of AAV5 and FVIII specific cellular immunity)				X	
Von Willebrand Factor Antigen (VWF:Ag)				X	
Thrombin Generation Assay				X	
PCR of vector DNA in blood, saliva, urine, semen, and stools				X	X[m]
Biomarker testing[i]		X			
Exploratory biomarker assessments[j]				X	X
Cytokine bead array assay				X	
Haemo-QOL-A assessment				X	
EQ-5D-5L assessment				X	
HAL assessment				X	
WPAI+CIQ:HS assessment				X	
BMN 270 Infusion					X
Complement panel					X[o]
Hypersensitivity blood assessments				X	(X)[p]

[a] Complete physical examination should be done at Screening. Brief physical examination may be done at Baseline and at the BMN 270 Infusion Visit.

[b] Diaries should be distributed to subjects who have consented to participate in the study and who have been determined to meet all study eligibility criteria.

[c] Includes baseline FVIII activity (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), coagulation exploratory assay, hFVIII inhibitor level (Bethesda assay with Nijmegen modification), hFVIII total antibody titer, and hFVIII antigen assay. Baseline activity should be assessed at trough (at least >72 hours after last dose of replacement FVIII therapy, or 5x the known half-life of the FVIII concentrates administered).

[d] Screening, Smart Re-screening, and Infusion Day samples will be tested using the ARUP AAV5 TAb assay. During Screening, the ARUP AAV5 TAb assay test may be done first, under a standalone informed consent form, before the main ICF for the study is signed and further screening procedures are performed. Sample collection on the day of the infusion visit must be performed before the BMN 270 infusion is given. If the ARUP AAV5 TAb assay test is done first with the standalone consent, it does not need to be repeated as part of regular Screening.

[e] Baseline and all post-dose samples will be tested in a different AAV5 TAb post-dose immunogenicity monitoring assay.

[f] Subjects with documented negative results within the last 30 days do not need to be retested. Hepatitis B screening should include hepatitis B surface antigen (HBsAg), hepatitis B surface antibody (HBsAb), and hepatitis B core antibody (HBcAb).

[g] COVID-19 RT-PCR testing is required during Screening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing.

[h] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. ABO blood typing assessment should be performed at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study.

[i] Includes HLA genotyping and FVIII genotyping.

[j] Blood samples will be collected from subjects to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for those evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[k] Smart rescreening should only be performed if a subject has been determined to be eligible for the study and is unable to complete the Baseline assessments and Infusion prior to the closing of the original Screening window (28 + 14 days). COVID-19 RT-PCR testing is required as part of smart rescreening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing. Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once.

[l] Should the screening visit occur within 30 days of the drug infusion, physical examination, blood chemistry, LTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline.

[m] With the exception of the collection of samples for PCR vector DNA analysis, assessments on the day of infusion must be performed prior to the infusion. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit. On the day of the BMN 270 Infusion, vital signs will be monitored prior to the infusion, during the infusion every 15 minutes (± 5 minutes), and following the infusion hourly (± 5 minutes) for at least 8 hours during the subject's stay in the clinic. Shedding samples for PCR of vector DNA analysis (blood, saliva, urine, semen, stool) should be collected between 2 and 24 hours after the infusion has been completed.

[n] Only the hFVIII inhibitor level (Bethesda assay with Nijmegen modification) assay must be done at smart rescreening.

[o] Complement panel should include C3, C3a, C4, Bb, and sC5b-9 and should be collected within 2 hours post-infusion.

[p] In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination, vital signs will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.

Table 9.1.2: Schedule of Activities – Post-Infusion Follow-Up (Week 1-16)

Assessment	Follow-Up After BMN 270 Infusion – Weeks*																	
	Week 1																	
	D2	D4	D8	2	3	4	5[g]	6	7[g]	8	9[g]	10	11[g]	12	13[g]	14	15[g]	16
Study Day*	2	4	8	15	22	29	36	43	50	57	64	71	78	85	92	99	106	113
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight						X				X				X				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X		X		X		X						X		
Urine Tests[b]														X				
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII antibody titer				X		X		X		X		X		X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]	X[d]	X	X	X	X	X		X		X				X				X
Exploratory biomarker assessments[e]				X				X				X				X		
Haemo-QOL-A assessment														X				
EQ-5D-5L assessment														X				
HAL assessment														X				
WPAI+CIQ:HS assessment														X				
AAV5 TAb Assay	X		X	X		X		X		X		X		X		X		X
AAV5 TI Assay	X		X	X		X		X		X		X		X		X		X

Assessment	Follow-Up After BMN 270 Infusion – Weeks*																	
	Week 1																	
	D2	D4	D8	2	3	4	5[g]	6	7[g]	8	9[g]	10	11[g]	12	13[g]	14	15[g]	16
Study Day*	2	4	8	15	22	29	36	43	50	57	64	71	78	85	92	99	106	113
Testing for reactivation of hepatitis B and hepatitis C																		X[f]
PBMC collection (for determination of AAV5 and FVIII specific immunity)	X			X		X		X		X		X		X		X		X
Complement panel[h]	X	X	X	X		X		X		X		X		X				
VWF:Ag						X				X				X				X
Cytokine bead array assay	X		X	X				X				X				X		

* Visit windows are ± 24 hours during Week 1 and ± 48 hours starting with the Week 2 visit.

[a] Brief physical examination should be done at all weekly visits.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs may be monitored more or less frequently (and in particular when ALT values are > ULN or ≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen assay. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Collection for each matrix to occur until at least 3 consecutive results below the limit of detection are obtained. Collection and testing of semen samples must continue through Week 12, even if 3 consecutive results below the limit of detection in that compartment have already been recorded. Separate collection on Day 2 is not required if vector shedding samples were collected on Day 1 post-BMN 270 infusion; if collection on Day 2 is needed, it must occur within 24 hours of the time of the BMN 270 infusion.

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] Testing for reactivation of hepatitis B and hepatitis C at Week 16, for subjects with a past medical history of hepatitis B or hepatitis C prior to study entry, should be performed only in subjects who have not received prophylactic oral corticosteroids; subjects who have received prophylactic oral corticosteroids will have hepatitis B and hepatitis C testing at the time points indicated in Table 9.1.6.

[g] The scheduled visits at Week 5, Week 7, Week 9, Week 11, Week 13, and Week 15 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits.

[h] Complement panel should include C3, C3a, C4, Bb, and sC5b-9.

Table 9.1.3: Schedule of Activities – Post-Infusion Follow-Up (Week 17-32)

Assessment	Follow-Up After BMN 270 Infusion – Weeks*															
	17[f]	**18**	**19[f]**	**20**	**21[f]**	**22**	**23[f]**	**24**	**25[f]**	**26**	**27[f]**	**28**	**29[f]**	**30**	**31[f]**	**32**
Study Day*	120	127	134	141	148	155	162	169	176	183	190	197	204	211	218	225
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight[a]				X						X						X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X						X						X
Urine Tests[b]										X						
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII antibody titer				X				X		X						X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]				X				X		X						X
Exploratory biomarker assessments[e]		X				X				X						X
Haemo-QOL-A assessment										X						
EQ-5D-5L										X						
HAL										X						
WPAI+CIQ:HS										X						
AAV5 TAb Assay		X		X		X		X		X		X		X		X
AAV5 TI Assay		X		X		X		X		X		X		X		X
PBMC collection (for determination of AAV5 and FVIII specific cellular immunity)		X		X		X		X		X		X		X		X
VWF:Ag				X						X						
Thrombin Generation Assay[e]				X				X		X						X

Assessment	Follow-Up After BMN 270 Infusion – Weeks*															
	17[f]	18	19[f]	20	21[f]	22	23[f]	24	25[f]	26	27[f]	28	29[f]	30	31[f]	32
Study Day*	120	127	134	141	148	155	162	169	176	183	190	197	204	211	218	225
Cytokine bead array assay				X				X		X						X

* Visit windows are ± 48 hours.

[a] Brief physical examination should be done at all weekly visits except Week 26, where a complete physical examination should be performed. Weight should be recorded at Week 20, Week 26, and Week 32.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs may be monitored more or less frequently (and in particular when ALT values are > ULN or ≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Collection for each matrix to occur until at least 3 consecutive results below the limit of detection are obtained.

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] The scheduled visits at Week 17, Week 19, Week 21, Week 23, Week 25, Week 27, Week 29, and Week 31 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits.

Table 9.1.4: Schedule of Activities – Post-Infusion Follow-Up (Week 33-52)

Assessment	Year 1 – Weeks*											
	33[e]	**34**	**35[e]**	**36**	**38[e]**	**40**	**42[e]**	**44**	**46[e]**	**48**	**50[e]**	**52**
Study Day[*]	232	239	246	253	267	281	295	309	323	337	351	365
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X
Weight[a]				X				X				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X				X				X
Urine Tests[b]				X								X
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]	X	X	X	X	X	X	X	X	X	X	X	X
AAV5 TAb Assay		X		X		X		X		X		X
AAV5 TI Assay		X		X		X		X		X		X
FVIII antibody titer				X				X				X
Exploratory biomarker assessments[d]				X		X		X		X		X
PBMC Collection (for determination of FVIII and Capsid specific cellular immunity)		X		X				X				X
VWF:Ag				X								X
Thrombin Generation Assay[d]				X		X		X		X		X
Cytokine bead array assay				X				X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools				X		X		X		X		X
Haemo-QOL-A assessment												X
EQ-5D-5L												X
HAL												X

Assessment	Year 1 – Weeks*											
	33[e]	34	35[e]	36	38[e]	40	42[e]	44	46[e]	48	50[e]	52
WPAI+CIQ:HS												X

* Visit windows are ± 48 hours through Week 36, then ±1 week until Week 52.

[a] Complete physical examination should be performed at Week 52; brief physical exam may be performed at other study visits. Weight should be recorded at Week 36, Week 44, and Week 52.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. LTs may be monitored more or less frequently (and in particular when ALT values are > ULN or ≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN or increased by > 10 U/L from prior assessment; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII protein assay. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[e] The scheduled visits at Week 33, Week 35, Week 38, Week 42, Week 46, and Week 50 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits.

Table 9.1.5: Schedule of Activities – Long-Term Follow-Up (Year 2 – Year 5)

Assessment	Years 2-5*		End of Year Visit				ETV
			Year 2	Year 3	Year 4	Year 5	
Study Week*	Q12W	Q6W[gh]	W104	W156	W208	W260	
Physical examination[a]	X[a]		X[a]				X
Weight[a]	X[a]		X[a]				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X				X
Vital Signs	X		X				X
Blood chemistry, hematology, and coagulation tests[b]	X[b]		X[b]				X
Urine Tests[b]	X[b]		X[b]				X
Liver Tests[b]	X	X	X				X
FVIII assays[c]	X	X	X				X
AAV5 TAb Assay	X		X				X
AAV5 TI Assay	X		X				X
FVIII antibody titer	X		X				X
Exploratory biomarker assessments[e]	X		X				X
PBMC Collection (for determination of FVIII and Capsid specific cellular immunity)	X		X				X
VWF:Ag	X		X				X
Thrombin Generation Assay[e]	X		X				X
Cytokine bead array assay	X		X				X
PCR of vector DNA in semen[d]	(X)[d]	(X)[d]	(X)[d]				(X)[d]
PCR of vector DNA in blood, saliva, urine, and stools[d]	(X)[d]		(X)[d]				(X)[d]
Haemo-QOL-A assessment			X[f]				X
EQ-5D-5L			X[f]				X

Assessment	Years 2-5*		End of Year Visit				ETV
			Year 2	Year 3	Year 4	Year 5	
Study Week*	Q12W	Q6W[gh]	W104	W156	W208	W260	
HAL			X[f]				X
WPAI+CIQ:HS			X[f]				X

* Visit windows are ± 2 weeks for visits in Years 2-5. The Q6W visits during Years 2-5 should restart after each End of Year visit (eg, the first Q6W visit during Year 3 should be ~6 weeks after the End of Year 2 visit).

[a] Brief physical examination should be performed at all visits during Years 2-5. Weight should be recorded at the second Q12W visit each year and at every End of Year visit during Years 2-5.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. LTs may be monitored more or less frequently (and in particular when ALT values are > ULN or ≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN or increased by > 10 U/L from prior assessment; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. During Years 2-5 of the Post-Infusion Follow-Up period, urine tests and blood, chemistry, and coagulation tests should be performed at the second Q12W visit each year and at every End of Year visit.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII protein assay. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism. If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.

[d] Sample testing during Long-Term Follow-Up is not required if at least 3 consecutive samples were below the limit of detection during the Post-Infusion Follow-Up period. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 52 should continue to have PCR testing of semen every 6 weeks during Years 2-5 until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for TGA assay and any other exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] PRO assessments during Years 2-5 of Long-Term Follow-up should be performed at every End of Year visit.

[g] Subjects who meet the definition of treatment failure to BMN 270 therapy after Week 52 (refer to Section 12.6) may omit the Q6W visits during Years 2-5, and must attend only the Q12W and End of Year visits. Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in semen must still provide samples Q6W during Years 2-5 until vector shedding has been cleared (but do not need to do other scheduled assessments at those visit dates).

[e] The scheduled Q6W visits during Years 2-5 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits.

Table 9.1.6: Suggested Schedule of Activities – Prophylactic Corticosteroids

Assessment	Corticosteroid Treatment Period[b]																Post-Corticosteroid Period[c]				
	Day 1	Week															Week				
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	1	2	3	4	13
Prophylactic corticosteroid dose (mg/day)	40 mg	40 mg	40 mg	40 mg	40 mg	35 mg	35 mg	30 mg	30 mg	25 mg	25 mg	20 mg	20 mg	15 mg	10 mg	5 mg					
FVIII activity testing	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Liver tests	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Hepatitis B testing[d]							X										X				X
HCV Viral Load[d]							X										X				X

[a] This table provides an example of a prophylactic corticosteroid course. Clinical judgment, weighing the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses, and discussions between the Investigator and Medical Monitor are advised for any questions or concerns.

[b] Following initiation or completion of corticosteroid regimen, if a recurrence of ALT values ≥ ULN or ≥ 2x baseline value is reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor. Modification of the corticosteroid regimen may take into consideration possible confounders for the ALT elevation, relationship between increases in ALT and FVIII activity, ALT/FVIII levels post-corticosteroid initiation, and adverse events related to corticosteroid dosing. Guidance for tapering oral corticosteroid dosing can be found in Section 9.4.8.2, although a discussion between the PI and Medical Monitor should take place prior to tapering the corticosteroid dose.

[c] After discontinuation of oral corticosteroids, weekly labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values. If these assessments are already being done as part of normal study follow-up, they do not need to be duplicated.

[d] Should only be performed in subjects with a history of hepatitis B or hepatitis C prior to study entry.

9.2 Discussion of Study Design, Including Choice of Control Group

Study 270-203 is designed to be a Phase 1/2, single-arm, open-label study in hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid. Hemophilia A patients who provide written informed consent and meet the entry criteria will be eligible to enroll in the study.

Approximately 10 subjects may be enrolled at the 6E13 vg/kg BMN 270 dose. Subjects will be followed for 26 weeks post-BMN 270 infusion during which safety and efficacy assessments will be taken. After the final analysis at 26 weeks post-infusion, safety and efficacy will then continue to be assessed long-term for approximately a total of 5 years.

9.3 Selection of Study Population

Approximately 10 hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid may enroll into the study.

Additional criteria for participation in the study are provided in Section 9.3.1 and Section 9.3.2.

9.3.1 Inclusion Criteria

Individuals eligible to participate in this study must meet all of the following criteria:

1. Males ≥ 18 years of age with hemophilia A and residual FVIII levels ≤ 1 IU/dL as evidenced by medical history, at the time of signing the informed consent
2. Detectable pre-existing antibodies against the AAV5 vector capsid as measured by AAV5 total antibody ELISA
3. Treated/exposed to FVIII concentrates or cryoprecipitate for a minimum of 150 exposure days (EDs)
4. Subject must have been on prophylactic FVIII replacement therapy for at least 12 months prior to study entry.
5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures.
6. No previous documented history of a detectable FVIII inhibitor, and results from a Bethesda assay or Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) (or less than 1.0 BU for laboratories with a historical lower sensitivity cutoff for inhibitor detection of 1.0 BU) on 2 consecutive occasions at least one week apart within the past 12 months (at least one of which should be tested at the central laboratory)

7. Sexually active participants must agree to use an acceptable method of effective contraception, either double barrier contraception (ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with viral vector DNA below the limit of detection.
8. Willing to abstain from consumption of alcohol for at least the first 52 weeks following BMN 270 infusion.

9.3.2 Exclusion Criteria

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Any evidence of active infection, including COVID-19, or any immunosuppressive disorder, including HIV infection.
2. Significant liver dysfunction with any of the following abnormal laboratory results:
    - ALT (alanine aminotransferase) > 1.25x ULN;
    - AST (aspartate aminotransferase) > 1.25x ULN;
    - GGT (gamma-glutamyltransferase) > 1.25x ULN;
    - Total bilirubin > 1.25x ULN;
    - Alkaline phosphatase > 1.25x ULN; or
    - INR (international normalized ratio) ≥ 1.4

    Subjects whose laboratory assessments fall outside of these ranges may undergo repeat testing of the entire liver test panel within the same Screening window and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor.
3. Most recent, prior FibroScan or liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used.
4. Evidence of any bleeding disorder not related to hemophilia A
5. Platelet count of < 100 x 10^9/L
6. Creatinine ≥ 1.5 mg/dL
7. Liver cirrhosis of any etiology as assessed by liver ultrasound/FibroScan.

8. Chronic or active hepatitis B as evidenced by positive serology testing (hepatitis B surface antigen [HBsAg], hepatitis B surface antibody [HBsAb], and hepatitis B core antibody [HBcAb]) and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results in the Laboratory Manual.
9. Active Hepatitis C as evidenced by detectable HCV RNA, or currently on antiviral therapy
10. Active malignancy, except non-melanoma skin cancer
11. History of hepatic malignancy
12. History of arterial or venous thromboembolic events (eg, deep vein thrombosis, non-hemorrhagic stroke, pulmonary embolism, myocardial infarction, arterial embolus), with the exception of catheter-associated thrombosis for which anti-thrombotic treatment is not currently ongoing.
13. Known inherited or acquired thrombophilia, including conditions associated with increased thromboembolic risk, such as atrial fibrillation.
14. A history of known inflammatory, connective tissue, or autoimmune disorders (eg, vasculitis).
15. Treatment with any Investigational Product within 30 days or 5 half-lives of the investigational product (whichever is longer) prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study
16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including corticosteroid treatment and/or use of alternative immunosuppressive agents outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.
17. Prior treatment with any vector or gene transfer agent
18. Major surgery planned in the 52-week period following the infusion with BMN 270
19. Use of systemic immunosuppressive agents, not including corticosteroids, or live vaccines within 30 days before the BMN 270 infusion
20. Concurrent enrollment in another clinical study, unless it is an observational (non-interventional) clinical study that does not interfere with the requirements of the current protocol or have the potential to impact the evaluation of safety and efficacy of BMN 270 and with prior consultation with the Medical Monitor
21. Known allergy or hypersensitivity to investigational product formulation
22. Unwilling to receive blood or blood products for treatment of an adverse event and/or a bleed

9.3.3 Removal of Subjects from Treatment or Assessment

Subjects may withdraw their consent to participate in the study at any time without prejudice. The Investigator must withdraw from the study any subject who requests to be withdrawn. Such subjects will always be asked about the reason(s) for withdrawal. The Investigator will discuss with the subject appropriate procedures for withdrawal from the study. The Investigator should ask the subject's consent to perform the procedures listed under the early termination visit. Should a subject withdraw from the study, all efforts will be made to complete and report the observations as thoroughly as possible up to the date of the withdrawal.

A subject's participation in the study may be discontinued at any time at the discretion of BioMarin or of the Investigator and in accordance with his/her clinical judgment. When possible, the tests and evaluations listed for the termination visit should be carried out and every effort will be made to gather follow-up safety data if possible.

BioMarin must be notified of all subject withdrawals as soon as possible. BioMarin also reserves the right to discontinue the study at any time for either clinical or administrative reasons and to discontinue participation by an individual Investigator or site for poor enrollment or noncompliance.

Reasons for which the Investigator or BioMarin may withdraw a subject from the study include, but are not limited to, the following:

- Subject requires medication or medical procedure prohibited by the protocol
- Subject was erroneously enrolled into the study or does not meet entry criteria and not yet been dosed with BMN 270; subjects who do not meet entry criteria but who erroneously receive BMN 270 should remain in the study for safety monitoring
- Subject is lost to follow-up

If a subject fails to return for scheduled visits, a documented effort must be made to determine the reason. If the subject cannot be reached by telephone, a certified letter should be sent to the subject requesting contact with the Investigator. This information should be recorded in the study records.

Subjects may be considered lost to follow-up if the subject has missed 3 consecutive visits in the study and has failed to communicate a reason for this to the site. In addition, the site has documented at least 4 attempted contacts by key research personnel to reach the subject without success in the following manner:

- 2 attempts by telephone or email (if possible); then

- If telephone/email contacts are unsuccessful, 2 attempts must be made by certified letter or by appropriate local process.

Where communication has been made by phone, this should be documented in the subject source notes.

The Investigator or designee must explain to each subject, before enrollment into the study, that for evaluation of study results, the subject's protected health information obtained during the study may be shared with the study Sponsor, regulatory agencies, and IRB/EC It is the Investigator's (or designee's) responsibility to obtain written permission to use protected health information per country-specific regulations, such as the Health Insurance Portability and Accountability Act (HIPAA) in the US, from each subject. If permission to use protected health information is withdrawn, it is the Investigator's responsibility to obtain a written request, to ensure that no further data will be collected from the subject and the subject will be removed from the study.

9.3.3.1 Study Safety Evaluation Criteria

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC evaluation of safety and efficacy in treated subjects with the following triggers for pausing further enrollment:

- any related SAE;
- an related AE with a severity > Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

Additionally, the DMC should consider whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction. Following a temporary halt of enrolment, the DMC may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

If a Grade 4 or Grade 5 adverse event assessed as related to study drug occurs in a study subject, further enrollment into the trial will be halted until a full safety review of the data by the DMC has taken place. Relevant reporting and discussion with the Sponsor and the DMC will take place before resumption of dosing.

If any of the following events occur in a subject in the study who has received BMN 270 infusion, further enrollment into the trial will be temporarily put on hold pending prompt evaluation by the DMC.

1. Liver dysfunction (criteria do not apply to ALT elevations with an extra-hepatic etiology):
    - ALT >5x ULN, for more than 2 weeks
    - ALT >3x ULN and (total bilirubin >2x ULN **or** INR >1.5)
    - ALT >3x ULN with signs and symptoms of liver dysfunction
2. The occurrence of an AE of hepatic failure.
3. The detection of high titer neutralizing antibodies (>5 BU) to hFVIII following BMN 270 infusion in two subjects.
4. The occurrence of any cancer (except non-melanoma skin cancer) at any point after BMN 270 infusion.
5. The occurrence of a thromboembolic event with FVIII activity > 150 IU/dL in one subject.

If any of the following events occurs in a subject in the study who has received BMN 270 infusion, a prompt evaluation by the DMC will be required. Further enrollment into the trial will continue while DMC evaluation is ongoing, unless deemed otherwise by the DMC.

1. Acute hypersensitivity assessed as related to BMN 270
2. The detection of high titer neutralizing antibodies (>5 BU) to hFVIII following BMN 270 infusion in one subject
3. Occurrence of a thromboembolic event

9.3.4 Subject Identification and Replacement of Subjects

Each subject will be assigned a unique subject identifier. This unique identifier will be on all eCRF pages. In legal jurisdictions where use of initials is not permitted, a substitute identifier will be used.

Subjects who withdraw from the study after receiving BMN 270 will not be replaced.

9.3.5 Duration of Subject Participation

The duration of participation for each subject will be approximately 264 weeks. This includes 4 weeks of screening, 1 day of BMN 270 infusion, 26 weeks of Post-Infusion Follow-Up, and 234 weeks of Long-Term Follow-Up.

9.4 Treatments

9.4.1 Treatments Administered

BioMarin and/or its designee will provide the study site with a supply of IP sufficient for the completion of the study. BioMarin is responsible for shipping study drug to clinical sites.

9.4.2 Identity of Investigational Product

9.4.2.1 Product Characteristics and Labeling

BMN 270 is a sterile, clear, colorless-to-pale yellow solution for IV infusion and is supplied in a 10 mL Crystal Zenith® (CZ) vial. Each CZ vial contains 8.5 mL (extractable volume 8 mL) of AAV5-hFVIII-SQ at a concentration of 2E13 vector genomes per mL in a pH 7.4 phosphate buffer.

The study drug is labelled according to the particulars approved by the relevant regulatory agencies.

9.4.3 Storage

At the study site, all IP must be stored under the conditions specified in the Pharmacy Manual in a secure area accessible only to the designated pharmacists and clinical site personnel. All IP must be stored and inventoried and the inventories must be carefully and accurately documented according to applicable state, federal and local regulations, ICH GCP, and study procedures.

9.4.4 Directions for Administration

On the day of infusion, the subject will come to the infusion site, where a physical examination will be performed by the Investigator or designee. If the subject is found to have an active acute illness at the time of planned infusion, then the infusion should be deferred until the illness has resolved; screening procedures may require repetition if outside the specified window. An IV catheter or butterfly needle will be inserted into a suitable peripheral vein (eg, the median cubital vein) and flushed with saline. FVIII replacement therapy will not be given since venipuncture is a minimally invasive procedure in these individuals under ordinary conditions.

BMN 270 will be prepared and infused as a pure solution over a dose-dependent time. Prepared drug will be kept at room temperature prior to administration. An electric syringe pump will be used to infuse through an in-line, low protein binding 0.22 micron filter. BMN 270 will be infused through the catheter using an appropriate infusion pump at an initial rate of 1 mL/min. The infusion rate should be increased every 30 minutes by 1 mL/min up to a maximum of 4 mL/min, provided that the subject's clinical condition permits such an increase. Of note, the IP has been shown to be stable at room temperature for approximately 10 hours following completion of product thaw. Vital signs (pulse, blood pressure, respiration rate and temperature) should be monitored at 15 minute (±5 minutes) intervals throughout the period of the infusion.

As with any infused biological product, there is a potential risk of acute, systemic hypersensitivity reactions (including anaphylaxis) with BMN 270. Dosing will be administered at a qualified infusion site, with appropriate resuscitation equipment and medication available and easily accessible.

Clinical staff administering BMN 270 should be trained appropriately in recognizing and managing the signs and symptoms associated with potential hypersensitivity, anaphylactic, and anaphylactoid reactions. Additionally, the Investigator should be familiar with Sampson's criteria for defining anaphylaxis (Sampson, 2006; Appendix 1).

Should symptoms of potential hypersensitivity occur, the infusion may be slowed or halted at the Investigator's discretion, with consideration of the subject's clinical condition. If the infusion is halted, it should only be restarted if the Investigator considers it safe and appropriate to do so. Antihistamines, anti-pyretic, and/or corticosteroid administration is permitted prior to restarting an interrupted infusion by an infusion-related reaction. At the restart, the infusion rate may be adjusted (ie, to a slower rate [minimum of 1 mL/min], with the rate increased every 30 minutes by 1 mL/min up to a maximum rate of 4 mL/min, if the subject's clinical condition permits such an increase) with careful monitoring of the subject. In the event of an infusion rate reaction with more than one dosing interruption, the infusion rate would not go beyond 1 mL/min.

In case of hypersensitivity reactions, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.

Following completion of the infusion, vital signs will be monitored hourly (± 5 minutes). If the vital signs are stable the catheter will be removed 8 hours after the infusion. Hemostasis at the puncture site will be established by applying pressure according to standard protocol for infusing FVIII concentrates. Subjects will remain in the clinic for at least 24 hours to observe for any immediate toxicity of the procedure; in-patient observation can be extended beyond 24 hours if needed per Investigator discretion. After 24 hours, subjects will be

discharged from the clinic unless toxicity has been observed in which case the stay in the clinic may be extended or the subject may transfer to a separate facility based on the evaluation and judgment of the Principal Investigator after consultation with the Medical Monitor.

Prior to discharging subjects from the clinic, the Investigator or designee should instruct subjects how to recognize signs and symptoms of potential (delayed) hypersensitivity reactions and anaphylaxis, and to contact a medical practitioner or seek emergency care in case of such an event.

9.4.5 Method of Assigning Subjects to Treatment Groups

Subjects who meet all eligibility criteria (refer to Section 9.3.1 and Section 9.3.2) may be enrolled into the study. Approval by the Medical Monitor will be required prior to enrollment of each study subject. Upon their enrollment into the study, subjects will be assigned a unique subject number.

9.4.6 Selection of Dose Used in the Study

Data from an ongoing first in human study (270-201) indicates that following single escalated doses of BMN 270 (6E12, 2E13, 4E13, 6E13 vg/kg), dose-related increases in FVIII activity were observed, with concurrent improvements in bleeding episodes and exogenous FVIII utilization, particularly at the 4E13 and 6E13 vg/kg dose levels. At all dose levels, BMN 270 is considered to be well-tolerated with mild increases in ALT as the most common adverse event. Please refer to the IB for detailed efficacy and safety data. The 6E13 vg/kg dose has been selected for this study to maximize the likelihood of transduction in the face of pre-existing AAV5 antibodies.

9.4.6.1 Selection of Timing of Dose for Each Subject

9.4.7 Blinding

This is an open-label study.

9.4.8 Prior and Concomitant Medications

All prescription and over-the-counter medications (including dietary and herbal supplements) taken by a subject for 30 days before Screening will be recorded on the designated eCRF. The Investigator may prescribe additional medications, deemed necessary to provide adequate prophylactic or supportive care, during the study, as long as the prescribed medication is not prohibited by the protocol. In the event of an emergency, any needed medications may be prescribed without prior approval, but the Medical Monitor must be notified of the use of any contraindicated medications immediately thereafter. Any

concomitant medications added or discontinued during the study should be recorded on the eCRF. Medications should, whenever possible, not be recorded in the electronic database with a frequency of as needed (PRN).

The following medications are prohibited starting 30 days before Screening and through the end of the study, and the Sponsor must be notified if a subject receives any of these during the study:

- Any investigational therapy
- Emicizumab
- Fitusiran
- Concizumab
- Efavirenz

Subjects who begin antiretroviral therapy following BMN 270 infusion and during their participation in 270-203 must have their antiretroviral medication regimen approved by the Investigator and Medical Monitor prior to starting antiretroviral treatment.

The following medications should be avoided, starting 30 days prior to and for at least 52 weeks after BMN 270 infusion and minimized throughout the remaining duration of the study.

- Alcohol
- Herbal and natural remedies and dietary supplements
- Medications which may be hepatotoxic, including isotretinoin and dextroamphetamine/amphetamine
- Medications which may reduce or increase the plasma concentration of corticosteroids

Subjects should be counseled to avoid starting potentially hepatotoxic therapies and to inform the Investigator of any new medications prescribed by other physicians. Investigators should carefully consider both the mechanism of action and potential hepatotoxicity of any new medication prior to initiation. If a potentially concerning new medication is started, Investigators should closely monitor both FVIII activity and ALT levels (eg, weekly to every 2 weeks for the first month) in order to determine if any detrimental effects on the efficacy or safety of BMN 270 have occurred. If co-medications are required during the course of the study, where possible, please check the National Center for Biotechnology Information LiverTox website for potential hepatotoxicity issues prior to prescribing (NCBI, 2020).

Vaccines should also be avoided during this period, but in particular during the first 26 weeks unless clinically indicated.

The following medications should be avoided during oral corticosteroid therapy:

- Vaccines
- NSAIDs

9.4.8.1 Concomitant Hemophilia Treatments

Subjects on prophylactic FVIII therapy will discontinue their regular treatment regimen starting 4 weeks after the day of infusion and switch to an "on-demand" schedule. FVIII replacement therapy can always be taken as needed by the subject for treatment of an acute bleeding episode; the subject must carefully record his treatment and bleeding episodes in his diary. Prophylactic FVIII can be used on a case-by-case basis and in consultation with the Medical Monitor to prevent bleeding in extenuating circumstances (eg, peri-operative).

In addition, information on FVIII usage and bleeding episodes by medical history will be collected from subjects for the 12-month period immediately preceding study enrollment.

9.4.8.2 Therapeutic Corticosteroid Treatment and/or Immunosuppressive Agent Treatment of Elevated Hepatic Transaminases

Refer to corticosteroid prescription guidelines for recommended monitoring for, and management of, potential side effects of corticosteroids, including guidance on medications that should be avoided during corticosteroid treatment.

All subjects will be started on prophylactic corticosteroids starting on the day of infusion (Day 1). Table 9.1.6 provides an example of a possible prophylactic corticosteroid course, including taper and post-corticosteroid additional monitoring of FVIII activity, LTs, and hepatitis B/hepatitis C reactivation. Clinical judgment, weighting the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses. Discussions between the Investigator and Medical Monitor are advised for any questions or concerns.

Following initiation or completion of the prophylactic corticosteroid regimen, if ALT levels become increased (eg, > ULN or ≥ 1.5x baseline value) and alternative etiologies have been ruled out, prompt institution of therapeutic or on-demand oral corticosteroids (prednisone or converted equivalent) should be considered after consultation with the Medical Monitor (refer to Table 9.7.6.3.2).

- Whenever possible, a confirmatory lab draw for ALT should be performed within 72 hours, along with FVIII activity, prior to initiating oral corticosteroids.

- Corticosteroids may be delayed if elevations in ALT are clearly not related to BMN 270 (eg, elevated in ALT with concurrent increase in CPK due to intensive exercise)
- Alternative immunosuppressive agents may also be considered for use on a case-by-case basis and following consultation with the Medical Monitor (eg, if prolonged corticosteroid use is contraindicated).

Therapeutic corticosteroid treatment should be initiated at a dose of 60 mg/day. At minimum, the recommended duration of therapeutic corticosteroids is 60 mg/day for 3 weeks, 40 mg/day for 4 weeks, and 30 mg/day for 4 weeks, followed by a gradual taper thereafter. Should a scenario arise in which a deviation from the minimum recommended dose and/or duration of therapeutic corticosteroids may be clinically indicated, a discussion should take place between the Investigator and Medical Monitor regarding corticosteroid dose adjustments. Tapering of corticosteroid dosages should be guided by the following (Table 9.4.8.2.1):

Table 9.4.8.2.1: Adjustments to Corticosteroid Regimen

Corticosteroids should be tapered on an individual subject basis with the following guiding principles:	Corticosteroids may be tapered **if**: • ALT ≤ 1.5x baseline value; and • FVIII activity levels > 90% of the pre-decline FVIII activity levels; and • There is no concern for adrenal insufficiency post-withdrawal
Increasing Corticosteroid Dose	If ALT level is increasing or FVIII activity level is decreasing while on oral corticosteroids, any increases in oral corticosteroid dosing should be made only upon consultation with the Medical Monitor

For any scenarios that are not accounted for in the above table, a discussion should take place between the Investigator and Medical Monitor regarding corticosteroid dose adjustments.

After discontinuation of oral corticosteroids, labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values.

Following initiation or completion of therapeutic oral corticosteroids, if increased ALT levels (eg, > ULN or ≥1.5x baseline value) are reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor. Modification of the corticosteroid regimen may take into consideration possible confounders for the ALT elevation and impact on FVIII expression.

Management and monitoring of reactions to corticosteroids should be determined by the Investigator's clinical judgment in consultation with the Sponsor's Medical Monitor. This includes the contraindicated use of NSAIDs during corticosteroid treatment and specific

monitoring not already covered by the SoA. The use of COX-2 inhibitors, while not contraindicated during corticosteroid treatment, should be limited, if possible. Practical management to prevent complications related to oral corticosteroid therapy may be undertaken at the discretion of the Investigator (eg, evaluation of glucose intolerance, hyperlipidemia). Alternative, non-steroidal systemic immunosuppressive agents may be used, following a discussion between the Investigator and the Medical Monitor, should corticosteroid use be deemed by an Investigator to be clinically ineffective, not tolerated, and/or contraindicated. Hepatitis B status and HCV viral load will be rechecked 6 weeks after the start of oral corticosteroid/immunosuppressive agent treatment and then 1 week and 13 weeks after the completion of oral corticosteroid/immunosuppressive agent treatment in subjects with a history of hepatitis B or hepatitis C. All adverse events (including any adverse events suspected to be caused by or related to corticosteroid/immunousuppressive agent use) should be reported as outlined in Section 10 of the protocol.

Subjects on corticosteroids should receive appropriate counselling and support regarding side effects from the Investigator or the treating institution (eg, listings of side effects and when to notify carers, wallet card for emergencies if on steroids, etc.). Additional management, including the co prescription of additional medications to prevent complications related to corticosteroid therapy, may be undertaken at the discretion of the investigator, including, but not limited to, prophylaxis against the occurrence of gastric ulcers, osteoporosis, and infections. The above guidance should also be followed in the event that an alternative immunosuppressive agent is used, as applicable.

9.4.9 Treatment Compliance

Study drug will be administered to subjects at the dosing facility by a qualified health care professional. The quantity dispensed, returned, used, lost, etc. must be recorded on a dispensing log. Sites will be instructed to return or destroy all used and unused study drug containers.

9.5 Investigational Product Accountability

The Investigator or designee is responsible for maintaining accurate records (including dates and quantities) of IP(s) received and IP lost or accidentally or deliberately destroyed. The Investigator or designee must retain all unused or expired study supplies until the study monitor (on-site CRA) has confirmed the accountability data, if allowed by local SOPs.

9.5.1 Return and Disposition of Clinical Supplies

Unused study drug must be kept in a secure location for accountability and reconciliation by the study monitor. The Investigator or designee must provide an explanation for any

destroyed or missing study drug or study materials (or must be referenced in their institution SOPs).

Unused study drug may be destroyed on site, per the site's standard operating procedures, but only after BioMarin has granted approval for drug destruction. The monitor must account for all study drug in a formal reconciliation process prior to study drug destruction. All study drug destroyed on site must be documented. Documentation must be provided to BioMarin or designee and retained in the Investigator study files. If a site is unable to destroy study drug appropriately, the site can return unused study drug to BioMarin upon request. The return of study drug or study drug materials must be accounted for on a Study Drug Return Form provided by BioMarin.

All study drug and related materials should be stored, inventoried, reconciled, and destroyed or returned according to applicable state and federal regulations and study procedures. For additional information, please refer to the Study Pharmacy Manual.

9.6 Dietary or Other Protocol Restrictions

There are no dietary or other protocol restrictions for this study. Alcohol should be avoided starting 30 days prior to and for at least the first 52 weeks of the study, and particularly within 48 hours prior to lab work. Alcohol use should be minimized throughout the remaining duration of the study.

Subjects should be advised to abstain from any blood, organ, or sperm donation after BMN 270 infusion, until there is no further evidence of vector shedding from PCR analysis of samples.

9.7 Efficacy and Safety Variables

9.7.1 Efficacy and Safety Measurements Assessed

The SoA (Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5) describe the timing of required evaluations.

9.7.2 Efficacy Variables

9.7.2.1 FVIII Activity

Efficacy (response to treatment) will be defined as FVIII activity ≥ 5 IU/dL at Week 26 following BMN 270 infusion.

Values for FVIII activity will be excluded from analysis if obtained within 72 hours since the last infusion of exogenous FVIII protein concentrates. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity

measurements when a 72-hour interval without FVIII use is achieved; The 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy.

In the event of an FVIII activity level decline during the study:

- If FVIII activity has declined at least 20% from the peak but less than 35% and has declined for at least 2 consecutive assessments, FVIII activity and LTs should be repeated every 7 days until FVIII activity is stable or increasing
- If FVIII activity has declined >35% from the peak and has declined for at least 2 consecutive assessments, FVIII activity and LTs should be repeated every 72 hours until FVIII activity is stable or increasing.

Note that fluctuations in FVIII activity are common, and if no clear trend indicating a decline in FVIII activity is observed, then this additional testing may be deferred (upon consultation between the Investigator and the Medical Monitor) until either a more clear trend of decline has been demonstrated or until the FVIII activity levels stabilize or increase.

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 52, as applicable and at the discretion of the Investigator. Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.

Details on collecting FVIII activity samples are included in the Laboratory Manual.

9.7.2.2 Factor VIII Replacement Therapy/Bleeding Episodes

Additional efficacy variables are:

- Change of annualized utilization (IU/kg) of exogenous FVIII replacement therapy during Week 5 to Week 26 post BMN 270 infusion from the baseline utilization of exogenous FVIII replacement therapy.
- Change of ABRs requiring exogenous FVIII replacement treatment during Week 5 to Week 26 of the study post BMN 270 infusion from the baseline ABR.

During the study, subjects will be asked at each study visit to report the use of factor replacement therapy and the number of bleeding episodes since the previous visit. This information will be captured on the subject's diary or other subject records.

Subjects are strongly encouraged to immediately consult Investigator for guidance regarding exogenous FVIII administration for suspected bleeds or bleeding episodes within the first 6 weeks post BMN 270 infusion.

In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis.

9.7.2.3 Patient-Reported Outcomes (PRO)

The Haemo-QoL-A questionnaire is a validated hemophilia-specific health-related quality of life questionnaire for adults (Rentz, 2008). It consists of 41 questions covering six domains (Physical Functioning, Role Functioning, Worry, Consequences of Bleeding, Emotional Impact and Treatment Concerns). Items are answered on a 6-point Likert-type scale, ranging from 0 (None of the time) to 5 (All of the time). Higher scores mean better health-related quality of life or less impairment for a particular subscale (Haemo-QoL Study Group, 2017). Details regarding the Haemo-QoL-A assessment will be included in the Study Reference Manual.

The EQ-5D-5L instrument is a self-reported questionnaire designed to measure general health status (The EuroQol Group, 1990) (Brooks, 1996). The EQ-5D-5L is composed of 2-parts: a descriptive system that assesses 5 levels of perceived problems (mobility, self-care, usual activities, pain/discomfort, and anxiety/depression) in 5 dimensions and the EQ visual analogue scale (EQ VAS) assessment for overall health. A sample copy of the EQ-5D-5L and additional information are provided in the Study Reference Manual.

The Haemophilia Activities List (HAL) measures the impact of hemophilia on self-perceived functional abilities in adults (van Genderen, 2006). The instrument consists of multiple domains including lying/sitting/kneeling/standing, leg and arm function, use of transportation, self-care, household tasks, and leisure activities where subjects are asked to rate their level of difficulty with activities of daily living on a 6-point Likert-type scale from 1 (Impossible) to 6 (Never). For some items, subjects are given the choice to answer 'Not applicable'. A sample copy of the HAL and additional information are provided in the Study Reference Manual.

The Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) instrument is designed to measure the effect of disease symptom severity on work productivity and classroom productivity (if applicable) (Recht, 2014). The WPAI+CIQ:HS questionnaire yields scores related to work/classroom absenteeism, reduced on-the-job effectiveness, overall work/classroom impairment, and activity impairment. WPAI+CIQ:HS outcomes are expressed as impairment percentages, with higher numbers indicating greater impairment and less productivity (Reilly, 2002).

A sample copy of the WPAI+CIQ:HS and additional information are provided in the Study Reference Manual.

9.7.3 Immunogenicity

Immunogenicity assays will be performed on plasma and PBMCs. The assays will include detection of anti-AAV5 vector capsid and anti-FVIII total antibodies, as well as determination of neutralizing antibodies against FVIII (FVIII inhibitors) and against the AAV5 vector capsid (Transduction Inhibitors, TI). FVIII Inhibitors will be assessed using the Bethesda assay with Nijmegen modification. Any abnormality of the liver parameters will lead to a retrospective immunogenicity assessment to evaluate FVIII-and vector capsid-specific cellular immunogenicity. FVIII- and vector capsid-specific cellular immunity will be assessed by stimulated cytokine secretion using an ELISpot assay performed on collected PBMCs.

9.7.4 Pharmacodynamics

The FVIII protein concentration and activity level as measured by a validated immunoassay and by a validated FVIII activity assay, respectively, will be used for plasma profiles; FVIII protein and activity will be used to determine PD parameters.

9.7.5 Exploratory Assessments

A cytokine bead array assay assessment will be performed at Baseline and then at the timepoints listed in the Schedule of Activities.

In addition, blood samples will be collected from subjects at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5 to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV5 gene transfer, and to develop assays used for these evaluations. Subject may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A.

All biomarker samples collected in this study may be used for exploratory biomarker research, including evaluation of additional biomarkers not specifically listed in the protocol. In addition, samples collected for other purposes in this study may be used for exploratory research once testing for the primary purpose has been completed.

9.7.6 Safety Variables

Safety in this study will be determined from evaluation of AEs, clinical laboratory assessments with a particular attention to the liver function, vital signs assessments, physical examinations, and immunogenicity.

9.7.6.1 Adverse Events

The determination, evaluation and reporting of AEs will be performed as outlined in Section 10.

9.7.6.2 Clinical Laboratory Assessments

The scheduled clinical laboratory tests are listed in Table 9.7.6.2.1. Refer to the Study Reference Manual for instructions on obtaining and shipping samples. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample.

Any abnormal test results determined to be clinically significant by the Investigator should be repeated (at the Investigator's discretion) until: (1) the cause of the abnormality is determined; (2) the value returns to baseline or to within normal limits; or (3) the Investigator determines that the abnormal value is no longer clinically significant.

All abnormal clinical laboratory results should be initialed and dated by an Investigator, along with a comment regarding whether or not the result is clinically significant. Each clinically significant laboratory result should be recorded as an adverse event.

The diagnosis, if known, associated with abnormalities in clinical laboratory tests that are considered clinically significant by the Investigator will be recorded on the AE eCRF.

Table 9.7.6.2.1: Clinical Laboratory Tests

Blood Chemistry	Hematology	Urine Tests	Coagulation Screen including:
Albumin	Hemoglobin	Appearance	APTT
BUN	Hematocrit	Color	PT/INR
Calcium	WBC count	pH	TT
Chloride	RBC count	Specific gravity	
Total cholesterol	Platelet count	Ketones	
CPK	Differential cell count	Protein	
Creatinine	RBC indices (MCV and MCH)	Glucose	
CRP		Bilirubin	**Other Tests:**
Glucose		Nitrite	ABO blood typing*
Phosphorus		Urobilinogen	
Potassium		Hemoglobin	
Total protein			
Sodium			
Uric Acid			

BUN, blood urea nitrogen; CPK, creatinine phosphokinase; CRP, C-reactive protein; PT, prothrombin time; APTT, activated partial thromboplastin time; RBC, red blood cell; WBC, white blood cell; TT, thrombin time; INR, international normalized ratio; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin.

* ABO blood typing assessment should be performed at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study.

In addition to scheduled clinical laboratory assessments, a fasting blood lipid panel (including triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) and fasting FibroTest will be assessed at the BMN 270 infusion visit. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit.

In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.

At applicable sites, certain study assessments may be performed by a mobile nursing (MN) professional at the patient's home or another suitable location, such as their school or office, to improve access and convenience for patients participating in the study. The Sponsor may select a healthcare company that will be responsible for providing MN services for participating sites (the MN vendor). The MN vendor is responsible for ensuring that all MN professionals are licensed, qualified, and in good standing, as per applicable regulations, and that appropriate background checks have been performed. If the investigator at a participating site determines that MN services are appropriate for a patient and the patient gives written informed consent to participate in MN visits, the MN network will communicate with the patient and the patient's site. Potential MN visits are designated in the Schedules of Events.

9.7.6.3 Liver and Hepatitis Testing

Subjects will be screened for evidence of previous or active hepatitis B or hepatitis C infection at Screening; hepatitis B screening should include HBsAg, HBsAb, and HBcAb. Subjects with documented results showing an absence of active hepatitis B or hepatitis C infection (as measured by negative surface antigen or DNA for hepatitis B or negative RNA testing for hepatitis C) 30 days prior to providing signed informed consent do not need to repeat those tests during the screening period.

Evidence of ongoing hepatitis B or hepatitis C infection is exclusionary. Subjects with history of hepatitis B or hepatitis C infection prior to study entry will be tested for hepatitis B and hepatitis C reactivation at Week 16. Subjects with a history of hepatitis B or hepatitis C will be asked for information about the treatments received as part of their medical history assessment at Screening.

Subjects with a previous history of hepatitis B or hepatitis C who receive therapeutic oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.6.

A liver ultrasound/FibroScan and liver tests (LTs) during Screening will identify any significant hepatic dysfunction.

Where a biopsy has been taken for safety-related reasons or was available from a past procedure, the Sponsor may request the biopsy information to help evaluate the impact of BMN 270 on the liver. The Sponsor may request that slides from a liver biopsy be made available for additional histopathological review.

Liver tests will be monitored on a regular basis; at each time point specified in the SoA, the following LTs should be assessed:

Table 9.7.6.3.1: Liver Tests

Liver Tests (LTs)			
Alkaline Phosphatase	AST (SGOT)	Total Bilirubin	LDH
ALT (SGPT)	Direct Bilirubin	GGT	

ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma-glutamyltransferase; LDH, lactate dehydrogenase; SGOT, serum glutamic-oxaloacetic transaminase; SGPT, serum glutamic-pyruvic transaminase

Elevated ALT levels should be evaluated according to the following plan (note that these evaluations may indicate additional testing of LTs and FVIII levels at unscheduled visits; these unscheduled laboratory tests may be completed by a mobile nursing professional at sites where the use of MN services has been approved):

Table 9.7.6.3.2: Evaluation of ALT Elevations

ALT Level	Work-Up
≥1.5x Baseline - <2x Baseline	• Continue to monitor LTs and FVIII per protocol (repeat within 24-72 hours if next protocol scheduled visit is >24-72 hours from the time of the reported ALT elevation) • Consider evaluation to rule out alternative etiology (eg, concomitant medications, viral or autoimmune hepatitis, alcohol use, recreational drug use, special diets, strenuous exercise, prior and/or concurrent illnesses, exposure to environmental and/or industrial chemicals, etc.) (refer to Table 9.7.6.3.3) • If ALT is > ULN or > 2x baseline in 2 consecutive assessments within 24-72 hours and alternative etiologies have been ruled out, start oral corticosteroids upon consultation with the Medical Monitor (refer to Section 9.4.8.2)
≥2x Baseline or ≥ ULN - <3x ULN	• Repeat LTs and FVIII within 24-72 hours • Continue to monitor LTs weekly until ALT is stable or improving • Evaluate and rule out alternative etiologies (as above) • Consult with Medical Monitor • If ALT is ≥ 2x baseline or ≥ ULN - < 3x ULN in 2 consecutive assessments within 72 hours and alternative etiologies have been ruled out, start oral corticosteroids upon consultation with the Medical Monitor (refer to Section 9.4.8.2) • Obtain other possibly relevant laboratory evaluations (albumin, PT/INR, CRP, etc.) • Obtain complete blood count with differential to assess for eosinophilia • Obtain PBMC to evaluate potential immune response (prior to starting oral corticosteroids) • If no improvement in 14 days, consider gastroenterology and/or hepatology consult, abdominal workup, imaging (including MRI or ultrasound), and/or liver biopsy as appropriate
≥3x ULN	• Consult with Medical Monitor • Evaluate and rule out alternative etiologies (as above) • Repeat LTs and FVIII within 24-48 hours, and continue with monitoring of LTs at least twice weekly for as long as the subject's ALT remains ≥ 3x ULN • In the event that ALT or AST is ≥3x ULN and total bilirubin is ≥2x ULN, albumin and PT/INR should also be obtained. • If ≥3x ULN in 2 consecutive assessments within 48 hours, start oral corticosteroids (refer to Section 9.4.8.2) • Obtain other possibly relevant laboratory evaluations (albumin, PT/INR, CRP, etc.) • Obtain complete blood count with differential to assess for eosinophilia • Obtain PBMC to evaluate potential immune response (prior to starting oral corticosteroids) • If no improvement in 14 days, consider gastroenterology and/or hepatology consult, abdominal workup, imaging (including MRI or ultrasound), and/or liver biopsy as appropriate

When ruling out alternative viral or autoimmune hepatitis as part of the elevated ALT workup, the following tests should be performed:

Table 9.7.6.3.3: Viral and Autoimmune Hepatitis Testing

Viral Hepatitis Workup Testing	Autoimmune Hepatitis Workup Testing
Hepatitis A	Smooth muscle antibody
Hepatitis B	Mitochondrial antibody
Hepatitis C	Liver/kidney microsomal antibodies
Hepatitis E	Antinuclear antibody (ANA) HEP-2
Cytomegalovirus (CMV)	
Epstein-Barr virus (EBV)	
Herpes simplex virus (HSV) 1 & 2	

9.7.6.4 HIV Testing

HIV testing will be performed at Screening. Subjects with documented negative results within the last 30 days prior to screening do not need to be retested.

9.7.6.5 Vital Signs, Physical Examinations and Other Observations Related to Safety

Vital signs will include seated systolic and diastolic blood pressure, heart rate, respiration rate, and temperature. Any clinically significant change in vital signs will be recorded as an AE.

Systolic blood pressure, diastolic blood pressure, heart rate, respiration rate, and temperature will be assessed at Screening, Baseline, and at the beginning of each visit during the Post-Infusion Follow-Up and Long-Term Follow-Up periods. On the day of the BMN 270 Infusion, vital signs will be monitored prior to infusion, during the infusion every 15 minutes (± 5 minutes), following the infusion hourly (± 5 minutes) for at least 8 hours during the subject's stay in the clinic. Any abnormal vital sign assessments should be repeated, and both values should be recorded in the eCRF.

A complete physical examination should be performed at Screening, Week 26, Week 52, and at the End of Year visits. A complete physical examination will include general appearance (head, eyes, ears, nose, and throat), cardiovascular, dermatologic, lymphatic, respiratory, gastrointestinal, genitourinary, musculoskeletal, and neurologic systems.

At other visits, brief physical examinations may be performed at the discretion of the Investigator based on the subject's clinical condition. A brief physical examination will include general appearance, cardiovascular, dermatologic, respiratory, gastrointestinal, musculoskeletal, and neurologic assessments. Particular attention should be given to signs of bleeding, as well as assessing possible hemarthroses.

Height will be recorded at Screening only. Weight will be recorded at Screening and then every 4 weeks thereafter through Week 20, at Week 26, Week 32, Week 36, Week 44, and Week 52, and then at the second Q12W visit each year and at every End of Year visit during Years 2-5.

At visits where the MN services are used or shortened lab draw-only visits are conducted at the sites, the physical examination and vital signs assessments indicated in the Schedule of Activities will not be performed.

9.7.6.6 Vector Shedding

During the Post-Infusion Follow-Up period, subjects will undergo testing of various bodily samples to look for evidence of vector shedding for possible viral transmission. Bodily fluids will be tested by polymerase chain reaction (PCR). Fluids tested will include:

- Blood
- Saliva
- Semen
- Urine
- Stool

Vector shedding will also be extensively studied in the present clinical trial, at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5. Testing will continue until at least 3 consecutive results below the limit of detection are obtained. If a positive result is obtained in a matrix after 3 consecutive results below the limit of detection have already been recorded, testing in that matrix should restart and continue until an additional 3 consecutive results below the limit of detection have been obtained in order to confirm clearance.

Testing of semen will continue at least through Week 12, even if 3 consecutive results below the limit of detection have been recorded in that compartment prior to that time point. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 26 should continue to have PCR testing in semen at the timepoints designated in the SoA until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor). Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule (refer to Section 12.5.3) but who have not cleared vector shedding from a matrix must still provide samples for assessment until vector shedding has cleared: at Weeks 32, 36, 40, 44, 48, and 52 (during Year 1) and every 6 weeks (for semen samples) or every 12 weeks (for all uncleared

matrices) during Years 2-5. Such subjects may provide samples on the designated study visit dates either at the sites or through use of a MN professional.

Samples may be fractionated prior to shedding analysis in order to better characterize the presence, structure, and location of vector DNA and/or vector capsid within each matrix. If needed, the fractionation may be performed with samples collected specifically for shedding analysis (saliva, blood, semen, urine, feces). Alternatively, the vector DNA characterization during shedding analysis may utilize already fractionated exploratory samples obtained from the above biofluids, such as exploratory plasma samples, exploratory PBMC samples, and red blood cells recovered during PBMC/plasma isolations.

Fractionation of semen to collect purified sperm separately from non-sperm cells may be performed in parallel at any visit where semen samples are collected. The shedding analysis of a fractionated semen sample will only be performed if vector DNA was detected in the whole semen sample for the same visit. Fractionation of semen during shedding analysis may be stopped if purified sperm tested positive for vector DNA on at least three visits, or if purified sperm tested below the limit of detection for vector DNA on at least three consecutive visits.

Contraception use may need to be extended beyond 12 weeks in individual subjects based on observed vector shedding in semen. After 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples below the limit of detection (upon consultation between the Investigator and Medical Monitor).

Details for sample collection and storage are provided in the Laboratory Manual.

10 REPORTING ADVERSE EVENTS

10.1 Safety Parameters and Definitions

Safety assessments will consist of monitoring and recording protocol-defined AEs and SAEs; measurement of protocol-specified hematology, clinical chemistry, and urinalysis variables; measurement of protocol-specified vital signs; and other protocol-defined events of special interest that are deemed critical to the safety evaluation of the study drug.

10.1.1 Adverse Events

For this protocol, an adverse event (AE) is any untoward medical occurrence in a subject, temporally associated with the use of study intervention, whether or not considered related to the study intervention.

An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of study intervention.

Events meeting the AE definition include:

- Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition.
- New conditions detected or diagnosed after study intervention administration even though it may have been present before the start of the study.
- Signs, symptoms, or the clinical sequelae of a suspected drug-drug interaction.

Events not meeting the AE definition include:

- Any clinically significant abnormal laboratory findings or other abnormal safety assessments which are associated with the underlying disease, unless judged by the investigator to be more severe than expected for the subject's condition.
- The disease/disorder being studied or expected progression, signs, or symptoms of the disease/disorder being studied, unless more severe than expected for the subject's condition.
- Medical or surgical procedure (eg, endoscopy, appendectomy): the condition that leads to the procedure is the AE.
- Situations in which an untoward medical occurrence did not occur (social and/or convenience admission to a hospital).
- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

10.1.1.1 Bleeding and Suspected Bleeding Events

All bleeding events and suspected bleeding events, regardless of the need for exogenous FVIII therapy as treatment, should be captured in subject diaries and recorded on the designated bleeding eCRF. Bleeding events and suspected bleeding events should not be reported as adverse events, with the following exception:

- All bleeding events and suspected bleeding events which meet one or more of the criteria for being serious (refer to Section 10.1) should be reported as serious adverse events (whether or not they are bleeding events that are normal sequelae of hemophilia, and whether or not they required exogenous FVIII as treatment).

10.2 Serious Adverse Events

A serious adverse event (SAE) is any untoward medical occurrence that, at any dose:

- Results in death
- Is life-threatening

Note: Life-threatening refers to an event in which the subject was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.

- Requires inpatient hospitalization or prolongation of existing hospitalization
    - Note: In general, hospitalization signifies that the subject has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization, the event is serious.

Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE.

- Results in persistent or significant disability or incapacity
- Is a congenital anomaly or birth defect in the child or fetus of a subject exposed to IP prior to conception or during pregnancy
- Is an important medical event or reaction – that, based on medical judgment, may jeopardize the subject or require medical/surgical intervention to prevent one of the other outcomes listed above (eg, anaphylaxis)

10.2.1 Events of Special Interest (EOSI)

The following EOSI need to be reported to the Sponsor within 24 hours of site awareness, irrespective of seriousness, severity or causality:

- Elevation of ALT > ULN or ≥1.5x baseline value, regardless of whether that elevation triggers an initiation or modification of oral corticosteroid treatment
- Events potentially meeting the criteria for Hy's law (ALT or AST elevation ≥ 3x ULN plus total bilirubin ≥ 2x ULN)
- Thromboembolic event
- Systemic hypersensitivity, anaphylactic, or anaphylactoid reactions (refer to Appendix 1)
- Development of anti-FVIII inhibitory antibodies (inhibitors)

10.3 Methods and Timing for Capturing and Assessing Safety Parameters

10.3.1 Adverse Event Reporting Period

The study AE reporting period is as follows: After informed consent but prior to initiation of study drug, only SAEs associated with any protocol-imposed interventions will be collected. After informed consent is obtained and following infusion of study drug, the reporting period for all non-serious AEs and SAEs begins and continues for approximately 5 years or until study discontinuation/termination, whichever is longer. The criteria for determining, and the reporting of SAEs is provided in Section 10.1.

10.3.2 Eliciting Adverse Events

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and non-leading verbal questioning of the subject is the preferred method to inquire about AE occurrences. The Investigator will record all relevant AE/SAE/EOSI information in the subject's medical record and AE Case Report Form (eCRF).

10.3.3 Assessment of Seriousness, Severity, and Causality

The Investigator responsible for the care of the subject or medically qualified designee will assess AEs for severity, relationship to study drug and/or corticosteroids and/or other immunosuppressive agents, and seriousness (refer to Section 10.2 for SAE definitions). These assessments must be made by a study clinician with the training and authority to make a diagnosis (eg, MD/DO, physician's assistant, nurse practitioner, or DDS).

10.3.3.1 Seriousness

The Investigator will assess if an AE should be classified as "serious" based on the seriousness criteria enumerated in Section 10.2. Seriousness serves as a guide for defining regulatory reporting obligations.

10.3.3.2 Severity

Severity (as in mild, moderate, or severe headache) is not equivalent to seriousness, which is based on subject/event outcome or action criteria usually associated with events that pose a threat to a subject's life or functioning. The Investigator will determine the severity of each AE, SAE and EOSI using the NCI CTCAE v4.03. Adverse events that do not have a corresponding CTCAE term will be assessed according to the general guidelines for grading used in the CTCAE v4.03 as stated in Table 10.3.3.2.1.

Table 10.3.3.2.1: Adverse Event Grading (Severity) Scale

Grade	Description	
1	Mild: asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated	
2	Moderate: minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living (ADL)[a]	
3	Severe or medically significant but not immediately life-threatening: hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL[b]	
4	Life threatening consequences; urgent intervention indicated	Grade 4 and 5 AEs should always be reported as SAEs
5	Death related to AE	

[a] Instrumental ADL refer to the following examples: preparing meals, shopping for groceries or clothes, using the telephone, managing money.

[b] Self-care ADL refer to the following examples: bathing, dressing and undressing, feeding oneself, using the toilet, taking medications, not bedridden.

10.3.3.3 Causality

The Investigator will determine the relationship of an AE to the study drug and/or corticosteroids and/or other immunosuppressive agents and will record it on the source documents and AE eCRF. To ensure consistency of causality assessments, Investigators should apply the guidance in Table 10.3.3.3.1.

Table 10.3.3.3.1: Causality Attribution Guidance

Relationship	Description
Not Related	• Exposure to the IP and/or corticosteroids and/or other immunosuppressive agents has not occurred **OR** • The administration of the IP and/or corticosteroids and/or other immunosuppressive agents and the occurrence of the AE are not reasonably related in time **OR** • The AE is considered likely to be related to an etiology other than the use of the IP and/or corticosteroids and/or other immunosuppressive agents; that is, there are no facts, evidence, or arguments to suggest a causal relationship to the IP and/or corticosteroids and/or other immunosuppressive agents.
Related	• The administration of the IP and/or corticosteroids and/or other immunosuppressive agents and the occurrence of the AE are reasonably related in time **AND** • The AE could not possibly be explained by factors or causes other than exposure to the IP and/or corticosteroids and/or other immunosuppressive agents **OR** • The administration of IP and/or corticosteroids and/or other immunosuppressive agents and the occurrence of the AE are reasonably related in time **AND** • The AE is more likely explained by exposure to the IP and/or corticosteroids and/or other immunosuppressive agents than by other factors or causes.

Factors suggestive of a causal relationship could include (but are not limited to):

- Plausible temporal relationship
- Absence of alternative explanations
- Rarity of event in a given patient or disease state
- Absence of event prior to study drug and/or corticosteroid and/or other immunosuppressive agent exposure
- Consistency with study product pharmacology

- Known relationship to underlying mechanism of study drug and/or corticosteroid and/or other immunosuppressive agent action
- Similarity to adverse reactions seen with related drug products
- Abatement of AE with discontinuation of study drug and/or corticosteroids and/or other immunosuppressive agents, and/or recurrence of AE with reintroduction of study drug and/or corticosteroids and/or other immunosuppressive agents

The Investigator's assessment of causality for individual AE reports is part of the study documentation process. Regardless of the Investigator's assessment of causality for individual AE reports, the Sponsor will promptly evaluate all reported SAEs against cumulative product experience to identify and expeditiously communicate possible new safety findings to Investigators and applicable regulatory authorities.

10.4 Procedures for Recording Adverse Events

10.4.1 Recording Adverse Events on a eCRF

Investigators should use precise medical terminology when recording AEs or SAEs on the AE eCRF. Avoid colloquialisms and abbreviations.

Record only one diagnosis, sign, or symptom per event field on the AE eCRF (eg, nausea and vomiting should not be recorded in the same entry, but as 2 separate entries).

In order to classify AEs and diseases, preferred terms will be assigned by the Sponsor to the original terms entered on the AE eCRF, using MedDRA (Medical Dictionary for Regulatory Activities) terminology.

10.4.1.1 Diagnosis versus Signs and Symptoms

The Investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE. Using accepted medical terminology, enter the diagnosis (if known). If not known, enter sign(s) and/or symptom(s). If a diagnosis subsequently becomes available, then this diagnosis should be entered on the AE (or SAE, as appropriate) eCRF, replacing the original entries where appropriate.

10.4.1.2 Persistent or Recurrent Adverse Events

A persistent AE (duration of adverse event > 7 days) is one that extends continuously, without resolution, between subject evaluation time points. Events that change in severity necessitate the recording of an additional AE. AEs that do not have a change in severity should be recorded only once on the eCRF.

A recurrent AE is one that occurs and resolves between subject evaluation time points, but then subsequently recurs. All recurrences of the AE should be recorded on the AE eCRF. For example, if a subject has an adverse event of ALT increased that subsequently resolves, but the subject's ALT increases again, that should be reported as two adverse events – the initial ALT increase, and the second ALT increase.

10.4.1.3 Abnormal Laboratory Values

Laboratory test results (including any local FVIII activity or liver test results) will be recorded on the laboratory results pages of the eCRF, or appear on electronically produced laboratory reports submitted directly from the central laboratory, if applicable.

Any laboratory result abnormality fulfilling the criteria for a SAE or EOSI should be reported as such, and recorded in the AE eCRF unless associated with an AE that has already been reported.

Any laboratory result abnormality of CTCAE Grade 4 or 5 should be recorded as an SAE in the AE eCRF unless associated with an AE that has already been reported.

A clinical laboratory abnormality is considered clinically significant and should be documented as an AE if not refuted by a repeat test to confirm the abnormality and **any** one or more of the following conditions is met:

- Accompanied by clinical symptoms
- Requiring a change in concomitant therapy (eg, addition of, interruption of, discontinuation of, or any other change in a concomitant medication, therapy or treatment).
- The abnormality suggests a disease and/or organ toxicity
- The abnormality is of a degree that requires active management (eg, change of dose, discontinuation of study drug, more frequent follow-up assessments, further diagnostic investigation, etc.)

This applies to any protocol and non-protocol specified safety and efficacy laboratory result from tests performed after the first dose of study medication that falls outside the laboratory reference range and meets the clinical significance criteria.

This does not apply to any abnormal laboratory result that falls outside the laboratory reference range but that does not meet the clinical significance criteria (these will be analyzed and reported as laboratory abnormalities), those that are considered AEs of the type explicitly exempted by the protocol, or those which are a result of an AE that has already been reported.

For purposes of this study, laboratory tests showing a decreased level of FVIII activity should not be reported as adverse events unless there is an impact to clinical outcomes (eg, increased rate of bleeding, worsening of joint disease).

10.4.1.4 Pre-existing Conditions

A pre-existing condition is one that is present prior to administration of BMN 270. Such conditions should be recorded as medical history on the appropriate eCRF.

A pre-existing condition should be recorded as an AE or SAE during the study **only** if the frequency, intensity, or character of the condition worsens during the study period. It is important to convey the concept that a pre-existing condition has changed by including applicable language in the verbatim description of the event (eg, *more frequent* headaches).

10.4.1.5 General Physical Examination Findings

At screening, any clinically significant abnormality should be recorded as a pre-existing condition (refer to Section 10.4.1.5). During the study, any new clinically significant findings and/or abnormalities discovered on physical examination that meet the definition of an AE (or an SAE) must be recorded and documented as an AE or SAE on the AE eCRF.

10.4.1.6 Hospitalization, Prolonged Hospitalization, or Surgery

Any AE that results in hospitalization or prolonged hospitalization should be documented and reported as an SAE unless specifically instructed otherwise in this protocol (refer to Section 10.2).

There are some hospitalization scenarios that do not require reporting as an SAE when there is no occurrence of an AE. These scenarios include planned hospitalizations or prolonged hospitalizations to:

- Perform a protocol-mandated efficacy measurement
- Undergo a diagnostic or elective surgical procedure for a pre-existing medical condition that has not worsened
- Insert an in-dwelling IV catheter (such as a Port-a-Cath or other brand, if applicable) for administration of study drug or FVIII replacement therapy
- Receive scheduled therapy (study drug or otherwise) for the study indication

10.4.1.7 Deaths

All deaths that occur during the AE reporting period (refer to Section 10.3.1), regardless of attribution, will be recorded on the AE eCRF and expeditiously reported to the Sponsor as an SAE.

When recording a death, the event or condition that caused or contributed to the fatal outcome should be recorded as the single medical concept on the eCRF. If the cause of death is unknown and cannot be ascertained at the time of reporting, record "Unexplained Death" or "Death of Unknown Cause" on the AE eCRF.

10.4.1.8 Pregnancy

Although not an AE per se, pregnancy in the partner of a subject taking trial medication should be reported expeditiously to the Sponsor to facilitate outcome monitoring by the Sponsor. Pregnancy in partner should be reported during the period up to 5 years after viral infusion.

Pregnancy in a partner should be reported within 24 hours of the site becoming aware of the pregnancy by entering the information on the Pregnancy eCRF and submitting to BPV within 24 hours of the site becoming aware of the event. The Investigator must make every effort to follow the subject's partner (with that partner's consent) through resolution of the pregnancy (delivery or termination) and to report the resolution on the Pregnancy Follow-up eCRF. In the event of pregnancy in the partner of a study subject, the Investigator should make every reasonable attempt to obtain the woman's consent for release of protected health information.

Abortion, whether therapeutic or spontaneous, should always be classified as an SAE (as the Sponsor considers these to be medically significant), recorded on the AE eCRF, and expeditiously reported to the Sponsor as an SAE.

10.5 Reporting Requirements

10.5.1 Expedited Reporting Requirements

All SAEs and EOSI that occur during the course of the AE Reporting Period (refer to Section 10.3.1), whether or not considered related to study drug, must be reported by entering the information in the AE eCRF and submitting to BPV within 24 hours of the site becoming aware of the event. Investigators should not wait to collect information that fully documents the event before notifying BPV of an SAE. BioMarin may be required to report certain SAEs to regulatory authorities within 7 calendar days of being notified about the event; therefore, it is important that Investigators submit any information requested by BioMarin as soon as it becomes available. IND safety reports will be submitted within 7 calendar days for unexpected fatal or life-threatening unexpected suspected adverse reactions (SUSARs) and within 15 calendar days for other non-life-threatening SUSARs.

The Sponsor is responsible for identifying, preparing and reporting all SUSARs to the relevant competent authorities, ethics committees and Investigators in accordance with the requirements identified in the Clinical Trials Regulations.

If the EDC is unavailable, all SAEs should be reported to BPV by completing the SAE Report Form and faxing or emailing the completed form to BPV within 24 hours of the site becoming aware of the event. Once the EDC is available, the information should be entered in the AE eCRF.

10.5.2 Institutional Review Board or Independent Ethics Committee Reporting Requirements

Reporting of SAEs to the Independent Ethics Committee (EC) or Institutional Review Board (IRB) will be done in compliance with the standard operating procedures and policies of the IRB/EC and with applicable regulatory requirements. Adequate documentation must be obtained by BioMarin showing that the IRB/EC was properly and promptly notified as required.

10.6 Follow-up of Subjects after Adverse Events

After the initial AE/SAE/EOSI report, the Investigator is required to proactively follow each subject at subsequent visits/contacts. All AEs/SAEs/EOSI will be followed until resolution, stabilization, the event is otherwise explained, or the subject is lost to follow-up. Resolution of AEs/SAEs/EOSI (with dates) should be documented on the AE eCRF and submitted to BioMarin Pharmacovigilance and in the subject's medical record to facilitate source data verification.

For some SAEs and EOSI, the Sponsor may follow up by telephone, fax, electronic mail, and/or a monitoring visit to obtain additional case details (eg, hospital discharge summary, consultant report, or autopsy report) deemed necessary to appropriately evaluate the SAE or EOSI report.

10.7 Post-Study Adverse Events

At the last scheduled visit, the Investigator should instruct each subject to report, to the Investigator and/or to BPV directly, any subsequent SAEs that the subject's personal physician(s) believes might be related to prior study drug.

The Investigator should notify the study Sponsor of any death or SAE occurring at any time after a subject has discontinued or terminated study participation, if the Investigator believes that the death or SAE may have been related to prior study drug. The Sponsor should also be notified if the Investigator should become aware of the development of cancer or of a congenital anomaly in a subsequently conceived offspring of a subject that participated in this study.

10.8 Urgent Safety Measures

The regulations governing clinical trials state that the Sponsor and Investigator are required to take appropriate urgent safety measures to protect subjects against any immediate hazards that may affect the safety of subjects, and that the appropriate regulatory bodies should be notified according to their respective regulations. According to the European Union (EU) Clinical Trial Directive 2001/20/EC, "…in the light of the circumstances, notably the occurrence of any new event relating to the conduct of the trial or the development of the investigational medicinal product where that new event is likely to affect the safety of the patients, the Sponsor and the Investigator shall take appropriate urgent safety measures to protect the patients against any immediate hazard. The Sponsor shall forthwith inform the competent authorities of those new events and the measures taken and shall ensure that the IRB/EC is notified at the same time."

The reporting period for these events which may require the implementation of urgent safety measures is the period from the time of signing of the ICF through the completion of the last study visit or at the Early Termination Visit (ETV). Investigators are required to report any events which may require the implementation of urgent safety measures to BioMarin within 24 hours.

Examples of situations that may require urgent safety measures include discovery of the following:

- Lack of study scientific value, or detrimental study conduct or management
- Discovery that the quality or safety of the IP does not meet established safety requirements

10.9 BioMarin Pharmacovigilance Contact Information

Contact information for BioMarin Pharmacovigilance is as follows:

BioMarin Pharmaceutical Inc.

Address	105 Digital Drive Novato, CA 94949
Phone:	PI
Fax:	PI
E-mail:	drugsafety@bmrn.com

The Investigator is encouraged to discuss with the medical monitor any AEs for which the issue of seriousness is unclear or questioned. Contact information for the medical monitor is as follows:

Name: PI , MA MB BChir MSc
Address: BioMarin (UK) Ltd.
10 Bloomsbury Way
London WC1A 2SL
Phone PI (office)
PI (mobile)
E-mail: PI

11 APPROPRIATENESS OF MEASUREMENTS

The measures of efficacy to be used in this study are standard, ie, widely used and generally recognized as reliable, accurate, and relevant (able to discriminate between effective and ineffective agents). The measures of safety used in this study are routine clinical and laboratory procedures.

The chromogenic substrate FVIII assay and the one-stage clotting FVIII assay are both validated and utilize CE marked reagents.

12 STUDY PROCEDURES

12.1 Prestudy

An ICF must be signed and dated by the subject, the Investigator or designee and witness (if required) before any study-related procedures are performed.

12.2 Screening Visit(s)

Screening assessments should be performed within 42 days of BMN 270 infusion, while baseline assessments will take place within 7 days prior to BMN 270 infusion (Day 1). Should the screening visit occur within 30 days of the drug infusion, physical examination, vital signs, blood chemistry, LTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline.

During the first part of the Screening Period (Day -42 to Day -29), testing for AAV5 TAb titers using the ARUP screening assay may be performed, so that subjects can verify their AAV5 TAb status. Subjects who agree to participate in this activity will be asked to sign a separate ICF documenting this decision. Subjects who do not agree will have the ARUP AAV5 TAb screening assay performed along with other assessments during the regular Screening period.

The following procedures will be performed during the Screening Period (Day -28 to Day -1):

- Demographics (age, sex, race, ethnicity)
- Full medical history, including hemophilia A history, Hepatitis B, Hepatitis C, and HIV. Subjects with a history of hepatitis B or hepatitis C will be asked for information about the treatments received. Any prior pharmacokinetics information obtained while the subject was receiving prophylactic or on-demand FVIII therapy prior to the study should also be collected.
- Complete Physical Examination
- Height and weight
- Vital Signs (systolic and diastolic blood pressure, heart rate, respiration rate, and temperature)
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information) for the previous 12 months
- Distribution of subject diaries and training in diary completion
- Electrocardiogram

- Liver Ultrasound/FibroScan
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay
    - hFVIII coagulation activity exploratory assay (collected but not tested prior to enrollment)
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
    - hFVIII total antibody assay (collected but not tested prior to enrollment)
    - hFVIII antigen assay (collected but not tested prior to enrollment)
- Blood sample for ARUP AAV5 TAb assay
    - Subjects who underwent the ARUP AAV5 TAb assay test earlier in the Screening period with the standalone consent do not need to repeat the test as part of regular Screening.
- Screen for Hepatitis B, Hepatitis C, and HIV if required (subjects with documented negative results 30 days prior to informed consent being obtained do not need to be retested)
    - Hepatitis B screening should include HBsAg, HBsAb, and HBcAb
- Screen for COVID-19 (local or central testing)
    - COVID-19 RT-PCR testing is required during Screening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing.
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)
- Blood samples for Biomarker testing (may include HLA genotyping and FVIII genotyping status)

12.2.1 "Smart Rescreening" Visit

Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once.

If a patient has to be screened again because the original assessments have fallen out of the 28 + 14 day period allowed for Screening (refer to Section 12.2), then only the following assessments need to be performed (rather than the full list indicated in Section 12.2) for the patient to be successfully re-screened for the study:

- Vital signs
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- hFVIII Assays (only the hFVIII inhibitor level (Bethesda assay with Nijmegen modification))
- AAV5 TAb assay (ARUP)
- Screen for COVID-19 (local or central testing)
    - COVID-19 RT-PCR testing is required during smart rescreening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing.
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)

12.3 Baseline Visit

Baseline values will be recorded from 1 to 7 days prior to the infusion visit. The following procedures will be performed during the Baseline Period:

- Brief physical examination
- Vital signs
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay
    - hFVIII coagulation activity exploratory assay
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)

  - hFVIII total antibody assay
  - hFVIII antigen assay
- Von Willebrand Factor Antigen (VWF:Ag)
- Blood sample for AAV5 Total Antibody assay
  - Baseline sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
  - ABO blood typing assessment should be performed at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study.
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)
- PBMC collection for CTL baseline
- Blood sample for AAV5 TI assay
- Thrombin Generation Assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools
- Exploratory biomarker assessments
- Cytokine bead array assay
- Hypersensitivity blood assessments
- Haemo-QoL-A assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.4 Treatment Visit/BMN 270 Infusion Visit (Day 1)

There will be one infusion visit for each subject. Subjects will remain in the clinic for at least 24 hours for the BMN 270 Infusion Visit. The following procedures will be performed during the BMN 270 Infusion Visit:

- Brief physical examination
- Start prophylactic corticosteroids (pre-infusion)
- Assessment of Adverse Events and Concomitant Medications

- AAV5 TAb assay (ARUP) (sample collected pre-infusion for analysis)
- Blood sample for AAV5 TI assay (sample collected pre-infusion for analysis)
- Fasting blood sample for future exploratory analysis (sample collected pre-infusion)
- Fasting lipid panel (blood triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) (sample collected pre-infusion)
    - Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit.
- Fasting FibroTest
- BMN 270 Infusion
- Complement panel
    - Complement panel should include C3, C3a, C4, Bb, and sC5b-9 and should be collected within 2 hours post-BMN 270 infusion.
- Vital Signs
    - Vital signs will be recorded prior to BMN 270 infusion and then every 15 minutes (± 5 minutes) during BMN 270 infusion. Following infusion, vital signs will be monitored every 1 hour (± 5 minutes) for at least 8 hours during the subject's stay in the clinic.
- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Collection of samples for PCR testing should occur between 2 and 24 hours after the BMN 270 infusion has been completed

In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine bead array. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.

Subjects should also start prophylactic corticosteroids, prior to BMN 270 infusion, on Study Day 1 (refer to Table 9.1.6 for a possible prophylactic corticosteroid regimen, and to

Section 9.4.8.2 further discussion). Clinical judgment, weighting the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses. Discussions between the Investigator and Medical Monitor are advised for any questions or concerns.

12.5 BMN 270 Infusion Follow-Up Visits

At applicable sites, certain study assessments may be performed by a mobile nursing (MN) professional at the patient's home or another suitable location, such as their school or office, to improve access and convenience for patients participating in the study. The Sponsor may select a healthcare company that will be responsible for providing MN services for participating sites (the MN vendor). The MN vendor is responsible for ensuring that all MN professionals are licensed, qualified, and in good standing, as per applicable regulations, and that appropriate background checks have been performed. If the investigator at a participating site determines that MN services are appropriate for a patient and the patient gives written informed consent to participate in MN visits, the MN network will communicate with the patient and the patient's site. Potential MN visits are designated in the Schedules of Events.

12.5.1 Week 1

During Week 1, the subject will be assessed on Study Day 2, Study Day 4, and Study Day 8.

12.5.1.1 Week 1, Study Day 2

On Study Day 2, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Separate collection on Day 2 is not required if vector shedding samples were collected on Day 1 post-BMN 270 infusion; if collection on Day 2 is needed, it must occur within 24 hours of the time of the BMN 270 infusion.
- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay

- PBMC collection
- Complement panel
    - Complement panel should include C3, C3a, C4, Bb, and sC5b-9.
- Cytokine bead array assay

12.5.1.2 Week 1, Study Day 4

On Study Day 4, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
- Complement panel
    - Complement panel should include C3, C3a, C4, Bb, and sC5b-9.
- PCR of vector DNA in blood, saliva, urine, semen, and stools

12.5.1.3 Week 1, Study Day 8

On Study Day 8, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.3.1)
- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- Samples for FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level
    - hFVIII antigen assay

- Complement panel
    - Complement panel should include C3, C3a, C4, Bb, and sC5b-9.
- PCR of vector DNA in blood, saliva, urine, semen, and stools
- Cytokine bead array assay

12.5.2 Weeks 2-26

After Week 1 (Day 8), subjects will return to the study site once a week (± 48 hours) during Weeks 2-26.

12.5.2.1 Once per week (Weeks 2 through 26)

The following procedures will be performed once per week from Weeks 1 through 26:

- Brief physical examination (complete physical examination at Week 26)
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, physical examination will not be performed.
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
    - For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.
- Vital Signs
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, vital signs will not be performed.
- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are > ULN or ≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
- Samples for FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level

- hFVIII antigen assay

12.5.2.2 Every Other Week (Weeks 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, and 26)

The following procedures will be performed every other week (at Weeks 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, and 26):

- Blood sample for AAV5 TAb Assay
  - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection

12.5.2.3 Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26

The following procedure will be performed at Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26:

- PCR of vector DNA in blood, saliva, urine, semen, and stools
  - Collection to occur until at least 3 consecutive results below the limit of detection are obtained. Semen samples should continue to be collected and tested through Week 12, even if 3 consecutive results below the limit of detection in that compartment have been recorded prior to that time point.

12.5.2.4 Weeks 2, 4, 6, 8, 10, and 12

The following procedure will be performed at Weeks 2, 4, 6, 8, 10, and 12:

- Complement panel
  - Complement panel should include C3, C3a, C4, Bb, and sC5b-9.

12.5.2.5 Weeks 2, 4, 6, 8, 10, 12, 16, 20, 24, and 26

The following procedure will be performed at Weeks 2, 4, 6, 8, 10, 12, 16, 20, 24 and 26:

- hFVIII total antibody assay

12.5.2.6 Weeks 2, 4, 6, 8, 14, 20, and 26

The following procedure will be performed at Weeks 2, 4, 6, 8, 14, 20, and 26:

- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)

12.5.2.7 Weeks 2, 6, 10, 14, 18, 22, and 26

The following procedure will be performed at Weeks 2, 6, 10, 14, 18, 22, and 26:

- Exploratory biomarker assessments

12.5.2.8 Weeks 2, 6, 10, 14, 20, 24, and 26

The following procedure will be performed at Weeks 2, 6, 10, 14, 20, 24, and 26:

- Cytokine bead array assay

12.5.2.9 Weeks 4, 8, 12, 16, 20, and 26

The following procedures will be performed at Weeks 4, 8, 12, 16, 20, and 26:

- Weight
- VWF:Ag

12.5.2.10 Weeks 12 and 26

The following procedures will be performed at Weeks 12 and 26:

- Urine tests (to be performed locally)
- Haemo-QoL-A QoL assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.5.2.11 Week 16

The following procedure will be performed at Week 16:

- Testing for reactivation of hepatitis B and hepatitis C (only in subjects with evidence of prior exposure to hepatitis B and/or hepatitis C)
    - Subjects who receive therapeutic oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.6.

12.5.2.12 Weeks 20, 24, and 26

The following procedure will be performed at Weeks 20, 24, and 26:

- Thrombin Generation Assay

12.5.3 Post-Infusion Follow-Up – Weeks 27-52

During Weeks 27-36, subjects will return to the study site weekly (± 48 hours). During Weeks 37-52, subjects will return to the study site every 2 weeks (Week 38, 40, 42, 44, 46, 48, 50, and 52) (± 1 week). At these visits, the following procedures will be completed:

12.5.3.1 Every Visit

At every visit (Weeks 27-36, 38, 40, 42, 44, 46, 48, 50, and 52), the following procedures will be performed:

- Physical examination
    - Brief physical examination should be performed at all weeks except Week 52, when a complete physical examination should be performed
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, physical examination will not be performed.
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
    - For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.
- Vital Signs
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, vital signs will not be performed.
- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are > ULN or ≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
        - FVIII coagulation activity exploratory assay
        - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
        - FVIII protein assay

12.5.3.2 Weeks 28, 30, 32, 34, 36, 40, 44, 48, and 52

At Weeks 28, 30, 32, 34, 36, 40, 44, 48, and 52, the following procedures will be performed:

- AAV5 TAb Assay
- AAV5 TI Assay

12.5.3.3 Weeks 28, 30, 32, 34, 36, 44, and 52

At Weeks 28, 30, 32, 34, 36, 44, and 52, the following procedure will be performed:

- PBMC collection

12.5.3.4 Weeks 32, 36, 44, and 52

At Weeks 32, 36, 44, and 52, the following procedures will be performed:

- Weight
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.4.8.2.1)
- FVIII antibody titer
- Cytokine bead array assay

12.5.3.5 Weeks 32, 36, 40, 44, 48, and 52

At Weeks 32, 36, 40, 44, 48, and 52, the following procedures will be performed:

- Exploratory biomarker assessments
- TGA Assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools
  - Sample testing to occur until at least 3 consecutive sample below the limit of detection results have been obtained. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 52 should continue to have PCR testing of semen every 6 weeks until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).
  - Subjects considered to be treatment failures must continue to provide samples for PCR assessment at these timepoints until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints)

12.5.3.6 Week 36 and 52

At Weeks 36 and 52, the following procedures will be performed:

- Urine Tests (refer to Table 9.7.6.2.1)
- VWF:Ag

12.5.3.7 Week 52

At Week 52, the following procedures will be performed:

- Haemo-QoL-A assessment

- EQ-5D-5L
- HAL
- WPAI+CIQ:HS

12.6 Post-Infusion Follow-Up – Years 2-5

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 52 of the study by attending only the Q12W and End of Year visits during Years 2-5.

Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from semen must still provide semen samples for assessment every 6 weeks during Years 2-5 until vector shedding has cleared (but do not have to perform other scheduled assessments at those study visits). Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.

During Years 2-5 of Post-Infusion Follow-up, the following procedures will be completed:

12.6.1 Years 2-5 – Every 6 Weeks (not required for treatment failure)

During Years 2-5, every 6 weeks (± 2 weeks), the following procedures will be performed:

- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
    - For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.
- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are > ULN or ≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)

- FVIII activity level (one-stage clotting FVIII assay)
- FVIII coagulation activity exploratory assay
- Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
  - If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.
- FVIII protein assay

- PCR of vector DNA in semen (if required)
  - Sample testing during Years 2-5 is not required if at least 3 consecutive samples are clear by the end of Year 1. Subjects who have not had 3 consecutive semen samples below the limit of detection by the end of Year 1 should continue to have PCR testing of semen every 6 weeks during Years 2-5 until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).
    - Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from semen must still provide semen samples for assessment every 6 weeks during Years 2-5 until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints).

12.6.2 Years 2-5 – Every 12 Weeks and End of Year Visits (required for all subjects)

During Years 2-5, subjects will be asked to return to the study site for visits at the following study weeks (±2 weeks):

- Year 2 – Week 64, Week 76, Week 88, Week 104
- Year 3 – Week 116, Week 128, Week 140, Week 156
- Year 4 – Week 168, Week 180, Week 192, Week 208
- Year 5 – Week 220, Week 232, Week 244, Week 260

For each of these years, the last study visit listed (Week 104, Week 156, Week 208, and Week 260) will serve as an End of Year visit.

At the every 12 week and End of Year visits, the following procedures will be performed:

- Brief physical examination (at every 12 week visits) or complete physical examination (at End of Year visits)
- Weight (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)

- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
- Liver Tests (refer to Table 9.7.6.3.1)
    - LTs may be monitored more or less frequently (and in particular when ALT values are > ULN or ≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥3x ULN.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
        - If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.
    - FVIII protein assay
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1) (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Urine Tests (refer to Table 9.7.6.2.1) (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Vital Signs
- AAV5 TAb Assay
- AAV5 TI Assay
- FVIII antibody titer
- Haemo-QoL-A assessment (at End of Year visits only)
- EQ-5D-5L (at End of Year visits only)
- HAL (at End of Year visits only)
- WPAI+CIQ:HS (at End of Year visits only)
- Exploratory biomarker assessments
- PBMC collection
- VWF:Ag
- TGA Assay

- PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)
    - Sample testing during Years 2-5 is not required if at least 3 consecutive samples are below the limit of detection during the Post-Infusion Follow-Up period in Weeks 1-52.

12.7 Early Termination Visit

The Early Termination visit will occur on the date the subject withdraws from the study, even if the date does not correspond to a protocol-specific visit.

If a subject leaves the study prior to the Week 260 visit, the subject will be asked to return to the study site and complete an Early Termination visit. At the Early Termination visit, as many of the following assessments as possible should be done:

- Complete physical examination
- Weight
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Vital signs
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.3.1)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay
    - hFVIII coagulation activity exploratory assay
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
    - hFVIII antigen assay
- hFVIII total antibody assay
- Exploratory biomarker assessments
- Blood sample for AAV5 Total Antibody assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay

- Cytokine bead array assay
- PBMC collection for CTL baseline
- VWF:Ag

- Thrombin Generation Assay
- Haemo-QoL-A QoL assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

- PCR of vector DNA in blood, saliva, urine, semen, and stool
    - Sample testing at the Early Termination Visit is not required if at least 3 consecutive samples are clear during the period of the subject's participation in the study.

12.8 End of Study

The study will end after the last subject yet to complete the last Long-Term Follow-Up visit (Week 260) does so, has transferred to another BMN 270 study, is withdrawn from the study, or discontinues from the study. BioMarin reserves the right to discontinue the study any time for clinical or administrative reasons and to discontinue participation of an individual Investigator or site for clinical or administrative reasons, including, but not limited to, poor enrollment or noncompliance with procedures of the protocol or GCP. In addition, the study may be terminated if, in the opinion of BioMarin, the safety of the study subjects may be compromised.

13 DATA QUALITY ASSURANCE

BioMarin personnel or designees will visit the study site prior to initiation of the study to review with the site personnel information about the IP, protocol and other regulatory document requirements, any applicable randomization procedures, source document requirements, CRFs, monitoring requirements, and procedures for reporting AEs, including SAEs.

At visits during and after the study, a CRA will monitor the site for compliance with regulatory documentation, with a focus on accurate and complete recording of data on CRFs from source documents, adherence to protocol, randomization (if applicable), SAE reporting and drug accountability records.

Sites will enter study data into eCRFs into the study EDC system. Data Quality Control will be performed by BioMarin Clinical Data Management or designee through implementation of quality control checks specified in the study data management plan and edit check specifications.

14 STATISTICAL METHODS AND DETERMINATION OF SAMPLE SIZE

14.1 Statistical and Analytical Plans

The statistical analysis plan (SAP) will provide additional details on the planned statistical analysis. Unless otherwise stated, all analyses will be performed using SAS.

14.1.1 Interim Analyses

No interim analysis is planned.

14.1.2 Procedures for Accounting for Missing, Unused and Spurious Data

Because the completeness of the data affects the integrity and accuracy of the final study analysis, every effort should be made to ensure complete, accurate, and timely data collection and, therefore, avoid missing data.

Sensitivity analyses will be conducted to assess the impact of missing data on the primary efficacy endpoint analysis. Additional details regarding the handling of missing data will be provided in the SAP.

14.2 Efficacy Analysis

The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using longitudinal methods, such as mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at $\geq$ 5 IU/dL at 26 weeks following study treatment. FVIII activity will be assessed weekly during the study period. We can only assess the true steady state of FVIII produced from BMN 270 after a minimum of 72 hours (or 5x the known half-life of the FVIII replacement therapy administered) has elapsed since the last infusion of FVIII concentrates.

14.3 Safety Analysis

The Medical Dictionary for Regulatory Activities terminology (MedDRA) will be used by the Sponsor to assign system organ class and preferred term classification to events and diseases, based on the original terms entered on the eCRF.

All AEs will be coded using the current version of MedDRA. The incidence of AEs will be summarized by system organ class, preferred term, relationship to study drug, and severity. A by-subject listing will be provided for those subjects who experience a serious AE (SAE), including death, or experience an AE associated with early withdrawal from the study or study drug.

Clinical laboratory data will be summarized by the type of laboratory test. For each clinical laboratory test, descriptive statistics will be provided on Baseline as well as all subsequent visits. Descriptive statistics for physical examination results and vital signs will also be provided.

Analysis of neutralizing antibody response and other immunological parameters as well as vector shedding will be primarily descriptive and involve both inter-subject and intra-subject comparisons across doses.

Detailed statistical methods will be provided in the SAP.

14.4 Determination of Sample Size

The sample size is based upon clinical considerations and is sufficient to detect a strong clinical efficacy signal. Approximately 10 subjects may be dosed in the study.

14.5 Analysis Populations

The efficacy analysis set will be comprised of all subjects who have received the BMN 270 infusion.

The safety population will consist of all subjects who receive BMN 270 infusion during the study.

14.6 Changes in the Conduct of the Study or Planned Analyses

Only BioMarin may modify the protocol. Any change in study conduct considered necessary by the Investigator will be made only after consultation with BioMarin, who will then issue a formal protocol amendment to implement the change. The only exception is when an Investigator considers that a subject's safety is compromised without immediate action. In these circumstances, immediate approval of the chairman of the IRB/EC must be sought, and the Investigator should inform BioMarin and the full IRB/EC within 2 working days after the emergency occurs.

With the exception of minor administrative or typographical changes, the IRB/EC must review and approve all protocol amendments. Protocol amendments that have an impact on subject risk or the study objectives, or require revision of the ICF, must receive approval from the IRB/EC prior to their implementation.

When a protocol amendment substantially alters the study design or the potential risks or burden to subjects, the ICF will be amended and approved by BioMarin and the IRB/EC, and all active subjects must again provide informed consent.

15 DATA MONITORING COMMITTEE

The Data Monitoring Committee (DMC) will consist of experts in clinical trials, statistics, and hemophilia. The DMC will review available safety and efficacy data during the study on an ongoing basis to determine, based on emerging data and the benefit/risk profile, whether further enrollment should continue.

Duties of the DMC include:

- Conducting an ongoing review of individual subject safety and efficacy data during the study;
- Recommending whether to proceed with enrollment of subjects at a different gating schedules based on emerging data from 270-203 and the overall risk/benefit analysis of BMN 270;
- If applicable, considering whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction.
- Making other recommendations on the conduct and reporting of the trial based on their evaluation of clinical data including institution of any pause or stopping stages.

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC determination of safety and efficacy in treated subjects with the following triggers for pausing further enrollment:

- any related SAE;
- any related AE with a severity > Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

16 COSTS, COMPENSATION, AND SUBJECT INJURY

BioMarin will pay the full costs of the study-related tests, procedures, and treatments set forth in the protocol. In addition, after IRB/EC approval, BioMarin may reimburse the reasonable cost of travel for study-related visits in accordance with BioMarin's travel and reimbursement policy.

The Investigator should contact BioMarin immediately upon notification that a study subject has been injured by the study drug or by procedures performed as part of the study. Any subject who experiences a study-related injury should be instructed by the Investigator to seek immediate medical treatment at a pre-specified medical institution if possible, or at the closest medical treatment facility if necessary. The subject should be given the name of a person to contact to seek further information about, and assistance with, treatment for study-related injuries. BioMarin or the institution may pay for reasonable and necessary medical services to treat the injuries caused by the study drug or study procedures. In some jurisdictions, BioMarin is obligated by law to pay for study-related injuries. If this is the case, BioMarin will comply with the law. BioMarin will not pay for any hospitalizations, tests, or treatments for medical problems of any sort related solely to the study subject's primary disease or any concurrent disease that are unrelated to this study.

17 CASE REPORT FORMS AND SOURCE DOCUMENTS

Electronic case report forms will be provided for each subject. The Investigator must review and electronically sign the completed eCRF casebook to verify its accuracy.

eCRFs must be completed using a web–based application developed and validated. Study site personnel will be trained on the application and will enter the clinical data from source documentation. Unless explicitly allowed in the eCRF instructions, blank data fields are not acceptable.

In the event of an entry error, or if new information becomes available, the value will be corrected by deselecting the erroneous response and then selecting or entering the factual response. In compliance with ICH GCP Guidelines and 21 CFR Part 11, the system will require the personnel making the correction to enter a reason for changing the value. The documented audit trail will include the reason for the change, the original value, the new value, the time of the correction and the identity of the operator.

BioMarin's policy is that study data on the eCRFs must be verifiable to the source data, which necessitates access to all original recordings, laboratory reports, and subject records. In addition, all source data should be attributable (signed and dated). The Investigator must therefore agree to allow direct access to all source data. Subjects must also allow access to their medical records, and subjects will be informed of this and will confirm their agreement when giving informed consent. If direct source document verification of study data by the site monitor is prohibited by institutional policy or local law, then the Investigator must make available facilities and/or personnel to allow GCP-compliant source verification to occur. Examples of such methods include certified copies of records which have study data visible but sensitive information redacted, or other GCP-compliant means agreed between the Investigator and the Sponsor.

A site monitor designated by BioMarin will compare the eCRFs with the original source documents at the study site and evaluate them for completeness and accuracy before designating them as "Source Data Verified" (SDV). If an error is discovered at any time or a clarification is needed, the site monitor, or designee, will create an electronic query on the associated field. Site personnel will then answer the query by either correcting the data or responding to the query. The site monitor will then review the response and determine either to close the query or re–query the site if the response does not fully address the question. This process is repeated until all open queries have been answered and closed.

Before a subject's eCRF casebook can be locked, data fields must be source data verified and all queries closed. Refer to the Study Monitoring Plan for details on which fields must be

source data verified. The Data Manager, or designee, will set the status of the forms, visits, and the entire casebook to Locked. The Investigator will then electronically sign the casebook, specifying that the information on the eCRFs is accurate and complete. Upon completion of the CSR, an electronic copy of each site's casebooks will be copied to a compact disk (CD) and sent to each site for retention with other study documents.

18 STUDY MONITORING AND AUDITING

Qualified individuals designated by BioMarin will monitor all aspects of the study according to GCP and standard operating procedures for compliance with applicable government regulations. The Investigator agrees to allow these monitors direct access to the clinical supplies, dispensing, and storage areas and to the clinical files, including original medical records, of the study subjects, and, if requested, agrees to assist the monitors. The Investigator and staff are responsible for being present or available for consultation during routinely scheduled site visits conducted by BioMarin or its designees.

Members of BioMarin's GCP Compliance Department or designees may conduct an audit of a clinical site at any time before, during, or after completion of the study. The Investigator will be informed if an audit is to take place and advised as to the scope of the audit. Representatives of the FDA or other Regulatory Agencies may also conduct an audit of the study. If informed of such an inspection, the Investigator should notify BioMarin immediately. The Investigator will ensure that the auditors have access to the clinical supplies, study site facilities, original source documentation, and all study files.

19 RETENTION OF RECORDS

The Investigator must retain all study records required by BioMarin and by the applicable regulations in a secure and safe facility. The Investigator must consult a BioMarin representative before disposal of any study records, and must notify BioMarin of any change in the location, disposition or custody of the study files. The Investigator /institution must take measures to prevent accidental or premature destruction of essential documents, that is, documents that individually and collectively permit evaluation of the conduct of a study and the quality of the data produced, including paper copies of study records (eg, subject charts) as well as any original source documents that are electronic as required by applicable regulatory requirements.

All study records must be retained for at least 2 years after the last approval of a marketing application in the US or an ICH region and until (1) there are no pending or contemplated marketing applications in the U.S. or an ICH region or (2) at least 2 years have elapsed since the formal discontinuation of clinical development of the IP. The Investigator /institution should retain subject identifiers for at least 15 years after the completion or discontinuation of the study. Subject files and other source data must be kept for the maximum period of time permitted by the hospital, institution or private practice, but not less than 15 years. These documents should be retained for a longer period, however, if required by the applicable regulatory requirements or by a BioMarin agreement. BioMarin must be notified and will assist with retention should Investigator /institution be unable to continue maintenance of subject files for the full 15 years. It is the responsibility of BioMarin to inform the Investigator /institution as to when these documents no longer need to be retained.

20 USE OF INFORMATION AND PUBLICATION

BioMarin recognizes the importance of communicating medical study data and therefore encourages the publication of these data in reputable, peer-reviewed scientific journals and at seminars or conferences. The details of the processes of producing and reviewing reports, manuscripts, and presentations based on the data from this study will be described in the Clinical Trial Agreement between BioMarin and the Investigator/Institution. Consideration for authorship of all publications will be based on compliance with the Uniform Requirements for Manuscripts Submitted to Biomedical Journals ("Uniform Requirements") of the International Committee of Medical Journal Editors (ICMJE) (http://www.icmje.org/ethical_1author.html) and good publication practices (GPP).

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22 INVESTIGATOR RESPONSIBILITIES

22.1 Conduct of Study and Protection of Human Subjects

In accordance with FDA Form 1572 and/or principles of ICH E6 GCP, the Investigator will ensure that:

- He or she will conduct the study in accordance with the relevant, current protocol and will only make changes in a protocol after notifying the sponsor, except when necessary to protect the safety, rights, or welfare of subjects.
- He or she will personally conduct or supervise the study.
- He or she will inform any potential subjects, or any persons used as controls, that the drugs are being used for investigational purposes, and he or she will ensure that the requirements relating to obtaining informed consent in 21 CFR Part 50 and/or ICH E6 Sections 2.9 and 4.8 are met. As well, he or she will ensure that IRB/EC review and approval in 21 CFR Part 56 and/or ICH E6 Section 2.6 are met.
- He or she will report to the sponsor adverse experiences that occur in the course of the investigation in accordance with 21 CFR 312.64 and/or ICH E6 Section 4.11.
- He or she has read and understands the information in the Investigator's Brochure, including potential risks and side effects of the drug.
- His or her staff and all persons who assist in the conduct of the study are informed about their obligations in meeting the above commitments
- Adequate and accurate records in accordance with 21 CFR 312.62 and/or ICH E6 Section 4.9 are kept, and those records are available for inspection in accordance with 21 CFR 312.68 and/or ICH E6 Section 4.9.7.
- The IRB/EC complies with the requirements of 21 CFR Part 56, ICH E6 Section 3.0, and other applicable regulations, and conducts initial and ongoing reviews and approvals of the study. He or she will also ensure that any change in research activity and all problems involving risks to human subjects or others are reported to the IRB/EC. Additionally, he or she will not make any changes in the research without IRB/EC approval, except where necessary to eliminate apparent immediate hazards to human subjects.
- He or she agrees to comply with all other requirements regarding the obligations of clinical Investigators and all other pertinent requirements in 21 CFR Part 312 and/or ICH E6.

23 SIGNATURE PAGE

Protocol Title: A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5

Protocol Number: 270-203 Amendment 3

I have read the foregoing protocol and agree to conduct this study as outlined. I agree to conduct the study in compliance with all applicable regulations and guidelines, including E6R2 ICH, as stated in the protocol, and other information supplied to me.

Investigator Signature Date

Printed name: _____

Accepted for the Sponsor:

DocuSigned by:
PI
Signer Name: PI
Signing Reason: I approve this document
Signing Time: 24-Aug-2020 | 5:14 AM PDT
D81F76E32EA74F83864C63F8E75F1EBE

Medical Monitor Signature Date

Printed name: PI, MA MB BChir MSc, PI, Clinical Sciences___

24 APPENDIX 1: SAMPSON'S ANAPHYLAXIS CRITERIA

According to the National Institute of Allergy and Infectious Disease/Food Allergy and Anaphylaxis Network (NIAID/FAAN) Second Symposium on the definition and management of anaphylaxis, anaphylaxis is highly likely when any one of the following 3 criteria are fulfilled:

1. Acute onset of an illness (minutes to several hours) with involvement of the skin, mucosal tissue, or both (eg, generalized hives, pruritus or flushing, swollen lips-tongue-uvula)
2. AND AT LEAST ONE OF THE FOLLOWING

a. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced peak expiratory flow [PEF], hypoxemia)

b. Reduced blood pressure (BP) or associated symptoms of end-organ dysfunction (eg, hypotonia [collapse], syncope, incontinence)

3. Two or more of the following that occur rapidly after exposure *to a likely allergen for that patient* (minutes to several hours):

a. Involvement of the skin-mucosal tissue (eg, generalized hives, itch-flush, swollen lips-tongue-uvula)

b. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced PEF, hypoxemia)

c. Reduced BP or associated symptoms (eg, hypotonia [collapse], syncope, incontinence)

d. Persistent gastrointestinal symptoms (eg, crampy abdominal pain, vomiting)

4. Reduced BP after exposure to *known allergen for that patient* (minutes to several hours):

a. Infants and children: low systolic blood pressure (age specific) or greater than 30% decrease is systolic BP

b. Adults: systolic BP of less than 90 mmHg or greater than 30% decrease from that person's baseline.

Source: Sampson, 2006.

25 PROTOCOL AMENDMENT TEXT REVISIONS

The following table summarizes the revisions made to Protocol Amendment 1 and relates the changes to the appropriate rationale (refer to pages 2-4). Added text is indicated by underlined font and deleted text is indicated by ~~strikethrough~~ font.

Section No./Title	Revision	Rationale
2/Synopsis (Study Design and Plan)	Additional blood samples will be collected (tryptase, C3, C3a, C4, ~~C5, C5a~~Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine bead array (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. ... Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.	2, 10
2/Synopsis (Exclusion Criteria)	Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study: 1. Any evidence of active infection, including COVID-19, or any immunosuppressive disorder, including HIV infection. 3. ~~Prior~~Most recent, prior FibroScan or liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used. 7. Liver cirrhosis of any etiology as assessed by liver ultrasound/FibroScan. 16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including ~~possible~~ corticosteroid treatment and/or the use of alternative immunosuppressive agents outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.	1, 6, 8
2/Synopsis (Criteria for Evaluation)	**Safety:** • Liver tests (LTs, including ALT, AST, GGT, LDH, total bilirubin, and alkaline phosphatase)	20
7.4/Summary of Overall Risks and Benefits	~~The majority of subjects in the ongoing 270-201 clinical study who have received 4E13 or 6E13 vg/kg doses of BMN 270 have had Grade 1 asymptomatic elevations in ALT that have reached only slightly above the upper limit of normal (ULN). Subjects in the 6E13 vg/kg cohort received corticosteroids, predominantly on a prophylactic basis, starting 2-4 weeks following BMN 270 infusion, whereas those in the 4E13 vg/kg cohort received corticosteroids only if they experienced an elevation in their ALT ≥ 1.5x ULN (ie, "therapeutic"). Based on the effectiveness of transient oral corticosteroids used starting 8-10 weeks after vector infusion to suppress a presumed Class 1 (cytotoxic T cell) response in prior studies with hepatic transduction with AAV vectors (Mingozzi, 2013), subjects were treated with 7-32 weeks of oral corticosteroids prophylactically or in response to the elevations in ALT to limit hepatotoxicity, ensure preservation of the transduced hepatocytes, and minimize any associated~~	17

Section No./Title	Revision	Rationale
	~~impact of such on FVIII levels. Using this approach, no sustained loss of FVIII activity has been observed in subjects with ALT elevations. Moreover, the rise in ALT levels was not accompanied by significant or lasting abnormalities in other liver tests such as AST, bilirubin or albumin, indicating that extent of toxicity is limited. At the highest dose tested in 270-201 (6E13 vg/kg), the majority of subjects achieved chromogenic FVIII activity levels above 50 IU/dL at 52 weeks post-infusion. Subjects in that cohort also reported markedly decreased bleeding compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. Subjects at all dose levels continue to be followed.~~ ~~In the ongoing 270-301 clinical study, a majority of subjects, all dosed with 6E13 vg/kg BMN 270, have experienced asymptomatic elevations in ALT. Subjects received therapeutic corticosteroids if they experienced an ALT elevation and/or had a FVIII activity level that had declined >35% from peak values. While sporadically observed cytotoxic T-cell responses have not been correlated with ALT elevations, the majority of subjects with transient rises in ALT levels had associated declines in their FVIII activity that subsequently increase to higher than pre-ALT elevation levels following initiation of therapeutic corticosteroids. Similar to 270-201, the rise in ALT levels in 270-301 were not accompanied by significant or lasting aberrations in other liver tests such as AST, bilirubin or albumin, indicating that extent of toxicity is limited. At the time of the 270-301 interim analysis, the modified intent-to-treat population (n=16, who had completed ≥26 weeks on study) had mean and median chromogenic FVIII activity levels of 36.1 and 33.2 IU/dL at 26 weeks, respectively, which are lower than the corresponding values observed for the 270-201 6E13 vg/kg cohort.~~ ~~There has been one HIV-positive subject in the ongoing 270-302 clinical study who experienced Grade 3 asymptomatic elevations in ALT and AST, which has been attributed to an interaction between one or more of his antiretroviral therapy medications and/or unsuspected underlying hepatic disease with BMN 270. In addition, there have been three subjects, including one with Gilbert's syndrome, in the ongoing 270-301 clinical study who have experienced Grade 3 asymptomatic elevations in ALT and AST. These cases have led to the exclusion of subsequent HIV-positive subjects and requirement of liver tests at Screening that are <1.25 times the upper limit of the normal range in the ongoing 270-301 and 270-302 clinical studies. Of note, two HIV-positive subjects in 270-301 and one presumed Gilbert's syndrome subject in 270-201 have received BMN 270 without experiencing any elevations in ALT beyond Grade 1 to date.~~Overall, 151 subjects have received a BMN 270 infusion at one of 4 dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, or 6E13 vg/kg) in one of the four ongoing BMN 270 clinical studies (270-201, 270-301, 270-302, 270-203). Single infusions have been generally well-tolerated across all investigated doses. All subjects have successfully completed their full-dose infusion of BMN 270, with no discontinuation of dosing due to adverse events observed during the infusion. No deaths have been reported in any of the BMN 270 studies, and no participants have discontinued from studies as a result of an adverse event.	

Section No./Title	Revision	Rationale
	Transient ALT elevation (Grade 1 to 3 in severity) has been observed in most subjects administered BMN 270 shortly after dosing, with no evidence for major impacts upon liver function; no events meeting the Hy's Law criteria have been identified. Liver function has remained stable over time. ALT elevations have been reported as events of interest in 13 subjects in 270-201, 1 subject in 270-302, and 91 subjects in 270-301. Although the majority of events have been Grade 1 or Grade 2 in severity, 11 subjects (1 in 270-302 and 10 in 270-301) had a reported Grade 3 ALT elevation. Only one serious event of ALT increased has been reported by investigators (in addition to one event that BioMarin conservatively assessed as serious based on the details of the case). While the majority of ALT elevations responded rapidly to corticosteroids, given current interest in the field of AAV gene therapy for the use of non-steroidal approaches to managing or preventing ALT elevations, alternate non-steroidal systemic immunosuppressive agents have also been used to manage hepatic reactions where corticosteroids have proven to be ineffective or where high doses/and or prolonged exposure to corticosteroids have led to unwanted side effects. Overall, the literature and clinical experience with BMN 270 suggests that transient elevations in liver enzymes are expected following AAV-based gene therapy for the treatment for hemophilia A or B without any long-term concerns of hepatic injury (Manno, 2006; Nathwani, 2011; George, 2016; Miesbach, 2016; Pasi, 2017). ~~As with any infused biological product, there is a potential risk of acute, systemic hypersensitivity reactions (including anaphylaxis) with BMN 270. No hypersensitivity reactions were observed during dosing of BMN 270 in the 270-201 clinical study, although infusion-related hypersensitivity reactions (including anaphylaxis) have been observed during dosing of BMN 270 in the 270-301 clinical study (refer to Investigator's Brochure for full details). All of the infusion-related reactions were effectively managed clinically and resolved without any clinical sequelae.~~ Short-lived infusion reactions associated with one-time BMN 270 administration have included symptoms such as nausea, maculopapular rash, urticaria, diarrhea, watery eyes, rigors, chills, myalgia, fever, tachycardia and hypotension emerging within 24 hours of receiving BMN 270. Most infusion-related reactions were Grade 1 or Grade 2 in severity, and all events resolved, typically within 48 hours following medical management. Three of these cases required temporary interruption of the infusion, followed by re-initiation at a slower rate. All subjects completed their infusions. The reactions with onset during or within approximately 5 hours after the end of infusion responded to treatment with systemic antihistamines and/or corticosteroids, where administered. Infusion-related reactions were effectively mitigated by managing infusion rate and medications. No subjects have experienced thromboembolic events or developed inhibitors to FVIII following BMN 270 infusion.	

Section No./Title	Revision	Rationale
	At the highest dose tested in 270-201 (6E13 vg/kg), the majority of subjects achieved FVIII levels above 50 IU/dL at 52 weeks post-infusion. Subjects in that cohort also reported markedly decreased bleeding compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. Subjects at all dose levels continue to be followed. In 270-301, an interim analysis has shown increased FVIII activity in the majority of subjects to mild HA or normal levels at 26 weeks post-infusion, also with markedly decreased bleeding compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. All subjects who will be included in the final analysis have been dosed with 6E13 vg/kg and continue to be followed.	
9.1/Overall Study Design and Plan	Additional blood samples will be collected (tryptase, C3, C3a, C4, ~~C5, C5a~~Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine bead array (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. ... Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.	2, 10
Schedules of Activities (Table 9.1.1 through Table 9.1.5)	Updates have been made in Table 9.1.1 through Table 9.1.5 consistent with changes made to the table notes and elsewhere in the protocol.	1, 2, 3, 6, 9, 14, 18, 20
Table 9.1.1 (notes)	[d] Screening, Smart Re-screening, and Infusion Day samples will be tested using the ARUP AAV5 TAb assay. During Screening, the ARUP AAV5 TAb assay test may be done first, under a standalone informed consent form, before the main ICF for the study is signed and further screening procedures are performed. Sample collection on the day of the infusion visit must be performed before the BMN 270 infusion is given. If the ARUP AAV5 TAb assay test is done first with the standalone consent, it does not need to be repeated as part of regular Screening. [g] COVID-19 RT-PCR testing is required during Screening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing. [k] Smart rescreening should only be performed if a subject has been determined to be eligible for the study and is unable to complete the Baseline assessments and Infusion prior to the closing of the original Screening window (28 + 14 days). COVID-19 RT-PCR testing is required as part of smart rescreening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing... [o] Complement panel should include C3, C3a, C4, Bb, and sC5b-9 and should be collected within 2 hours post-infusion.	1, 2, 3

Section No./Title	Revision	Rationale
	[g] In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination, vital signs will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, ~~C5, C5a~~Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible...	
Table 9.1.2 (notes)	[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs may be monitored more or less frequently (and in particular when ALT values are ~~≥ 1.5x~~> ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ~~≥ 1.5x~~> ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator... [h] Complement panel should include C3, C3a, C4, Bb, and sC5b-9.	3, 9
Table 9.1.3 (notes)	[a] Brief physical examination should be done at all weekly visits except Week 26, where a complete physical examination should be performed. Weight should be recorded at Week 20, Week 26, and Week ~~26~~32. [b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs may be monitored more or less frequently (and in particular when ALT values are ~~≥ 1.5x~~> ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ~~≥ 1.5x~~> ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator...	9, 14
Table 9.1.4 (notes)	[a] Complete physical examination should be performed at Week 52; brief physical exam may be performed at other study visits. Weight should be recorded at Week 36, Week 44, and ~~every 4 weeks through~~ Week 52. [b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. LTs may be monitored more or less frequently (and in particular when ALT values are ~~≥ 1.5x~~> ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ~~≥ 1.5x~~> ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator...	9, 14

Section No./Title	Revision	Rationale
Table 9.1.5 (notes)	[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.3.1 for liver tests. LTs may be monitored more or less frequently (and in particular when ALT values are ~~≥ 1.5x~~≥ ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. Subjects with ALT ~~≥ 1.5x~~≥ ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator... [g] Subjects who meet the definition of treatment failure to BMN 270 therapy after Week 52 (refer to Section 12.6) may omit the Q6W visits during Years 2-5, and must attend only the Q12W and End of Year visits. Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in ~~all fluids~~semen must still provide samples Q6W during Years 2-5 until vector shedding has been cleared (but do not need to do other scheduled assessments at those visit dates).	9, 10, 18
9.3.1/Inclusion Criteria	Individuals eligible to participate in this study must meet all of the following criteria: 7. Sexually active participants must agree to use an acceptable method of effective contraception, either double barrier contraception (ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with viral vector DNA below the ~~level~~limit of detection.	20
9.3.2/Exclusion Criteria	Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study: 1. Any evidence of active infection, including COVID-19, or any immunosuppressive disorder, including HIV infection. 3. ~~Prior~~Most recent, prior FibroScan or liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used. 7. Liver cirrhosis of any etiology as assessed by liver ultrasound/FibroScan. 16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including ~~possible~~ corticosteroid treatment and/or use of alternative immunosuppressive agents outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.	1, 6, 8

Section No./Title	Revision	Rationale
9.3.3/Removal of Subjects from Treatment or Assessment	Subjects may be considered lost to follow-up if the subject has missed 3 consecutive visits in the study and has failed to communicate a reason for this to the site. In addition, the site has documented at least 4 attempted contacts by key research personnel to reach the subject without success in the following manner: • 2 attempts by telephone or email (if possible); then • If telephone/email contacts are unsuccessful, 2 attempts must be made by certified letter or by appropriate local process. Where communication has been made by phone, this should be documented in the subject source notes.	15
9.4.4/Directions for Administration	In case of hypersensitivity reactions, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, ~~C5, C5a~~Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible.	2
9.4.8/Prior and Concomitant Medications	The following medications are prohibited starting 30 days before Screening and through the end of the study, and the Sponsor must be notified if a subject receives any of these during the study: • ~~Systemic immunosuppressive agents, except for corticosteroids~~ • ~~Lamivudine~~ The following medications should be avoided, starting 30 days prior to and for at least 52 weeks after BMN 270 infusion and minimized throughout the remaining duration of the study. • Medications which may be hepatotoxic, including isotretinoin and dextroamphetamine/amphetamine Subjects should be counseled to avoid starting potentially hepatotoxic therapies and to inform the Investigator of any new medications prescribed by other physicians. Investigators should carefully consider both the mechanism of action and potential hepatotoxicity of any new medication prior to initiation. If a potentially concerning new medication is started, Investigators should closely monitor both FVIII activity and ALT levels (eg, weekly to every 2 weeks for the first month) in order to determine if any detrimental effects on the efficacy or safety of BMN 270 have occurred. If co-medications are required during the course of the study, where possible, please check the National Center for Biotechnology Information LiverTox website for potential hepatotoxicity issues prior to prescribing (NCBI, 2020).	8, 12, 20
9.4.8.2/Therapeutic Corticosteroid and/or Immunosuppressive	Refer to corticosteroid prescription guidelines for recommended monitoring for, and management of, potential side effects of corticosteroids, including guidance on medications that should be avoided during corticosteroid treatment.	8, 9, 20

Section No./Title	Revision	Rationale
Agent Treatment of Elevated Hepatic Transaminases	Following initiation or completion of the prophylactic corticosteroid regimen, if ALT levels become increased (eg, ≥~~>~~ ULN or ≥ ~~2x~~1.5x baseline value) and alternative etiologies have been ruled out, prompt institution of therapeutic or on-demand oral corticosteroids (prednisone or converted equivalent) should be considered after consultation with the Medical Monitor (refer to Table 9.7.6.3.2). • Alternative immunosuppressive agents may also be considered for use on a case-by-case basis and following consultation with the Medical Monitor (eg, if prolonged corticosteroid use is contraindicated). ... Following initiation or completion of therapeutic oral corticosteroids, if increased ALT levels (eg, ≥~~>~~ ULN or ≥ ~~2x~~1.5x baseline value) are reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor... Management and monitoring of reactions to corticosteroids should be determined by the Investigator's clinical judgment in consultation with the Sponsor's Medical Monitor. This includes the contraindicated use of NSAIDs during corticosteroid treatment and specific monitoring not already covered by the SoA. The use of COX-2 inhibitors, while not contraindicated during corticosteroid treatment, should be limited, if possible. Practical management to prevent complications related to oral corticosteroid therapy may be undertaken at the discretion of the Investigator (eg, evaluation of glucose intolerance, hyperlipidemia ~~etc.).~~). Alternative, non-steroidal systemic immunosuppressive agents may be used, following a discussion between the Investigator and the Medical Monitor, should corticosteroid use be deemed by an Investigator to be clinically ineffective, not tolerated, and/or contraindicated. Hepatitis B status and HCV viral load will be rechecked 6 weeks after the start of oral corticosteroid/immunosuppressive agent treatment and then 1 week and 13 weeks after the completion of oral corticosteroid/immunosuppressive agent treatment in subjects with a history of hepatitis B or hepatitis C. All adverse events (including any adverse events suspected to be caused by or related to corticosteroid/immunousuppressive agent use) should be reported as outlined in Section 10 of the protocol. Subjects on corticosteroids should receive appropriate counselling and support regarding side effects from the Investigator or the treating institution (eg, listings of side effects and when to notify carers, wallet card for emergencies if on steroids, etc.). Additional management, including the co prescription of additional medications to prevent complications related to corticosteroid therapy, may be undertaken at the discretion of the investigator, including, but not limited to, prophylaxis against the occurrence of gastric ulcers, osteoporosis, and infections. The above guidance should also be followed in the event that an alternative immunosuppressive agent is used, as applicable.	
9.6/Dietary or Other Protocol Restrictions	Alcohol should be avoided starting 30 days prior to and for at least the first 52 weeks of the study, and particularly within 48 hours prior to lab work. Alcohol use should be minimized throughout the remaining duration of the study.	20

Section No./Title	Revision	Rationale
9.7.2.1/FVIII Activity	Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as either failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 or inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 52, as applicable and at the discretion of the Investigator. Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.	10
9.7.5/Exploratory Assessments	A cytokine bead array assay assessment will be performed at Baseline and then ~~weekly through Week 26~~at the timepoints listed in the Schedule of Activities.	20
9.7.6.2/Clinical Laboratory Assessments	In addition to scheduled clinical laboratory assessments, a fasting blood lipid panel (including triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) and fasting FibroTest will be assessed at the BMN 270 infusion visit. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit. In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, ~~C5, C5a~~Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction.	2, 6
Table 9.7.6.2.1	The ABO blood typing assessment has been listed as an "other test" rather than as part of the Hematology assessment panel.	20
9.7.6.3/Liver and Hepatitis Testing	A liver ultrasound/FibroScan and liver tests (LTs) during Screening will identify any significant hepatic dysfunction. Where a biopsy has been taken for safety-related reasons or was available from a past procedure, the Sponsor may request the biopsy information to help evaluate the impact of BMN 270 on the liver. The Sponsor may request that slides from a liver biopsy be made available for additional histopathological review... Elevated ALT levels should be evaluated according to the following plan~~:~~ (note that these evaluations may indicate additional testing of LTs and FVIII levels at unscheduled visits; these unscheduled laboratory tests may be completed by a mobile nursing professional at sites where the use of MN services has been approved):	5, 6, 13
Table 9.7.6.3.2	For ALT ≥3x ULN: • In the event that ALT or AST is ≥3x ULN and total bilirubin is ≥2x ULN, albumin and PT/INR should also be obtained.	4

Section No./Title	Revision	Rationale
9.7.6.5/Vital Signs, Physical Examinations, and Other Observations Related to Safety	Height will be recorded at Screening only. Weight will be recorded at Screening and then every 4 weeks thereafter through Week 20, at Week 26, ~~every 4 weeks through~~Week 32, Week 36, Week 44, and Week 52, and then at the second Q12W visit each year and at every End of ~~Years~~Year visit during Years 2-5.	14, 20
9.7.6.6/Vector Shedding	Vector shedding will also be extensively studied in the present clinical trial, at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5. Testing will continue until at least 3 consecutive results below the limit of detection are obtained. If a positive result is obtained in a matrix after 3 consecutive results below the limit of detection have already been recorded, testing in that matrix should restart and continue until an additional 3 consecutive results below the limit of detection have been obtained in order to confirm clearance. Testing of semen will continue at least through Week 12, even if 3 consecutive results below the limit of detection have been recorded in that compartment prior to that time point. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 26 should continue to have PCR testing in semen ~~every 4 weeks~~at the timepoints designated in the SoA until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor). Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule (refer to Section 12.5.3) but who have not cleared vector shedding from ~~all fluids~~a matrix must still provide samples for assessment until vector shedding has cleared: at Weeks 32, 36, 40, 44, 48, and 52 (during Year 1) and every 4~~6~~ weeks (for semen samples) or every 12 weeks (for all uncleared matrices) during Years 2-5. Such subjects may provide samples on the designated study visit dates either at the sites or through use of a MN professional.	7, 18, 20
10.2.1/Events of Special Interest (EOSI)	The following EOSI need to be reported to the Sponsor within 24 hours of site awareness, irrespective of seriousness, severity or causality: • Elevation of ALT ~~≥ 1.5x~~> ULN or ~~ALT > ULN & >2x~~≥1.5x baseline value, regardless of whether that elevation triggers an initiation or modification of oral corticosteroid treatment • Events potentially meeting the criteria for Hy's law (ALT or AST elevation ≥ 3x ULN plus total bilirubin ≥ 2x ULN) • Development of anti-FVIII inhibitory antibodies (inhibitors)	4, 9, 20
10.3.3/Assessment of Seriousness, Severity, and Causality	The Investigator responsible for the care of the subject or medically qualified designee will assess AEs for severity, relationship to study drug and/or corticosteroids and/or other immunosuppressive agents, and seriousness (refer to Section 10.2 for SAE definitions)...	

Section No./Title	Revision	Rationale
10.3.3.3/Causality	The Investigator will determine the relationship of an AE to the study drug and/or corticosteroids and/or other immunosuppressive agents and will record it on the source documents and AE eCRF. To ensure consistency of causality assessments, Investigators should apply the guidance in Table 10.3.3.3.1. Factors suggestive of a causal relationship could include (but are not limited to): • Absence of event prior to study drug and/or corticosteroid and/or other immunosuppressive agent exposure • Known relationship to underlying mechanism of study drug and/or corticosteroid and/or other immunosuppressive agent action • Abatement of AE with discontinuation of study drug and/or corticosteroids and/or other immunosuppressive agents, and/or recurrence of AE with reintroduction of study drug and/or corticosteroids and/or other immunosuppressive agents	11
Table 10.3.3.3.1	Table 10.3.3.3.1 has been updated to include assessment of causality against alternative immunosuppressive agents	11
~~10.4.1.2/Adverse Events Occurring Secondary to Other Events~~	~~In general, AEs occurring secondary to other events (eg, cascade events) should be identified by their primary cause. For example, if severe diarrhea is known to have resulted in dehydration, it is sufficient to record only diarrhea as an AE or SAE on the AE eCRF. However, medically important events that may be linked and/or separated in time should be recorded as independent events on the AE eCRF. For example, if severe hemorrhage leads to renal failure, both events should be recorded separately on the AE eCRF.~~	20
10.4.1.4/Abnormal Laboratory Values	Any laboratory result abnormality of CTCAE Grade 4 or 5 should be recorded as an SAE in the AE eCRF unless associated with an AE that has already been reported.	20
10.9/BPV Contact Information	The Investigator is encouraged to discuss with the medical monitor any AEs for which the issue of seriousness is unclear or questioned. Contact information for the medical monitor is as follows: Name: PI ~~MD PhD~~ PI MA MB BChir MSc Address: BioMarin ~~Pharmaceutical Inc.~~ (UK) Ltd. ~~105 Digital Drive~~ 10 Bloomsbury Way ~~Novato, CA 94949~~ London WC1A 2SL Phone: PI (office) PI (mobile) E-mail: PI	19

Section No./Title	Revision	Rationale
12.2/Screening Visit(s)	During the first part of the Screening Period (Day -42 to Day -29), testing for AAV5 TAb titers using the ARUP screening assay may be performed, so that subjects can verify their AAV5 TAB status. Subjects who agree to participate in this activity will be asked to sign a separate ICF documenting this decision. Subjects who do not agree will have the ARUP AAV5 TAb screening assay performed along with other assessments during the regular Screening period. The following procedures will be performed during the Screening Period~~:~~ (Day -28 to Day -1): • Liver Ultrasound/FibroScan • Blood sample for ARUP AAV5 TAb assay o Subjects who underwent the ARUP AAV5 TAb assay test earlier in the Screening period with the standalone consent do not need to repeat the test as part of regular Screening. • Screen for COVID-19 (local or central testing) o COVID-19 RT-PCR testing is required during Screening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing.	1, 6, 14
12.2.1/"Smart Rescreening" Visit	If a patient has to be screened again because the original assessments have fallen out of the 28 + 14 day period allowed for Screening (refer to Section 12.2), then only the following assessments need to be performed (rather than the full list indicated in Section 12.2) for the patient to be successfully re-screened for the study: • Screen for COVID-19 (local or central testing) o COVID-19 RT-PCR testing is required during smart rescreening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing.	1
12.4/Treatment Visit (Day 1)	The following procedures will be performed during the BMN 270 Infusion Visit: • Fasting FibroTest • Complement panel o Complement panel should include C3, C3a, C4, Bb, and sC5b-9 and should be collected within 2 hours post-BMN 270 infusion. In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4,	2, 3, 6

Section No./Title	Revision	Rationale
	~~C5, C5a~~Bb, sC5b-9, and cytokine bead array, as well as possible additional exploratory testing) and samples for IgE and cytokine bead array (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible.	
12.5.1.1/Week 1, Study Day 2	On Study Day 2, the following assessments will be performed: • Complement panel o Complement panel should include C3, C3a, C4, Bb, and sC5b-9.	3
12.5.1.2/Week 1, Study Day 4	On Study Day 4, the following assessments will be performed: • Complement panel o Complement panel should include C3, C3a, C4, Bb, and sC5b-9.	3
12.5.1.3/Week 1, Study Day 8	On Study Day 8, the following assessments will be performed: • Complement panel o Complement panel should include C3, C3a, C4, Bb, and sC5b-9.	3
12.5.2.1/Once per Week (Weeks 2 through 26)	The following procedures will be performed once per week from Weeks 1 through 26: • Liver Tests (refer to Table 9.7.6.3.1) o LTs may be monitored more or less frequently (and in particular when ALT values are > ULN or ≥ 1.5x ~~ULN~~baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.	9
12.5.2.4/Weeks 2, 4, 6, 8, 10, and 12	The following procedure will be performed at Weeks 2, 4, 6, 8, 10, and 12: • Complement panel o Complement panel should include C3, C3a, C4, Bb, and sC5b-9.	3
12.5.2.8/Weeks 2, 6, 10, 14, 20, 24, and 26	The following procedure will be performed at Weeks 2, 6, 10, 14, 20, ~~22~~24, and 26: • Cytokine bead array assay	14
12.5.3.1/Every Visit (Weeks 27-52)	At every visit (Weeks 27-36, 38, 40, 42, 44, 46, 48, 50, and 52), the following procedures will be performed: • Liver Tests (refer to Table 9.7.6.3.1)	9

Section No./Title	Revision	Rationale
	○ LTs may be monitored more or less frequently (and in particular when ALT values are ~~≥ 1.5x~~≥ ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.	
12.5.3.2/Weeks 28, 30, 32, 34, 36, 40, 44, 48, and 52	At Weeks 28, 30, 32, 34, 36, 40, 44, 48, and 52, the following procedures will be performed: • AAV5 TAb Assay • AAV5 TI Assay	14
12.5.3.5/Weeks 32, 36, 40, 44, 48, and 52	At Weeks 32, 36, 40, 44, 48, and 52, the following procedures will be performed: • ~~AAV5 TAb Assay~~ • ~~AAV5 TI Assay~~	14
12.6/Post-Infusion Follow-Up – Years 2-5	Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from ~~all fluids~~semen must still provide semen samples for assessment every 6 weeks during Years 2-5 until vector shedding has cleared (but do not have to perform other scheduled assessments at those study visits). Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.	10, 18
12.6.1/Years 2-5 – Every 6 Weeks (not required for treatment failure)	During Years 2-5, every 6 weeks (± 2 weeks), the following procedures will be performed: • Liver Tests (refer to Table 9.7.6.3.1) ○ LTs may be monitored more or less frequently (and in particular when ALT values are ~~≥ 1.5x~~≥ ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. • PCR of vector DNA in ~~blood, saliva, urine,~~ semen~~, and stools~~ (if required) ○ Sample testing during Years 2-5 is not required if at least 3 consecutive samples are clear by the end of Year ~~2~~1. Subjects who have not had 3 consecutive semen samples below the limit of detection by the end of Year 1 should continue to have PCR testing of semen every 6 weeks during Years 2-5 until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).	9, 18, 20

Section No./Title	Revision	Rationale
	○ Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from ~~all fluids~~semen must still provide semen samples for assessment every 6 weeks during Years 2-5 until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints).	
12.6.2/Years 2-5 – Every 12 Weeks and End of Year Visits (required for all subjects)	At the every 12 week and End of Year visits, the following procedures will be performed: • Liver Tests (refer to Table 9.7.6.3.1) ○ LTs may be monitored more or less frequently (and in particular when ALT values are ~~≥ 1.5x~~≥ ULN or ~~> ULN & > 2x~~≥ 1.5x baseline value) based on discussion between the Medical Monitor and the Investigator and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN. • PCR of vector DNA in blood, saliva, urine, semen, and stools (if required) ○ Sample testing during Years 2-5 is not required if at least 3 consecutive samples are below the limit of detection during the Post-Infusion Follow-Up period in Weeks 1-52. ~~Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 52 should continue to have PCR testing of semen every 6 weeks during Years 2-5 until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).~~	9, 20
12.7/Early Termination Visit	At the Early Termination visit, as many of the following assessments as possible should be done: • Cytokine bead array assay	14
16/Costs, Compensation, and Subject Injury	~~There will be no charge to study subjects to be in this study.~~ BioMarin will pay ~~all~~the full costs of the study-related tests, procedures, and treatments ~~that are part of this study~~set forth in the protocol. In addition, after IRB/EC approval, BioMarin may reimburse the reasonable cost of travel for study-related visits in accordance with BioMarin's travel and reimbursement policy. ~~BioMarin will not pay for any hospitalizations, tests, or treatments for medical problems of any sort related solely to the study subject's disease. Costs associated with such hospitalizations, tests, and treatments should be billed and collected in the way that such costs are usually billed and collected outside the study.~~ The Investigator should contact BioMarin immediately upon notification that a study subject has been injured by the study drug or by procedures performed as part of the study. Any subject who experiences a study-related injury should be instructed by the Investigator to seek immediate medical treatment at a pre-specified medical institution if possible, or at the closest medical treatment facility if necessary. The subject should be given the name of a person to contact to seek further information about, and assistance with, treatment for study-related injuries. ~~The treating physician should bill the subject's health insurance~~	16

Section No./Title	Revision	Rationale
	~~company or other third party payer for the cost of this medical treatment. If the cost of the medical treatment is not covered by health insurance or another third party that usually pays these costs, then either~~ BioMarin or the institution may pay for reasonable and necessary medical services to treat the injuries caused by the study drug or study procedures. In some jurisdictions, BioMarin is obligated by law to pay for study-related injuries~~ without prior recourse to third party payer billing and/or regardless of fault.~~ If this is the case, BioMarin will comply with the law. BioMarin will not pay for any hospitalizations, tests, or treatments for medical problems of any sort related solely to the study subject's primary disease or any concurrent disease that are unrelated to this study.	
21/References	~~Angus B, Brook G, Awosusi F, Barker G et al. British HIV Association guidelines for the routine investigation and monitoring of adult HIV-1-positive individuals. 2016. Available at http://www.bhiva.org/documents/Guidelines/Monitoring/2016-BHIVA-Monitoring-Guidelines.pdf. Last accessed 12 September 2017.~~ National Center for Biotechnology Information (NCBI). LiverTox: Clinical and Research Information on Drug-Induced Liver Injury. Available at: https://livertox.nih.gov (last accessed 20 August 2020).	20

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CLINICAL STUDY PROTOCOL

Study Title:	A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5
Protocol Number:	270-203
Active Investigational Product:	AAV5-hFVIII-SQ
European Union Drug Regulating Authorities Clinical Trials (EudraCT) Number:	2017-000662-29
Indication:	Hemophilia A
Sponsor:	BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949
Development Phase:	Phase 1/2
Sponsor's Responsible Medical Monitor:	PI , MD PI , Clinical Sciences BioMarin (UK) Ltd. 10 Bloomsbury Way London, WC1A 2SL
Study Design:	Single-arm, open-label
Duration of Subject Participation:	Up to 264 weeks
Dose:	6E13 vg/kg as single infusion
Study Population:	Males ≥ 18 years of age with severe hemophilia A and detectable pre-existing antibodies against AAV5 vector capsid
Date of Original Protocol:	29 September 2017
Date of Amendment 1:	5 October 2018
Date of Amendment 2:	4 October 2019
Date of Amendment 3:	24 August 2020
Date of Amendment 4:	4 August 2021

CLINICAL STUDY PROTOCOL AMENDMENT SUMMARY

Amendment 4

Date: 4 August 2021

RATIONALE AND SUMMARY OF CHANGES

A summary of major changes covered by Amendment 4 to the 270-203 protocol is provided below:

1. Changes have been made to enhance screening for potential malignancies (including hepatic cancers) and establishing baseline liver health during the Screening Period.

Rationale: The changes made include:

- Clarifying that all patients should undergo a liver ultrasound at Screening to screen for clinically significant liver disease and hepatocellular carcinoma (HCC), and that a FibroScan can also be performed at the discretion of the Investigator;
- Moving the fasting FibroTest from Day 1 to Screening for assessment of liver fibrosis;

As no current guidelines regarding HCC screening exist for AAV gene therapy recipients, the Sponsor engaged experts regarding current and recommended screening for HCC prior to administration of gene therapy. Protocol amendments contained herein were made based upon expert opinion, literature review, and evaluation of best practices.

2. Changes have been made to enhance screening for potential malignancies (including hepatic cancers) after dosing with BMN 270.

Rationale: The changes made include:

- Including a targeted liver ultrasound at the End of Year visits for Year 1 through Year 5 to screen for HCC (additional interim liver ultrasounds may be performed at the discretion of the Investigator);
- Recommending genomic analyses for any malignancy (except non-melanoma skin cancer) diagnosed during the course of the study.

Year-end liver ultrasounds are being implemented to assess the theoretical risk of HCC. While no cases of HCC have been reported in the Sponsor's AAV gene therapy non-clinical or clinical trials (more than 150 patients dosed, with some dosed more than 5 years ago), these assessments will further inform this theoretical risk.

3. Malignancy (except non-melanoma skin cancer) has been added as an Event of Special Interest (EOSI).

Rationale: The occurrence of malignancy (as above) was added as an EOSI for purposes of expedited safety reporting and additional safety monitoring.

4. HIV-positive patients (serological evidence of HIV-1 or HIV-2 infection) may now enroll in the study, provided their HIV infection is stable and well-controlled with an adequate CD4 count (>200/mm^3) and an undetectable viral load, respectively, at Screening and they are on an antiretroviral therapy (ART) regimen that does not contain efavirenz or another potentially hepatotoxic ART.

Rationale: HIV-positive subjects were initially included in prior BMN 270 studies. However, after an HIV-positive subject in 270-302 developed markedly elevated liver enzyme levels after receiving 4E13 vg/kg of BMN 270, out of an abundance of caution for the long-term liver health of HIV-positive patients, further enrollment of HIV-positive subjects was suspended in 270-301 (Protocol Amendment 3) and 270-302 (Protocol Amendment 3). The subject in 270-302 referenced above was receiving efavirenz and lamivudine as part of his ART regimen. Following discussion with a liver advisory board and review of the accumulated 270-301 data, efavirenz and not lamivudine has been implicated as the most likely medication that interacted with BMN 270 and contributed to the 270-302 subject's elevated liver enzyme levels. Due to its hepatotoxicity, efavirenz is considered a prohibited medication in all BMN 270 studies.

The two HIV-positive subjects on stable, non-efavirenz-containing ART regimens who were enrolled in and dosed in 270-301 study prior to Amendment 3 have been monitored closely. Following BMN 270 infusion, these subjects continued their ART as prescribed and followed routine monitoring of CD4 count and viral load. Results from 270-301 show similar safety results for the two HIV-positive subjects compared to those who are HIV-negative. The Sponsor believes that HIV infection, in and of itself, is not a contraindication to receive BMN 270 and has therefore removed the exclusion of HIV-positive subjects, as long as they are not receiving efavirenz or other potentially hepatotoxic ART in their HIV treatment regimen.

5. Language has been added concerning the use of the SARS-CoV-2 vaccines.

Rationale: Due to the current worldwide SARS-CoV-2 pandemic and evolving availability and types of vaccines, language has been added to assist with timing and planning for vaccine administration. The Sponsor's recommendations reflect the risk assessment conducted on the currently available vaccines and guidance from multiple health agencies, and include information regarding different types of SARS-CoV-2 vaccines.

6. The reactive corticosteroid regimen for ALT elevation has been updated.

Rationale: The guidance for the reactive corticosteroid regimen, including management of ALT elevations and corticosteroid taper, reflects the data gathered from previous BMN 270 studies. This change attempts to promote the safe and effective use of reactive corticosteroids.

7. Thrombin generation assay (TGA) assessment has been removed.

Rationale: Assessment of TGA in other studies in the BMN 270 development program has provided sufficient data; additional assessments are not expected to be needed. Removing this assessment helps to alleviate patient and site burden. If TGA assessments from 270-203 are needed in the future, backup aliquots from other platelet-poor plasma samples can be utilized.

8. The definition of treatment failure has changed.

Rationale: The previous definition required either a failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 or an inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes; the revised definition requires both of these conditions to be present before a subject may be considered a treatment failure. The revision reflects the data seen as of the 52-week data cut for 270-301, where some subjects were still able to remain off of prophylactic FVIII replacement therapy (due to an absence of treated bleeding events) despite lower FVIII levels 1 or more years after BMN 270 infusion.

9. Frequency of several laboratory assessments during Years 2-5 has been decreased. These changes include:

- Reducing FVIII Antigen BDD Assay to Q12W for Years 2-5

  Rationale: Robust characterization is already available from Year 1 data, and more frequent sampling is not needed to understand long-term protein to activity ratios.

- Reducing AAV5 TAb to End of Year Visits only for Years 3-5

  Rationale: Antibody response is high and does not change much over time. Yearly testing should be sufficient to understand longer term responses, as little change is expected.

- Reducing FVIII TAb to End of Year Visits only for Years 3-5

  Rationale: FVIII Bethesda Inhibitor is the main safety assessment. After 3 years, if there is a need in specific instances for data more frequent than once a year, backup plasma from another assay may be used for testing.

- Reducing AAV5 TI assay to End of Year visits only for Years 2-5

  Rationale: Yearly testing is considered to be sufficient to understand longer term responses, as little change is expected.

- Reducing PBMC collection to every other Q12W visit during Years 3-5

  Rationale: Cellular immune responses appear to taper off after the first year. After Year 2, sampling every 6 months should be sufficient to monitor late or more durable responses.

10. The identity of the Medical Monitor has been updated.
11. The option for a legally authorized representative to provide informed consent where needed has been added.
12. Additional minor changes have been made for consistency and clarity.

Refer to Section 25 for a summary of revisions to Amendment 3 (dated 24 August 2020).

2 SYNOPSIS

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	**SUMMARY TABLE** Referring to Part of the Dossier: Volume: Page: Reference:	**FOR NATIONAL AUTHORITY USE ONLY:**
TITLE OF STUDY: A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5		
PROTOCOL NUMBER: 270-203		
STUDY SITES: Approximately 5-14 sites globally		
PHASE OF DEVELOPMENT: Phase 1/2		
STUDY RATIONALE: Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. The clinical phenotype of HA patients generally correlates tightly with the level of residual expression. Severe HA is classified as FVIII activity less than 1% of wild-type (< 1 IU/dL), moderate disease comprises 1-5% of wild-type activity, and the mild form is 5-40% activity. The clinical manifestations of severe HA are frequent spontaneous bleeding episodes, predominantly in joints and soft tissues, with a substantially increased risk of death from hemorrhage when the brain is involved. Treatment of severe HA presently mainly consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with episodic (on-demand-only) therapy in the same studies, respectively. Continued repeated bleeding events compound debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death.		

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

Extended half-life products created through chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) or bioengineering of FVIII (eg, FVIII-Fc fusion proteins) can only extend half-life by approximately 50% (half-life 18-19 hours), and thus, show promise in reduced dosing and maintaining activity levels above a 1% trough for a greater proportion of the dosing interval. However, patients with severe HA who are treated with extended half-life FVIII remain dependent on multiple infusions to maintain critical levels of FVIII activity. More recently, emicizumab has been approved for the treatment of HA. Emicizumab is a humanized, bispecific monoclonal antibody that binds to both activated coagulation factor IX (FIX) and coagulation factor X (FX), thereby mimicking the function of activated FVIII. Emicizumab represents an incremental improvement in therapeutic burden over replacement FVIII products since it can be administered subcutaneously as opposed to intravenously; however, it is still a chronic treatment that requires repeat administration every 1-4 weeks, and sustained FVIII activity levels above 5-10 IU/dL have rarely been attained even with extended half-life products. Therefore, there is a strong unmet need for a treatment that can alter the fundamental problem for patients with HA, namely attainment of sustained, hemostatically effective FVIII activity levels long-term (ie, over multiple years) without the need for treatment.

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of active FVIII following a single intravenous infusion of a vector encoding the appropriate gene sequence. Hemophilia A is well-suited for a gene replacement approach because clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in minute amounts (200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and even modest increases in the level of FVIII (any increase of the plasma level by 2 ng/ml induces an increase in activity of 1%) can ameliorate the severe form of hemophilia A. Thus, relatively small changes in endogenous FVIII activity can result in clinically relevant improvements in disease phenotype. Moreover, the circulating FVIII response to gene transduction can be assessed using validated quantitative endpoints that are easily assayed using established laboratory techniques.

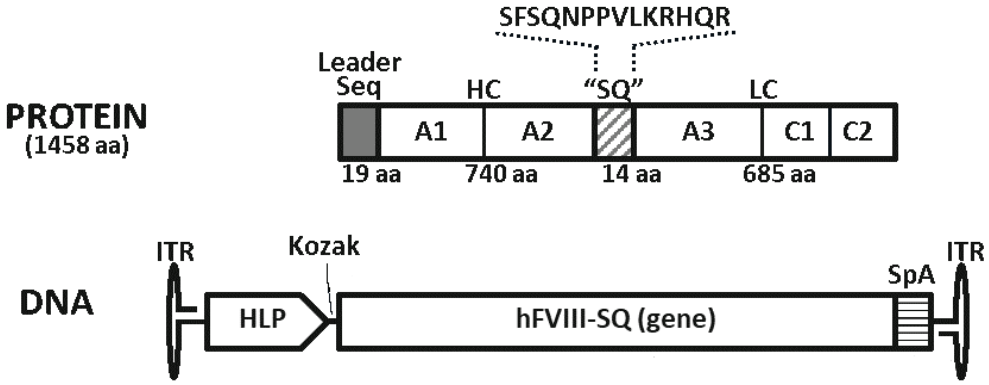
Several different gene transfer strategies for FVIII replacement have been evaluated, with adeno-associated viral (AAV) vectors used in most hemophilia clinical trials. AAV vectors have an excellent and well-defined safety profile, and can direct long-term transgene expression with tropism and promoter specificity for specific tissues, such as the liver (eg serotypes 2, 5, and 8). Indeed, an ongoing gene therapy clinical trial for a related disorder, hemophilia B, has established over 7 years of stable human factor IX (hFIX) expression at levels sufficient for conversion of their bleeding phenotype from severe to moderate or mild, following a single peripheral vein infusion of AAV8-hFIX vector. Several participants in this trial have been able to discontinue factor prophylaxis without suffering spontaneous hemorrhages, even when they undertook activities that

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

previously resulted in bleeding. Thus, gene therapy treatment has resulted in a substantial improvement in their quality of life.

BMN 270 is an AAV5-based gene therapy vector that expresses the SQ form of hFVIII under the control of a hybrid human liver-specific promoter (Figure 1).

Figure 1: AAV5-hFVIII-SQ Vector Genome and Encoded Protein



Legend –Note that schematic is not to scale; aa = amino acids; ITR = inverted terminal repeat; HLP = human liver promoter; Kozak = Kozak concensus sequence (GCCACC); SpA = Synthetic poly(A) signal

BMN 270 is delivered by a single IV dose and is designed to achieve stable, durable expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.

BMN 270 is being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. An additional study has been undertaken at the 6E13 vg/kg dose (270-301 in subjects with severe HA), as well as a study in subjects receiving the 4E13 vg/kg dose (270-302).

Four-year results from 270-201 and one-year results from 270-301 have demonstrated that following gene transfer, mean and median FVIII activity levels above 15% (15 IU/dL), as measured by a chromogenic substrate assay, are achievable and sustained following a single infusion of 6E13 vg/kg of BMN 270, with administration of prophylactic corticosteroids and an acceptable safety profile. Preliminary results from optional liver biopsies (in subjects receiving

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

lower doses of BMN 270 in 270-201) confirm dose-dependent pan-lobular and otherwise healthy liver transduction at 2.7-4.1 years.

Subjects receiving 6E13 vg/kg in 270-201 received a different corticosteroid regimen than subjects in 270-301; in 270-201, subjects were scheduled to start corticosteroids by Week 3 (either before Week 3, in response to an alanine aminotransferase (ALT) elevation, or at Week 3 otherwise, per protocol), whereas in 270-301 subjects received corticosteroids only in response to an ALT elevation. Possibly as a result of this difference, subjects receiving 6E13 vg/kg in 270-201 started corticosteroids at an earlier date in reference to the date of BMN 270 infusion and showed later onset of first ALT elevations when compared with subjects in 270-301. Recently published data from 270-201 and recent analysis of 270-301 data suggest that corticosteroids may have assisted in rescue or protection of FVIII activity levels during elevations of ALT and in resolution of elevated ALT levels in some subjects.

Subjects enrolled and infused in 270-201 screened negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. Subjects enrolled and infused in 270-301 also screened negative for AAV5 antibodies. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes.

Current data describing the extent of neutralization or inhibited transduction has been inconsistent across studies, each employing one of several varying methodologies to evaluate both vector-specific antibodies and transduction inhibition (TI). Early studies using AAV2 for the treatment of hemophilia B described inhibited transduction in patients with neutralizing antibody titers of 2 or 3, but more recent studies have described successful transduction in patients who, in retrospect, were determined to have pre-existing antibody to the vector capsid. Importantly, no drug-related adverse events were described in these patients. As the sensitivity of assays used to detect pre-existing antibody has increased greatly in recent years, the biological relevance of any given titer determination, and comparisons across studies using assays with differing titer sensitivities, requires additional study.

Data from a non-clinical study conducted by BioMarin in cynomolgus monkeys further demonstrated that transduction efficiency can vary greatly in the presence of pre-existing anti-AAV5 antibody. In non-human primates, the ability to attain FVIII levels in the face of measurable AAV5 antibody titers without significant safety risks supports the possibility of achieving a similar response in patients with AAV5 antibody positivity. The totality of available data shows the effect of pre-existing immunogenicity to AAV delivery vectors is not completely

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

understood, and indicates that some patients may benefit from AAV5 gene therapy who are currently excluded.

In this study, patients with severe HA and pre-existing antibodies against AAV5 will be enrolled to evaluate the clinical response to BMN 270 gene therapy. The aim is to evaluate the safety, tolerability, and efficacy of BMN 270 in patients with severe HA and pre-existing antibodies against AAV5 vector capsid at various levels of AAV5 antibody titers.

OBJECTIVES:

The primary objective of the study is to:

- Assess the safety of a single intravenous administration of BMN 270 in severe HA subjects with pre-existing antibody to AAV5 vector capsid, including development of FVIII neutralizing antibody

Secondary objectives of the study are to:

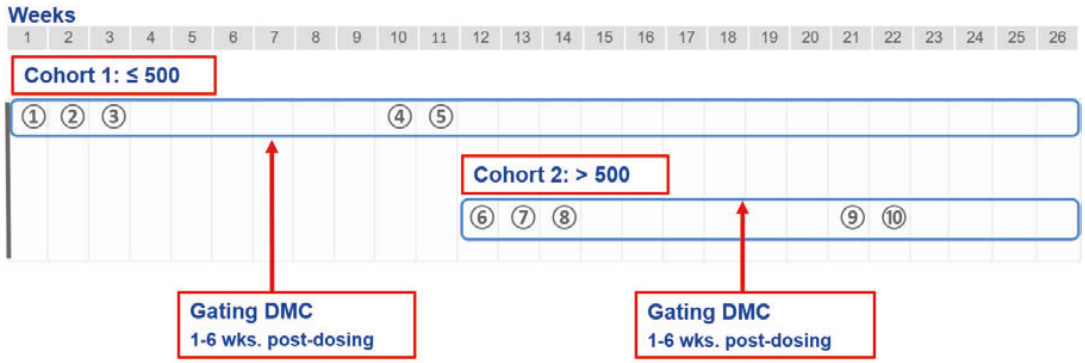
- Assess the efficacy of BMN 270 defined as FVIII activity at or above 5 IU/dL at Week 26
- Assess the impact of BMN 270 on usage of exogenous FVIII replacement therapy
- Assess the impact of BMN 270 on the number of bleeding episodes requiring exogenous FVIII therapy
- Evaluate the pharmacodynamics of FVIII expression following IV infusion of BMN 270
- Assess the impact of BMN 270 on patient-reported outcomes (PROs)

STUDY DESIGN AND PLAN:

This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to the AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects may be enrolled at 5-14 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb titer ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb titer > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date.

An independent Data Monitoring Committee (DMC) will consist of experts in clinical trials, statistics, and hemophilia. The DMC will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 2 represents one dosing schedule scenario):

Figure 2: 270-203 Dosing Schedule (One Possible Scenario)



① - ⑩ Enrollment sequence of subjects
Gating Rationale: Limit exposure to patients who may not benefit

Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from 1-6 weeks post-infusion) reviewed. FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response, based on data from 270-201. Between 1 and 6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness).

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC evaluation of safety and efficacy in treated subjects, with the following triggers that may potentially pause further enrollment:

- any related SAE;
- any related AE with a severity > CTCAE Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

Following a temporary halt of enrolment, the DMC may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed. Additional blood samples will be collected (tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine profiling, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine profiling (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine profiling. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.

Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DMC based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL. In subjects who experience recurring bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis.

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 and inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor or Sponsor-designated Data Monitor, follow an abbreviated visit schedule after Week 52 by attending only the Q12W and End of Year visits during Years 2-5. Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.

The study analysis will be performed after all subjects have been followed for 26 weeks post-BMN 270 infusion, with presentation of safety, efficacy, and FVIII PD assessments. After the safety, efficacy, and FVIII PD analyses at 26 weeks post-BMN 270 infusion, long-term safety and efficacy will be assessed in all subjects for up to a total of 5 years post-infusion. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the DMC.

NUMBER OF SUBJECTS PLANNED:

Approximately 10 subjects

DIAGNOSIS AND ALL CRITERIA FOR INCLUSION AND EXCLUSION:

Individuals eligible to participate in this study must meet all of the following criteria:

1. Males ≥ 18 years of age with hemophilia A and residual FVIII levels ≤ 1 IU/dL as evidenced by medical history, at the time of signing the informed consent.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

2. Detectable pre-existing antibodies against the AAV5 vector capsid as measured by AAV5 total antibody ELISA
3. Treated/exposed to FVIII concentrates or cryoprecipitate for a minimum of 150 exposure days (EDs)
4. Subject must have been on prophylactic FVIII replacement therapy for at least 12 months prior to study entry.
5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures. If the subject is unable to provide consent, a legally authorized representative may provide written informed consent.
6. No previous documented history of a detectable FVIII inhibitor, and results from a Bethesda assay or Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) (or less than 1.0 BU for laboratories with a historical lower sensitivity cutoff for inhibitor detection of 1.0 BU) on 2 consecutive occasions at least one week apart within the past 12 months (at least one of which should be tested at the central laboratory)
7. Sexually active participants must agree to use an acceptable method of effective contraception, either double-barrier contraception (ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with viral vector DNA below the limit of detection.
8. Willing to abstain from consumption of alcohol for at least the first 52 weeks following BMN 270 infusion.

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Any evidence of active infection, including COVID-19, or any immunosuppressive disorder, except for HIV infection. HIV-positive patients who meet all other eligibility criteria may be included if they have a CD4 count > 200/mm^3 and an undetectable viral load (unquantifiable viral load as defined as less than the limit of quantification by the testing laboratory's assay is permitted) while receiving an antiretroviral therapy (ART) regimen that does not contain efavirenz or another potentially hepatotoxic ART.
2. Significant liver dysfunction with any of the following abnormal laboratory results:
    - ALT (alanine aminotransferase) > 1.25x ULN;

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

- AST (aspartate aminotransferase) > 1.25x ULN;
- GGT (gamma-glutamyltransferase) > 1.25x ULN;
- Total bilirubin > 1.25x ULN;
- Alkaline phosphatase > 1.25x ULN; or
- INR (international normalized ratio) ≥ 1.4

Subjects whose liver laboratory assessments fall outside of these ranges may undergo repeat testing of the entire liver test panel within the same Screening window and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor.

3. Most recent, prior FibroScan or liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig or METAVIR scoring systems, or an equivalent grade of fibrosis if an alternative scale is used.
4. Evidence of any bleeding disorder not related to hemophilia A
5. Platelet count of < 100 x 10^9/L
6. Creatinine ≥ 1.5 mg/dL
7. Liver cirrhosis or other clinically significant liver disease of any etiology as assessed by liver ultrasound/FibroScan.
8. Chronic or active hepatitis B as evidenced by positive serology testing (hepatitis B surface antigen [HBsAg], hepatitis B surface antibody [HBsAb], and hepatitis B core antibody [HBcAb]) and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results.
9. Active Hepatitis C as evidenced by detectable HCV RNA, or currently on antiviral therapy
10. Active malignancy, except non-melanoma skin cancer
11. History of hepatic malignancy
12. History of arterial or venous thromboembolic events (eg, deep vein thrombosis, non-hemorrhagic stroke, pulmonary embolism, myocardial infarction, arterial embolus), with the exception of catheter-associated thrombosis for which anti-thrombotic treatment is not currently ongoing.
13. Known inherited or acquired thrombophilia, including conditions associated with increased thromboembolic risk, such as atrial fibrillation.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	**SUMMARY TABLE** Referring to Part of the Dossier: Volume: Page: Reference:	**FOR NATIONAL AUTHORITY USE ONLY:**

14. A history of known inflammatory, connective tissue, or autoimmune disorders (eg, vasculitis).
15. Treatment with any Investigational Product within 30 days or 5 half-lives of the investigational product (whichever is longer) prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study
16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including corticosteroid treatment and/or the use of alternative immunosuppressive agents outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.
17. Prior treatment with any vector or gene transfer agent
18. Major surgery planned in the 52-week period following the infusion with BMN 270
19. Use of systemic immunosuppressive agents, not including corticosteroids, or live vaccines within 30 days before the BMN 270 infusion
20. Concurrent enrollment in another clinical study, unless it is an observational (non-interventional) clinical study that does not interfere with the requirements of the current protocol or have the potential to impact the evaluation of safety and efficacy of BMN 270 and with prior consultation with the Medical Monitor
21. Known allergy or hypersensitivity to investigational product formulation
22. Unwilling to receive blood or blood products for treatment of an adverse event and/or a bleed

INVESTIGATIONAL PRODUCT(S), DOSE, ROUTE AND REGIMEN:
Each subject will receive a single IV infusion of BMN 270 at 6E13 vg/kg. The volume of infusion will depend on the subject's weight.

REFERENCE THERAPY(IES), DOSE, ROUTE AND REGIMEN:
No reference therapy will be evaluated in this study.

DURATION OF TREATMENT:
BMN 270 is given as a single dose by IV infusion.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

CRITERIA FOR EVALUATION:

Safety:

The following safety outcome measurements will be assessed:

- Incidence of adverse events (AEs), including serious AEs (SAEs)
- Change in clinical laboratory tests (serum chemistry and hematology)
- Change in vital signs
- Change in physical examination
- Vector shedding (blood, urine, semen, feces, saliva)
- Liver tests (LTs, including ALT, AST, GGT, LDH, total bilirubin, and alkaline phosphatase)
- Immune response to FVIII transgene product and AAV5 vector capsid

No major toxicity is expected based on 270-201 data and non-clinical studies. Each subject will have comprehensive surveillance monitoring of LTs (once per week for Weeks 1-26). During the long-term safety evaluation, LTs will be monitored every three months for up to 5 years post-infusion; the frequency and duration of LT testing may be changed based on discussion between the Medical Monitor and the Investigator and review of subject data.

There will be a detailed assessment of cellular and humoral responses to AAV5 vector capsid and FVIII.

Efficacy:

The efficacy measure will be to assess plasma FVIII activity. The efficacy goal is to achieve FVIII activity ≥ 5 IU/dL at 26 weeks post-BMN 270 administration. Other efficacy measures include assessing the impact of BMN 270 on the use of FVIII replacement therapy and on the number of bleeding episodes during the study. Subjects will be asked to keep a subject diary, provided by the sponsor, to record the relevant details.

Other efficacy endpoints:

- Change from baseline in the total score of HAEMO-QoL-A at Week 26 of the study post-BMN 270 infusion
- Change from baseline in the EQ-5D-5L score at Week 26 of the study post-BMN 270 infusion.
- Change from baseline in the Haemophilia Activities List (HAL) score at Week 26 of the study post-BMN 270 infusion.

NAME OF COMPANY BioMarin Pharmaceutical Inc. 105 Digital Drive Novato, CA 94949 NAME OF FINISHED PRODUCT: BMN 270 NAME OF ACTIVE INGREDIENT: AAV5-hFVIII-SQ	SUMMARY TABLE Referring to Part of the Dossier: Volume: Page: Reference:	FOR NATIONAL AUTHORITY USE ONLY:

- Change from baseline in the Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) score at Week 26 of the study post-BMN 270 infusion.

Pharmacodynamics:

The FVIII antigen and activity level, as measured by a validated immunoassay and a validated FVIII activity assay, respectively, will be used for plasma profiles; FVIII antigen and activity will be used to determine PD parameters.

STATISTICAL METHODS:

Approximately 10 subjects may be dosed in the study. The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using longitudinal methods, such as a mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at ≥ 5 IU/dL at 26 weeks following study treatment. Assessment of the true steady state of FVIII will require that FVIII activity is measured after a minimum of 72 hours (or 5x the known half-life of the FVIII replacement therapy administered) has elapsed since the last infusion of FVIII concentrates.

Analysis of neutralizing antibody response, other immunological parameters, and vector shedding will be primarily descriptive and involve both inter-subject and intra-subject comparisons across cohorts.

3 TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

4 LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviations

AAV	adeno-associated virus
ABR	annualized bleeding rate
ADL	activities of daily living
ADR	adverse drug reaction
AE	adverse event
ALT	alanine aminotransferase
APTT	activated partial thromboplastin time
ART	anti-retroviral therapy
AST	aspartate aminotransferase
BPV	BioMarin Pharmacovigilance
BU	Bethesda Unit
CFR	Code of Federal Regulations
CRA	clinical research associate
CRF	case report form
CSR	clinical study report
CTCAE	Common Terminology Criteria for Adverse Events
DMC	Data Monitoring Committee
EC	ethics committee
eCRF	electronic case report form
ED	exposure days
EOSI	events of special interest
ETV	early termination visit
EudraCT	European Union Drug Regulating Authorities Clinical Trials
FDA	Food and Drug Administration
FIH	first-in-human
FVIII	coagulation factor VIII
GCP	Good Clinical Practice
GGT	gamma-glutamyltransferase
HA	Hemophilia A
HAL	Haemophilia Activities List
HBcAb	hepatitis B core antibody
HBsAb	hepatitis B surface antibody
HBsAg	hepatitis B surface antigen

hFIX	human coagulation factor IX
hFVIII	human coagulation factor VIII
HIPAA	Health Insurance Portability and Accountability Act
IB	investigator brochure
ICF	informed consent form
ICH	International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
ICH E6 [R2]	ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6
IND	Investigational New Drug (application)
INR	international normalized ratio
IP	investigational product
IRB	institutional review board
IV	intravenous
LT	liver test
MedDRA	Medical Dictionary for Regulatory Activities
MN	mobile nursing
PBMC	peripheral blood mononuclear cells
PCR	polymerase chain reaction
PD	pharmacodynamics
PEG	polyethylene glycol
PK	Pharmacokinetics
PRO	patient-reported outcome
rhFVIII	recombinant human FVIII protein
SAE	serious adverse event
SAP	statistical analysis plan
SDV	source data verification
SoA	schedule(s) of activities
ULN	upper limit of normal
vg	vector genomes
VWF:Ag	von Willebrand factor Antigen
WPAI+CIQ:HS	Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific

Definition of Terms:

Investigational Product (IP):

"A pharmaceutical form of an active ingredient or placebo being tested or used as a reference in a clinical trial, including a product with a marketing authorization when used or assembled (formulated or packaged) in a way different from the approved form, or when used for an unapproved indication, or when used to gain further information about an approved use" (from International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6 [ICH E6] (R2)]).

The terms "IP" and "study drug" may be used interchangeably in the protocol.

5 ETHICS

BioMarin Pharmaceutical Inc. (hereafter referred to as BioMarin or the Sponsor) conducts its studies according to the highest ethical and scientific standards. The following Sections articulate standards to which Investigators will be held accountable, as well as matters of compliance to document adherence to such standards.

5.1 Institutional Review Board or Independent Ethics Committee

Investigators are expected to interact with Ethics Committees (ECs) promptly, as required, during the course of the study. This includes, but is not limited to, providing appropriate documentation to support study initiation and maintaining appropriate flow of safety and other information during the course of the study and for study close-out activities. BioMarin (or designee) will assist Investigators with access to timely and accurate information and with assurance of prompt resolution of any queries.

Prior to initiating the study, the Investigator will obtain written confirmation that the institutional review board (IRB) or independent ethics committee (EC) is properly constituted and compliant with International Conference on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use (ICH) and Good Clinical Practice (GCP) requirements, applicable laws, and local regulations. A copy of the confirmation from the IRB/EC will be provided to BioMarin Pharmaceutical Inc. (BioMarin) or its designee. The Investigator will provide the IRB/EC with all appropriate material, including the protocol, Investigator's Brochure (IB), the Informed Consent Form (ICF) including compensation procedures, and any other written information provided to the subjects, including all ICFs translated for patients who do not speak the local language at the clinical site. The study will not be initiated and Investigational Product (IP) supplies will not be shipped to the site until appropriate documents from the IRB/EC confirming unconditional approval of the protocol, the ICF and all subject recruitment materials are obtained in writing by the Investigator and copies are received at BioMarin or its designee. The approval document should refer to the study by protocol title and BioMarin protocol number (if possible), identify the documents reviewed, and include the date of the review and approval. BioMarin will ensure that the appropriate reports on the progress of the study are made to the IRB/EC and BioMarin by the Investigator in accordance with applicable guidance documents and governmental regulations.

5.2 Ethical Conduct of Study

It is expected that Investigators understand and comply with the protocol. This includes, but is not limited to: establishing and meeting enrollment commitments, including providing

eligible subjects for study enrollment; adhering to adverse event reporting, diagnostic, or other procedures as specified in the protocol; and assuring appropriate compliance with study treatment administration and accountability.

This study will be conducted in accordance with the following:

- European Clinical Trial Directive 2001/20/EC and Good Clinical Practice Directive 2005/28/EC, for studies conducted within any European country
- US Code of Federal Regulations (CFR) Sections that address clinical research studies, and/or other national and local regulations, as applicable
- ICH Harmonised Tripartite Guideline: Guideline for Good Clinical Practice E6(R2) (ICH E6R2)
- The ethical principles established by the Declaration of Helsinki

Specifically, this study is based on adequately performed laboratory and animal experimentation. The study will be conducted under a protocol reviewed and approved by an IRB/EC and will be conducted by scientifically and medically qualified persons. The benefits of the study are in proportion to the risks. The rights and welfare of the subjects will be respected and the investigators conducting the study do not find the hazards to outweigh the potential benefits. Each subject (or his legally authorized representative, if required) will provide written, informed consent before any study-related tests or evaluations are performed.

5.3 Subject Information and Informed Consent

A properly written and executed informed consent form (ICF), in compliance with ICH E6R2 (Section 4.8), United States Code of Federal Regulations (CFR) 21 CFR §50, European Clinical Trial Directive 2001/20/EC and Good Clinical Practice Directive 2005/28/EC, and other applicable local regulations, will be obtained for each subject prior to entering the subject into the study. The Investigator will prepare the ICF and provide the documents to BioMarin for approval prior to submission to the IRB/EC approval. BioMarin and the IRB/EC must approve the documents before they are implemented. A copy of the approved ICF and if applicable, a copy of the approved subject information sheet and all ICFs translated to a language other than the native language of the clinical site must also be received by BioMarin or designee prior to any study-specific procedures being performed.

6 INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

During administration of informed consent, expectations regarding participation in the study should be made clear to subjects (and their legally authorized representatives, if required). Subjects who are not willing and/or are not able to comply with all aspects of the study should not be encouraged to participate.

Prior to beginning the study, the Investigator at each site must provide to BioMarin or designee a fully executed and signed Statement of Investigator (SOI) form. A US Food and Drug Administration (FDA) Form FDA 1572 serves as an acceptable SOI form. If Form FDA 1572 may not be used in a particular region, the Investigator must provide a fully executed SOI on the form provided by the Sponsor. All Investigators and Sub-Investigators must be listed on Form FDA 1572 or its equivalent SOI. Financial Disclosure Forms must also be completed for all Investigators and Sub-Investigators listed on the Form FDA 1572 or SOI who will be directly involved in the treatment or evaluation of subjects in this study.

The study will be administered by and monitored by employees or representatives of BioMarin. Clinical Research Associates (CRAs) or trained designees will monitor each site on a periodic basis and perform verification of source documentation for each subject as well as other required review processes. BioMarin's Regulatory Affairs Department (or designee) will be responsible for the timely reporting of serious adverse events (SAEs) to appropriate regulatory authorities as required.

In multicenter studies, a Coordinating Investigator will be identified who will be responsible for study overview. The Coordinating Investigator will read the clinical study report (CSR) and confirm that it accurately describes the conduct and results of the study, to the best of his or her knowledge. The Coordinating Investigator will be chosen on the basis of active participation in the study, ability to interpret data, and willingness to review and sign the report in a specified timeframe. The identity of the Coordinating Investigator and a list of all Investigators participating in the study will be provided in the CSR.

Clinical Laboratory assessments will be performed at a nominated central laboratory. Bioanalytical samples will be sent to the appropriate specialty laboratories for testing. Refer to laboratory manual for more details.

7 INTRODUCTION

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males (Iorio 2019). It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. Clinical manifestations of severe FVIII deficiency are frequent unprovoked bleeding episodes in joints and soft tissues causing permanent disability and occasionally death mostly after brain hemorrhage. Treatment in Western countries (Berntorp 2012) consists of intravenous injection of plasma-derived or recombinant FVIII protein concentrates at the time of a bleed to control it or prophylactically to prevent bleeding episodes. The short half-life for FVIII (12-18 hours) necessitates frequent infusions and makes this treatment prohibitively expensive for the majority of the world's hemophilia A patients. These individuals develop debilitating arthropathy and have a substantially increased risk of death from hemorrhage in life (Stonebraker 2010). Chemical modification or bioengineering of FVIII may improve half-life to 18-19 hours (Kaufman 2013). However, these extended half-life FVIII variants do not eliminate the need for lifelong FVIII protein administration (Hay 2012).

Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of human FVIII (hFVIII) following a single administration of vector. Hemophilia A is well-suited for this approach because its clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in low amounts (100-200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and a modest increase in the level of FVIII (a plasma level of 2 ng/ml protein leads to a 1% expression) can ameliorate the severe phenotype (Srivastava 2020); thus, the therapeutic goal for gene therapy is a modest increase in hFVIII. Finally, the consequences of gene transfer can be assessed using validated quantitative endpoints that can be easily assayed in most clinical laboratories.

BMN 270 contains the cDNA for the B-domain-deleted SQ FVIII with a liver-specific HLP transcription promoter. The expression cassette is inserted between AAV2 ITRs, and this genome is packaged in the AAV5 capsid. A comprehensive review of BMN 270 is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.1 Nonclinical Studies

The nonclinical program supports a single IV infusion of BMN 270, the planned clinical route of administration, for the treatment of hemophilia A in male patients. This nonclinical program took into account the guidelines and reflection papers for gene therapy medicinal products under EMA Advanced Therapies as well as FDA guidance. The primary pharmacodynamics (PD), pharmacokinetics (PK), and toxicity of IV BMN 270 were characterized in a series of single dose studies in species that were vector permissive and responsive to the transgene including normal CD-1 mice, a B- and T-cell deficient mouse model of hemophilia A (B6;129S-*F8*^{tm1Kaz}/J x B6.129S6-*Rag2*^{tm1Fwa} N12; FVIII KO x Rag2), and normal cynomolgus and rhesus monkeys. Some PD studies evaluated additional PK, immunogenicity and toxicity endpoints.

The comparative pharmacodynamics of BMN 270 in cynomolgus monkeys with varying pre-existing AAV5 transduction inhibition (TI) titer and AAV5 TAb status was evaluated in study BMN270-16-021. BMN 270 was administered to 4 groups of monkeys, a control group (Group 1, n=3) that tested negative for both TI and AAV5 TAb, Group 2 (n=4) that was AAV5 TAb negative, and low TI titer (2-5) positive. Group 3 (n=4) was also AAV5 TAb negative, but had higher TI titers (5-10). Group 4 (n=5) tested positive for both AAV5 TAb and TI (TI titers were >5). Administration of BMN 270 by a single intravenous bolus injection was well-tolerated in cynomolgus monkeys regardless of baseline TI titer or TAb status. After dosing, all monkeys showed FVIII-SQ levels above the LLOQ, with the exception of two monkeys in the group that presented with both positive TI and TAb titers at baseline. Though these TAb+ monkeys, regardless of TI titers, showed a significant mean reduction in FVIII expression (68% less) compared to TAb negative monkeys, three of five monkeys showed detectable levels of FVIII-SQ, with one having levels similar to that observed in the TI and TAb negative control group. Monkeys that were TI+ but TAb-at baseline had FVIII expression levels that were similar to those of the TI and TAb negative control group.

Results of the nonclinical program to date are detailed in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.2 Ongoing Clinical Studies

Ongoing clinical studies for BMN 270 include:

- 270-201, a phase 1/2, dose-escalation study in patients with severe HA
- 270-205, a phase 1/2 study in patients with severe HA who have active or prior FVIII inhibitors

- 270-301, a phase 3 study in patients with severe HA who receive BMN 270 at the 6E13 vg/kg dose level
- 270-302, a phase 3 study in patients with severe HA who receive BMN 270 at the 4E13 vg/kg dose level
- 270-303, a phase 3 study in patients with severe HA who received BMN 270 at the 6E13 vg/kg dose level along with prophylactic corticosteroids

A comprehensive review of safety, efficacy, and immunogenicity results as of the latest data cut is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.

7.3 Study Rationale

Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males. It is caused by deficiency in the activity of coagulation factor VIII (FVIII), an essential cofactor in the intrinsic coagulation pathway. This disorder can be either inherited, due to a genetic aberrancy, or an acquired immunologic process, leading to insufficient quantities of FVIII or a dysfunctional FVIII, but all are characterized by a defective coagulation process. The clinical phenotype of HA patients generally correlates tightly with the level of residual expression. Severe HA is classified as FVIII activity less than 1% of wild-type (< 1 IU/dL), moderate disease comprises 1-5% of wild-type activity, and the mild form is 5-40% activity. The clinical manifestations of severe HA are frequent spontaneous bleeding episodes, predominantly in joints and soft tissues, with a substantially increased risk of death from hemorrhage when the brain is involved.

Treatment of severe HA presently mainly consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study (Berntorp 2017) and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with episodic (on-demand-only) therapy in the same studies, respectively. Continued repeated bleeding events compound debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death.

Extended half-life products created through chemical modification (eg, direct conjugation of polyethylene glycol (PEG) polymers) or bioengineering of FVIII (eg, FVIII-Fc fusion

proteins) can only extend half-life by approximately 50% (half-life 18-19 hours), and thus, show promise in reduced dosing and maintaining activity levels above a 1% trough for a greater proportion of the dosing interval. However, patients with severe HA who are treated with extended half-life FVIII remain dependent on multiple infusions to maintain critical levels of FVIII activity. More recently, emicizumab has been approved for the treatment of HA. Emicizumab is a humanized, bispecific monoclonal antibody that binds to both activated coagulation factor IX (FIX) and coagulation factor X (FX), thereby mimicking the function of activated FVIII. Emicizumab represents an incremental improvement in therapeutic burden over replacement FVIII products since it can be administered subcutaneously as opposed to intravenously; however, it is still a chronic treatment that requires repeat administration every 1-4 weeks, and sustained FVIII activity levels above 5-10 IU/dL have rarely been attained even with extended half-life products. Therefore, there is a strong unmet need for a treatment that can alter the fundamental problem for patients with HA, namely attainment of sustained, hemostatically effective FVIII activity levels long-term (ie, over multiple years) without the need for treatment.

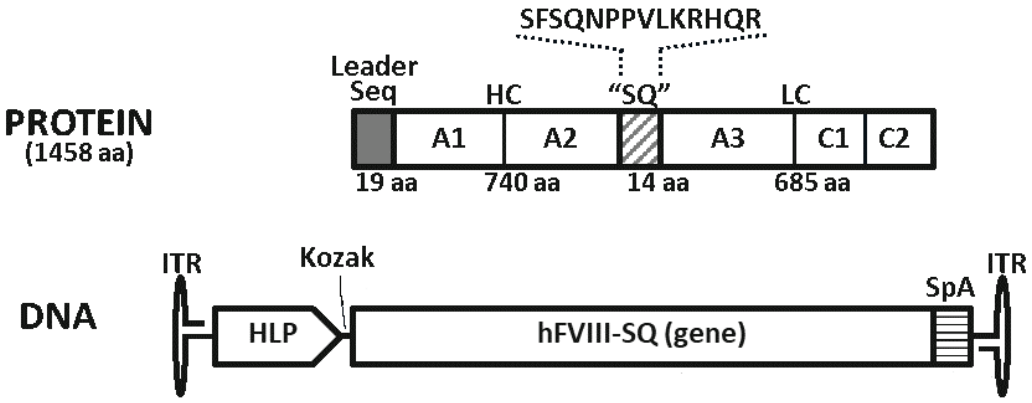
Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of active FVIII following a single intravenous infusion of a vector encoding the appropriate gene sequence. Hemophilia A is well-suited for a gene replacement approach because clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in minute amounts (200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and even modest increases in the level of FVIII (any increase of the plasma level by 2 ng/ml induces an increase in activity of 1%) can ameliorate the severe form of hemophilia A. Thus, relatively small changes in endogenous FVIII activity can result in clinically relevant improvements in disease phenotype. Moreover, the circulating FVIII response to gene transduction can be assessed using validated quantitative endpoints that are easily assayed using established laboratory techniques.

Several different gene transfer strategies for FVIII replacement have been evaluated, with adeno-associated viral (AAV) vectors used in most hemophilia clinical trials. AAV vectors have an excellent and well-defined safety profile, and can direct long-term transgene expression with tropism and promoter specificity for specific tissues, such as the liver (eg serotypes 2, 5, and 8). Indeed, an ongoing gene therapy clinical trial for a related disorder, hemophilia B, has established over 7 years of stable human factor IX (hFIX) expression at levels sufficient for conversion of their bleeding phenotype from severe to moderate or mild, following a single peripheral vein infusion of AAV8-hFIX vector (Nathwani 2018). Several participants in this trial have been able to discontinue factor prophylaxis without suffering spontaneous hemorrhages, even when they undertook activities

that previously resulted in bleeding. Thus, gene therapy treatment has resulted in a substantial improvement in their quality of life.

BMN 270 is an AAV5-based gene therapy vector that expresses the SQ form of hFVIII under the control of a hybrid human liver-specific promoter (Figure 7.3.1).

Figure 7.3.1: AAV5-hFVIII-SQ Vector Genome and Encoded Protein



Legend –Note that schematic is not to scale; aa = amino acids; ITR = inverted terminal repeat; HLP = human liver promoter; Kozak = Kozak concensus sequence (GCCACC); SpA = Synthetic poly(A) signal

BMN 270 is delivered by a single IV dose and is designed to achieve stable, durable expression of active hFVIII in the plasma, synthesized from vector-transduced liver tissue.

BMN 270 is being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. An additional study has been undertaken at the 6E13 vg/kg dose (270-301 in subjects with severe HA), as well as a study in subjects receiving the 4E13 vg/kg dose (270-302).

Four-year results from 270-201 and one-year results from 270-301 have demonstrated that following gene transfer, mean and median FVIII activity above 15% (15 IU/dL), as measured by a chromogenic substrate assay, are achievable and sustained following a single infusion of 6E13 vg/kg of BMN 270, with administration of prophylactic corticosteroids and an acceptable safety profile. Preliminary results from optional liver biopsies (in subjects

receiving lower doses of BMN 270 in 270-201) confirm dose-dependent pan-lobular and otherwise healthy liver transduction at 2.7-4.1 years.

Subjects receiving 6E13 vg/kg in 270-201 received a different corticosteroid regimen than subjects in 270-301; in 270-201, subjects were scheduled to start corticosteroids by Week 3 (either before Week 3, in response to an alanine aminotransferase (ALT) elevation, or at Week 3 otherwise, per protocol), whereas in 270-301 subjects received corticosteroids only in response to an ALT elevation. Possibly as a result of this difference, subjects receiving 6E13 vg/kg in 270-201 started corticosteroids at an earlier date in reference to the date of BMN 270 infusion and showed later onset of first ALT elevations when compared with subjects in 270-301. Recently published data from 270-201 and recent analysis of 270-301 data suggest that corticosteroids may have assisted in rescue or protection of FVIII activity levels during elevations of ALT and in resolution of elevated ALT levels in some subjects.

Subjects enrolled and infused in 270-201 were screened and negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. Subjects enrolled and infused in 270-301 also screened negative for AAV5 antibodies. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes (Hayes 2019; Boutin 2010).

Current data describing the extent of neutralization or inhibited transduction has been inconsistent across studies, each employing one of several varying methodologies to evaluate both vector-specific antibodies and transduction inhibition (TI). Early studies using AAV2 for the treatment of hemophilia B described inhibited transduction in patients with neutralizing antibody titers of 2 or 3 (Manno 2006), but more recent studies have described successful transduction in patients who, in retrospect, were determined to have pre-existing antibody to the vector capsid (Majowicz 2017). Importantly, no drug-related adverse events were described in these patients. As the sensitivity of assays used to detect pre-existing antibody has increased greatly in recent years, the biological relevance of any given titer determination, and comparisons across studies using assays with differing titer sensitivities, requires additional study.

Data from a non-clinical study conducted by BioMarin in cynomolgus monkeys further demonstrated that transduction efficiency can vary greatly in the presence of pre-existing anti-AAV5 antibody. In non-human primates, the ability to attain FVIII levels in the face of

measurable AAV5 antibody titers without significant safety risks supports the possibility of achieving a similar response in patients with AAV5 antibody positivity. The totality of available data shows the effect of pre-existing immunogenicity to AVV delivery vectors is not completely understood, and indicates that some patients may benefit from AAV5 gene therapy who are currently excluded.

In this study, patients with severe HA and pre-existing antibodies against AAV5 will be enrolled to evaluate the clinical response to BMN 270 gene therapy. The aim is to evaluate the safety, tolerability, and efficacy of BMN 270 in patients with severe HA and pre-existing antibodies against AAV5 vector capsid at various levels of AAV5 antibody titers.

7.4 Summary of Overall Risks and Benefits

Overall, 151 subjects have received a BMN 270 infusion at one of 4 dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, or 6E13 vg/kg) in one of the four ongoing BMN 270 clinical studies (270-201, 270-301, 270-302, 270-203). Single infusions have been generally well-tolerated across all investigated doses. All subjects have successfully completed their full-dose infusion of BMN 270, with no discontinuation of dosing due to adverse events observed during the infusion. No deaths have been reported in any of the BMN 270 studies, and no participants have discontinued from studies as a result of an adverse event.

Transient ALT elevation (Grade 1 to 3 in severity) has been observed in most subjects administered BMN 270 shortly after dosing, with no evidence for major impacts upon liver function; no events meeting the Hy's Law criteria have been identified. Liver function has remained stable over time. Across the 6E13 vg/kg cohort of 270-201 and 270-301, subjects enrolled in 270-201 developed ALT elevation about 5.5 weeks later than subjects in 270-301, generally once the first course of corticosteroids was being tapered, and experienced lower peak elevations in ALT (75.7 U/L) than subjects in 270-301 (112.5 U/L). The difference in the ALT profile seen between the 6E13 vg/kg subjects in 270-201 and the subjects in 270-301 could be attributed to the difference in the protocol-specified corticosteroid regimens in place in those studies, including the early use of corticosteroids (ie, by Week 3 post-BMN 270 infusion). While the majority of ALT elevations responded rapidly to corticosteroids, given current interest in the field of AAV gene therapy for the use of non-steroidal approaches to managing or preventing ALT elevations, alternate non-steroidal systemic immunosuppressive agents have also been used to manage hepatic reactions where corticosteroids have proven to be ineffective or where high doses/and or prolonged exposure to corticosteroids have led to unwanted side effects. Overall, the literature and clinical experience with BMN 270 suggests that transient elevations in liver enzymes are expected following AAV-based gene therapy for the treatment for hemophilia A or B without any

long-term concerns of hepatic injury (Manno 2006; Nathwani 2011; George 2016; Miesbach 2016; Pasi 2020).

Short-lived infusion reactions associated with one-time BMN 270 administration have included symptoms such as nausea, maculopapular rash, urticaria, diarrhea, watery eyes, rigors, chills, myalgia, fever, tachycardia and hypotension emerging within 24 hours of receiving BMN 270. Most infusion-related reactions were Grade 1 or Grade 2 in severity, and all events resolved, typically within 48 hours following medical management. While some cases required temporary interruption of the infusion, followed by re-initiation at a slower rate, all subjects completed their infusions. The reactions with onset during or within approximately 5 hours after the end of infusion responded to treatment with systemic antihistamines and/or corticosteroids, where administered. Infusion-related reactions were effectively mitigated by managing infusion rate and medications.

No subjects have experienced thromboembolic events or developed inhibitors to FVIII following BMN 270 infusion.

Subjects given the 6E13 vg/kg dose in 270-201 and 270-301 have achieved mean FVIII activity above 40 IU/dL at 49-52 weeks post-infusion, with markedly decreased bleeding compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. Subjects at all dose levels continue to be followed.

The current data available for BMN 270 has shown an established positive benefit:risk profile for BMN 270 at the 6E13 vg/kg dosing level, although impact of prophylactic corticosteroids requires further investigation. Given the monitoring measures in place in the clinical protocol(s) to minimize the risk to subjects participating in the existing studies, the identified risks are justified by the anticipated benefits that may be afforded to subjects. Each subject in 270-203 will have a comprehensive surveillance plan that monitors LTs during the study, and elevations in LT will be addressed according to the guidelines set forth in the protocol. Safety will be assessed by adverse event reporting and clinical laboratory assessments.

For additional information on findings from other BMN 270 clinical studies, refer to the current version of the IB.

8 STUDY OBJECTIVES

The primary objective of the study is to:

- Assess the safety of a single intravenous administration of BMN 270 in severe HA subjects with pre-existing antibody to AAV5 vector capsid, including development of FVIII neutralizing antibody

The secondary objectives of the study are to:

- Assess the efficacy of BMN 270 defined as FVIII activity at or above 5 IU/dL at Week 26
- Assess the impact of BMN 270 on usage of exogenous FVIII replacement therapy
- Assess the impact of BMN 270 on the number of bleeding episodes requiring exogenous FVIII therapy
- Evaluate the pharmacodynamics of FVIII expression following IV infusion of BMN 270
- Assess the impact of BMN 270 on patient-reported outcomes (PROs)

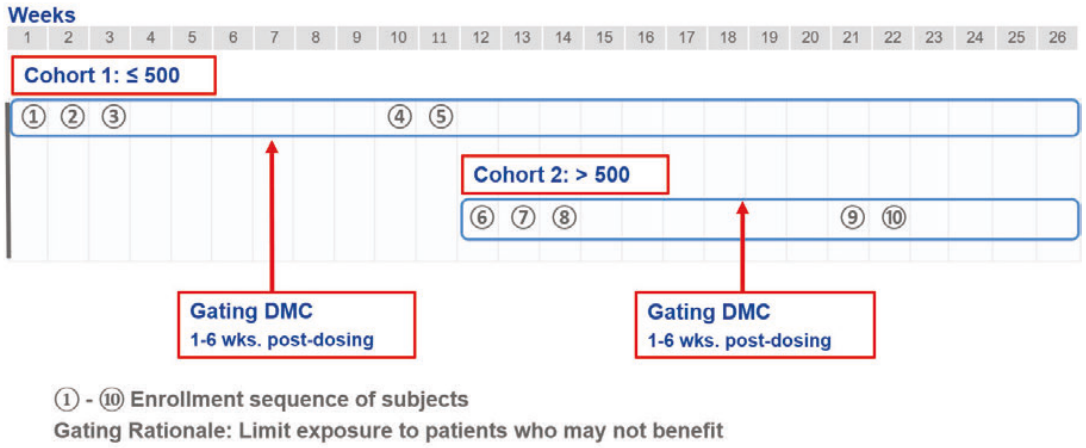
9 INVESTIGATIONAL PLAN

9.1 Overall Study Design and Plan

This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects may be enrolled at 5-14 sites globally in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date.

An independent Data Monitoring Committee (DMC), consisting of experts in clinical trials, statistics, and hemophilia, has been convened. The DMC will review available safety and efficacy data throughout the study and provide recommendations based on their review (Figure 9.1.1 represents one dosing schedule scenario):

Figure 9.1.1: 270-203 Dosing Schedule (One Scenario)



Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from 1-6 weeks post-infusion) reviewed. Based on data from 270-201, FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response. Between 1 and 6 weeks

post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness).

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC evaluation of safety and efficacy in treated subjects with the following triggers that may potentially pause further enrollment:

- any related SAE;
- any related AE with a severity > CTCAE Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects at 6 weeks post-BMN 270 infusion.

Following a temporary halt of enrolment, the DMC may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed. Additional blood samples will be collected (tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine profiling, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine profiling (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine profiling. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.

Data from 270-201 suggest that achievement of therapeutic FVIII levels ≥ 5 IU/dL occurs approximately 4 weeks after BMN 270 infusion, although the FVIIII PD in AAV+ subjects is unknown and requires close monitoring. As such, in order to provide adequate FVIII protection while subjects are projected to reach clinically relevant FVIII levels ≥ 5 IU/dL, prior FVIII prophylaxis for each subject will be continued at the discretion of the DMC based on individual subject status and data review or when FVIII activity has reached at least 5 IU/dL.

As the relationship between activity assay results of the BMN 270 gene product and bleeding remains to be established, Investigators should strive to minimize bias by avoiding consideration of FVIII activity levels by themselves or subjects in the reporting of bleeding episodes and FVIII usage.

In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis. Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 and inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor or Sponsor-designated Data Monitor, follow an abbreviated visit schedule after Week 52 by attending only the Q12W and End of Year visits during Years 2-5. Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.

The study analysis will be performed after all subjects have been followed for 26 weeks post-BMN 270 infusion, with presentation of safety, efficacy, and FVIII PD assessments. After the safety, efficacy, and FVIII PD analyses at 26 weeks post-BMN 270 infusion, long-term safety and efficacy will be assessed in all subjects for up to a total of 5 years post-infusion. During the trial, additional subjects may be recruited into each cohort at any time, if deemed necessary by the DMC.

A summary of all assessments is provided in the Schedule of Activities (SoA) in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5.

Table 9.1.1: Schedule of Activities – Screening/Baseline/Day 1

Assessment	Prior to BMN 270 Infusion				BMN 270 Infusion Visit (Day 1)[o]
	Screening		Smart Rescreening[m] (Day -28 to Day -1)	Baseline (Day -7 to Day -1)[n]	
	Screening (Day -42 to Day -29)	Screening (Day -28 to Day -1)			
Informed consent	X	X			
Demographics (age, sex, race, ethnicity)		X			
Medical History (including hemophilia A history, Hepatitis B, Hepatitis C, and HIV)		X			
Physical Examination[a]		X		X	X
Height and Weight		X			
Vital Signs		X	X	X	X
Assessment of Adverse Events and Concomitant Medications		X	X	X	X
Documentation of bleeding episodes and FVIII usage for previous 12 months (by either subject or clinical information)		X	X	X	
Distribution of subject diaries and training in their use[b]		X			
Electrocardiogram		X			
Liver Ultrasound (and FibroScan at discretion of Investigator)[c]		X			
hFVIII Assays[d]		X	X[p]	X	
AAV5 TAb Assay (ARUP)[e]	X	X[e]	X		X
AAV5 TAb Assays[f]				X	
AAV5 TI Assay[f]				X	X
Screen for Hepatitis B, Hepatitis C, HIV[g]		X			
SARS-CoV-2 screening (local or central)[h]		X	X		
Blood chemistry, hematology, and coagulation tests[i]		X	X	X	
Blood fasting lipid panel					X
Fasting FibroTest		X			
Urine Tests[i]		X	X	X	
Liver Tests[i]		X	X	X	

Assessment	Prior to BMN 270 Infusion				BMN 270 Infusion Visit (Day 1)[o]
	Screening		Smart Rescreening[m] (Day -28 to Day -1)	Baseline (Day -7 to Day -1)[n]	
	Screening (Day -42 to Day -29)	Screening (Day -28 to Day -1)			
PBMC collection (for baseline determination of AAV5 and FVIII specific cellular immunity)				X	
Von Willebrand Factor Antigen (VWF:Ag)				X	
PCR of vector DNA in blood, saliva, urine, semen, and stools				X	X[o]
Biomarker testing[j]		X			
Exploratory biomarker assessments[k]				X	X
Cytokine profiling				X	
Haemo-QOL-A assessment				X	
EQ-5D-5L assessment				X	
HAL assessment				X	
WPAI+CIQ:HS assessment				X	
BMN 270 Infusion					X
Complement panel and exploratory cytokine profiling[l]					X[l]
Hypersensitivity blood assessments				X	(X)[q]

[a] Complete physical examination should be done at Screening. Brief physical examination may be done at Baseline and at the BMN 270 Infusion Visit.

[b] Diaries should be distributed to subjects who have consented to participate in the study and who have been determined to meet all study eligibility criteria.

[c] All patients must have a liver ultrasound performed during the Screening period to screen for significant liver disease and hepatocellular carcinoma. A FibroScan may also be performed at the discretion of the Investigator.

[d] Includes baseline FVIII activity (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), coagulation exploratory assay, hFVIII inhibitor level (Bethesda assay with Nijmegen modification), hFVIII total antibody titer, and hFVIII antigen assay. Baseline activity should be assessed at trough (at least >72 hours after last dose of replacement FVIII therapy, or 5x the known half-life of the FVIII concentrates administered).

[e] Screening, Smart Re-screening, and Infusion Day samples will be tested using the ARUP AAV5 TAb assay. During Screening, the ARUP AAV5 TAb assay test may be done first, under a standalone informed consent form, before the main ICF for the study is signed and further screening procedures are performed. Sample collection on the day of the infusion visit must be performed before the BMN 270 infusion is given. If the ARUP AAV5 TAb assay test is done first with the standalone consent, it does not need to be repeated as part of regular Screening.

[f] Baseline and all post-dose samples will be tested in a different AAV5 TAb post-dose immunogenicity monitoring assay.

[g] Subjects with documented negative results within the last 30 days do not need to be retested. Hepatitis B screening should include hepatitis B surface antigen (HBsAg), hepatitis B surface antibody (HBsAb), and hepatitis B core antibody (HBcAb).

[h] SARS-CoV-2 RT-PCR testing is required during Screening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing. A SARS-CoV-2 vaccine is recommended for subjects if indicated and if available. If a two-step SARS-CoV-2 vaccine is being used, sites should consider using the flexible re-screen option to allow subjects to receive both doses at least 14 days prior to treatment with BMN 270 (or at least 30 days prior to treatment with BMN 270 for any live-virus vaccines). It is preferable for SARS-CoV-2 vaccination to occur prior to BMN 270 infusion. Investigators should use clinical judgment, taking into consideration local factors, individual risk factors, and benefit/risk related to timing of vaccine administration.

[i] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.4.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. ABO blood typing assessment should be performed at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study.

[j] Includes HLA genotyping and FVIII genotyping.

[k] Blood samples will be collected from subjects to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for those evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for any exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[l] Complement panel should include C3, C3a, C4, Bb, and sC5b-9 and should be collected within 2 hours post-infusion. While exploratory samples for cytokine profiling will be collected at the time points indicated above, testing of these samples will be performed only as deemed necessary by the Sponsor.

[m] Smart rescreening should only be performed if a subject has been determined to be eligible for the study and is unable to complete the Baseline assessments and Infusion prior to the closing of the original Screening window (28 + 14 days). SARS-CoV-2 RT-PCR testing is required as part of smart rescreening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing. Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once.

[n] Should the screening visit occur within 30 days of the drug infusion, physical examination, blood chemistry, LTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline.

[o] With the exception of the collection of samples for PCR vector DNA analysis and the collection of the complement panel/exploratory cytokine profiling sample, assessments on the day of infusion must be performed prior to the infusion. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of

the infusion visit. On the day of the BMN 270 Infusion, vital signs will be monitored prior to the infusion, during the infusion every 15 minutes (± 5 minutes), and following the infusion hourly (± 5 minutes) for at least 8 hours during the subject's stay in the clinic. Shedding samples for PCR of vector DNA analysis (blood, saliva, urine, semen, stool) should be collected between 2 and 24 hours after the infusion has been completed.

[p] Only the hFVIII inhibitor level (Bethesda assay with Nijmegen modification) assay must be done at smart rescreening.

[q] In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination, vital signs will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine profiling, as well as possible additional exploratory testing) and samples for IgE and cytokine profiling (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine profiling. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.

Table 9.1.2: Schedule of Activities – Post-Infusion Follow-Up (Week 1-16)

Assessment	Follow-Up After BMN 270 Infusion – Weeks*																	
	Week 1																	
	D2	D4	D8	2	3	4	5[g]	6	7[g]	8	9[g]	10	11[g]	12	13[g]	14	15[g]	16
Study Day*	2	4	8	15	22	29	36	43	50	57	64	71	78	85	92	99	106	113
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight						X				X				X				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X		X		X		X						X		
Urine Tests[b]														X				
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII antibody titer				X		X		X		X		X		X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]	X[d]	X	X	X	X	X		X		X				X				X
Exploratory biomarker assessments[e]				X				X				X				X		
Haemo-QOL-A assessment														X				
EQ-5D-5L assessment														X				
HAL assessment														X				
WPAI+CIQ:HS assessment														X				
AAV5 TAb Assay	X		X	X		X		X		X		X		X		X		X
AAV5 TI Assay	X		X	X		X		X		X		X		X		X		X

Assessment	Follow-Up After BMN 270 Infusion – Weeks*																	
	Week 1																	
	D2	D4	D8	2	3	4	5[g]	6	7[g]	8	9[g]	10	11[g]	12	13[g]	14	15[g]	16
Study Day*	2	4	8	15	22	29	36	43	50	57	64	71	78	85	92	99	106	113
Testing for reactivation of hepatitis B and hepatitis C																		X[f]
PBMC collection (for determination of AAV5 and FVIII specific immunity)	X			X		X		X		X		X		X		X		X
Complement panel[h]	X	X	X	X		X		X		X		X		X				
VWF:Ag						X				X				X				X
Cytokine profiling	X		X	X				X				X				X		

* Visit windows are ± 24 hours during Week 1 and ± 48 hours starting with the Week 2 visit.

[a] Brief physical examination should be done at all weekly visits.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.4.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs assessment is weekly, but may be checked more frequently when ALT values are > ULN or ≥ 1.5x baseline value or based upon discussion between the Medical Monitor and the Investigator. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: the ALT has increased to above ULN; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen assay. Chromogenic Nijmegen-Bethesda Assay for hFVIII inhibitor level will be tested as deemed necessary by the Sponsor. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted

based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Collection for each matrix to occur until at least 3 consecutive results below the limit of detection are obtained. Collection and testing of semen samples must continue through Week 12, even if 3 consecutive results below the limit of detection in that compartment have already been recorded. Separate collection on Day 2 is not required if vector shedding samples were collected on Day 1 post-BMN 270 infusion; if collection on Day 2 is needed, it must occur within 24 hours of the time of the BMN 270 infusion.

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for any exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] Testing for reactivation of hepatitis B and hepatitis C at Week 16, for subjects with a past medical history of hepatitis B or hepatitis C prior to study entry, should be performed only in subjects who have not received prophylactic oral corticosteroids; subjects who have received prophylactic oral corticosteroids will have hepatitis B and hepatitis C testing at the time points indicated in Table 9.1.6.

[g] The scheduled visits at Week 5, Week 7, Week 9, Week 11, Week 13, and Week 15 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject [or his legally authorized representative, if required] has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits.

[h] Complement panel should include C3, C3a, C4, Bb, and sC5b-9. While exploratory samples for cytokine profiling will be collected at the time points indicated above, testing of these samples will be performed only as deemed necessary by the Sponsor.

Table 9.1.3: Schedule of Activities – Post-Infusion Follow-Up (Week 17-32)

Assessment	Follow-Up After BMN 270 Infusion – Weeks*															
	17[f]	18	19[f]	20	21[f]	22	23[f]	24	25[f]	26	27[f]	28	29[f]	30	31[f]	32
Study Day*	120	127	134	141	148	155	162	169	176	183	190	197	204	211	218	225
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight[a]				X						X						X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X						X						X
Urine Tests[b]										X						
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
FVIII antibody titer				X				X		X						X
PCR of vector DNA in blood, saliva, urine, semen, and stools[d]				X				X		X						X
Exploratory biomarker assessments[e]		X				X				X						X
Haemo-QOL-A assessment										X						
EQ-5D-5L										X						
HAL										X						
WPAI+CIQ:HS										X						
AAV5 TAb Assay		X		X		X		X		X		X		X		X
AAV5 TI Assay		X		X		X		X		X		X		X		X
PBMC collection (for determination of AAV5 and FVIII specific cellular immunity)		X		X		X		X		X		X		X		X
VWF:Ag				X						X						
Cytokine profiling[g]				X				X		X						X

* Visit windows are ± 48 hours.

[a] Brief physical examination should be done at all weekly visits except Week 26, where a complete physical examination should be performed. Weight should be recorded at Week 20, Week 26, and Week 32.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.4.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs assessment may be checked more frequently when ALT values are > ULN or ≥ 1.5x baseline value or based upon discussion between the Medical Monitor and the Investigator. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: the ALT has increased to above ULN;; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen assay. Chromogenic Nijmegen-Bethesda Assay for hFVIII inhibitor level will be tested as deemed necessary by the Sponsor. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Collection for each matrix to occur until at least 3 consecutive results below the limit of detection are obtained.

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for any exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] The scheduled visits at Week 17, Week 19, Week 21, Week 23, Week 25, Week 27, Week 29, and Week 31 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject [or his legally authorized representative, if required] has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For

MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits.

[g] While exploratory samples for cytokine profiling will be collected at the time points indicated above, testing of these samples will be performed only as deemed necessary by the Sponsor.

Table 9.1.4: Schedule of Activities – Post-Infusion Follow-Up (Week 33-52)

Assessment	Year 1 – Weeks*											
	33[e]	34	35[e]	36	38[e]	40	42[e]	44	46[e]	48	50[e]	52
Study Day[*]	232	239	246	253	267	281	295	309	323	337	351	365
Physical examination[a]	X	X	X	X	X	X	X	X	X	X	X	X
Weight[a]				X				X				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X	X	X	X	X	X	X	X	X	X
Vital Signs	X	X	X	X	X	X	X	X	X	X	X	X
Blood chemistry, hematology, and coagulation tests[b]				X				X				X
Urine Tests[b]				X								X
Liver Tests[b]	X	X	X	X	X	X	X	X	X	X	X	X
FVIII assays[c]	X	X	X	X	X	X	X	X	X	X	X	X
AAV5 TAb Assay		X		X		X		X		X		X
AAV5 TI Assay		X		X		X		X		X		X
FVIII antibody titer				X				X				X
Exploratory biomarker assessments[d]				X		X		X		X		X
PBMC Collection (for determination of FVIII and Capsid specific cellular immunity)		X		X				X				X
VWF:Ag				X								X
Cytokine profiling[f]				X				X				X
PCR of vector DNA in blood, saliva, urine, semen, and stools				X		X		X		X		X
Haemo-QOL-A assessment												X
EQ-5D-5L												X
HAL												X
WPAI+CIQ:HS												X

Assessment	Year 1 – Weeks*											
	33[e]	34	35[e]	36	38[e]	40	42[e]	44	46[e]	48	50[e]	52
Liver ultrasound[g]												X

* Visit windows are ± 48 hours through Week 36, then ±1 week until Week 52.

[a] Complete physical examination should be performed at Week 52; brief physical exam may be performed at other study visits. Weight should be recorded at Week 36, Week 44, and Week 52.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.4.1 for liver tests. LTs assessment may be checked more frequently when ALT values are > ULN or ≥ 1.5x baseline value or based upon discussion between the Medical Monitor and the Investigator. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: the ALT has increased to above ULN or increased by > 10 U/L from prior assessment; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen assay. Chromogenic Nijmegen-Bethesda Assay for hFVIII inhibitor level will be tested as deemed necessary by the Sponsor. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.

[d] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for any exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[e] The scheduled visits at Week 33, Week 35, Week 38, Week 42, Week 46, and Week 50 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject [or his legally authorized representative, if required] has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via

phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits.

[f] While exploratory samples for cytokine profiling will be collected at the time points indicated above, testing of these samples will be performed only as deemed necessary by the Sponsor.

[g] Additional liver ultrasounds may be performed prior to Week 52 at the discretion of the Investigator.

Table 9.1.5: Schedule of Activities – Long-Term Follow-Up (Year 2 – Year 5)

Assessment	Years 2-5*		End of Year Visit				ETV
			Year 2	Year 3	Year 4	Year 5	
Study Week*	Q12W	Q6W[gh]	W104	W156	W208	W260	
Physical examination[a]	X[a]		X[a]				X
Weight[a]	X[a]		X[a]				X
Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)	X	X	X				X
Vital Signs	X		X				X
Blood chemistry, hematology, and coagulation tests[b]	X[b]		X[b]				X
Urine Tests[b]	X[b]		X[b]				X
Liver Tests[b]	X	X	X				X
FVIII assays[c]	X	X	X				X
FVIII antigen assay	X		X				X
AAV5 TAb Assay	X (Year 2 only)		X				X
AAV5 TI Assay			X				X
FVIII antibody titer	X (Year 2 only)		X				X
Exploratory biomarker assessments[e]	X		X				X
PBMC Collection (for determination of FVIII and Capsid specific cellular immunity)	X[k]		X				X
VWF:Ag	X		X				X
Cytokine profiling[i]	X		X				X
PCR of vector DNA in semen[d]	(X)[d]	(X)[d]	(X)[d]				(X)[d]
PCR of vector DNA in blood, saliva, urine, and stools[d]	(X)[d]		(X)[d]				(X)[d]
Haemo-QOL-A assessment			X[f]				X
EQ-5D-5L			X[f]				X

Assessment	Years 2-5*		End of Year Visit				ETV
	Q12W	Q6W[gh]	Year 2	Year 3	Year 4	Year 5	
Study Week*			W104	W156	W208	W260	
HAL			X[f]				X
WPAI+CIQ:HS			X[f]				X
Liver Ultrasound[j]			X				X

ETV: Early Termination Visit

* Visit windows are ± 2 weeks for visits in Years 2-5. The Q6W visits during Years 2-5 should restart after each End of Year visit (eg, the first Q6W visit during Year 3 should be ~6 weeks after the End of Year 2 visit).

[a] Complete physical examination should be performed at the End of Year visits (genitourinary examination may be deferred); brief physical examination may be performed at Q12W visits. Weight should be recorded at the second Q12W visit each year and at every End of Year visit during Years 2-5.

[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.4.1 for liver tests. LT assessment may be checked more frequently when ALT values are > ULN or ≥ 1.5x baseline value or based upon discussion between the Medical Monitor and the Investigator. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN or increased by > 10 U/L from prior assessment; or (2) after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. During Years 2-5 of the Post-Infusion Follow-Up period, urine tests and blood, chemistry, and coagulation tests should be performed at the second Q12W visit each year and at every End of Year visit.

[c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, and coagulation exploratory assay. Chromogenic Nijmegen-Bethesda Assay for hFVIII inhibitor level will be tested as deemed necessary by the Sponsor. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism. If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.

[d] Sample testing during Long-Term Follow-Up is not required if at least 3 consecutive samples were below the limit of detection during the Post-Infusion Follow-Up period. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 52 should continue to have PCR testing of semen every 6 weeks during Years 2-5 until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).

[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for any exploratory assessments) will be performed only as deemed necessary by the Sponsor.

[f] PRO assessments during Years 2-5 of Long-Term Follow-up should be performed at every End of Year visit.

[g] Subjects who meet the definition of treatment failure to BMN 270 therapy after Week 52 (refer to Section 12.6) may omit the Q6W visits during Years 2-5, and must attend only the Q12W and End of Year visits. Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage. Such subjects following the abbreviated schedule who have not yet cleared vector shedding in semen must still provide samples Q6W during Years 2-5 until vector shedding has been cleared (but do not need to do other scheduled assessments at those visit dates).

[h] The scheduled Q6W visits during Years 2-5 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject [or his legally authorized representative, if required] has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits.

[i] While exploratory samples for cytokine profiling will be collected at the time points indicated above, testing of these samples will be performed only as deemed necessary by the Sponsor.

[j] Additional liver ultrasounds may be performed at interim timepoints (ie, between the End of Year visits) at the discretion of the Investigator.

[k] PBMC collection should occur at each Q12W visit during Year 2, then at every other Q12W visit during Years 3-5, as well as at all End of Year Visits for Years 2-5.

Table 9.1.6: Suggested Schedule of Activities – Prophylactic Corticosteroids

Assessment	Corticosteroid Treatment Period[b]																Post-Corticosteroid Period[c]				
	Day 1	Week															Week				
	1	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	1	2	3	4	13
Prophylactic corticosteroid dose (mg/day)	40 mg	40 mg	40 mg	40 mg	40 mg	35 mg	35 mg	30 mg	30 mg	25 mg	25 mg	20 mg	20 mg	15 mg	10 mg	5 mg					
FVIII activity testing	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Liver tests	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	
Hepatitis B testing[d]							X										X				X
HCV Viral Load[d]							X										X				X

[a] This table provides an example of a prophylactic corticosteroid course. Clinical judgment, weighing the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses, and discussions between the Investigator and Medical Monitor are advised for any questions or concerns.

[b] Following initiation or completion of corticosteroid regimen, if a recurrence of ALT values ≥ ULN or ≥ 2x baseline value is reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor. Modification of the corticosteroid regimen may take into consideration timing of ALT elevation (prior to or after Week 52), as well as possible confounders for the ALT elevation and adverse events related to corticosteroid dosing. Guidance for tapering oral corticosteroid dosing can be found in Section 9.4.8.2, although a discussion between the PI and Medical Monitor should take place prior to tapering the corticosteroid dose.

[c] After discontinuation of oral corticosteroids, weekly labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values. If these assessments are already being done as part of normal study follow-up, they do not need to be duplicated.

[d] Should only be performed in subjects with a history of hepatitis B or hepatitis C prior to study entry.

9.2 Discussion of Study Design, Including Choice of Control Group

Study 270-203 is designed to be a Phase 1/2, single-arm, open-label study in hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid. Hemophilia A patients who provide written informed consent (or who have a legally authorized representative provide consent, if required) and meet the entry criteria will be eligible to enroll in the study.

Approximately 10 subjects may be enrolled at the 6E13 vg/kg BMN 270 dose. Subjects will be followed for 26 weeks post-BMN 270 infusion during which safety and efficacy assessments will be taken. After the final analysis at 26 weeks post-infusion, safety and efficacy will then continue to be assessed long-term for approximately a total of 5 years.

9.3 Selection of Study Population

Approximately 10 hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid may enroll into the study.

Additional criteria for participation in the study are provided in Section 9.3.1 and Section 9.3.2.

9.3.1 Inclusion Criteria

Individuals eligible to participate in this study must meet all of the following criteria:

1. Males ≥ 18 years of age with hemophilia A and residual FVIII levels ≤ 1 IU/dL as evidenced by medical history, at the time of signing the informed consent
2. Detectable pre-existing antibodies against the AAV5 vector capsid as measured by AAV5 total antibody ELISA
3. Treated/exposed to FVIII concentrates or cryoprecipitate for a minimum of 150 exposure days (EDs)
4. Subject must have been on prophylactic FVIII replacement therapy for at least 12 months prior to study entry.
5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures. If the subject is unable to provide consent, a legally authorized representative may provide written informed consent.
6. No previous documented history of a detectable FVIII inhibitor, and results from a Bethesda assay or Bethesda assay with Nijmegen modification of less than 0.6 Bethesda Units (BU) (or less than 1.0 BU for laboratories with a historical lower sensitivity cutoff for inhibitor detection of 1.0 BU) on 2 consecutive occasions at least one week apart within the past 12 months (at least one of which should be tested at the central laboratory)

7. Sexually active participants must agree to use an acceptable method of effective contraception, either double barrier contraception (ie, condom + diaphragm; or condom or diaphragm + spermicidal gel or foam) or their female partner either using hormonal contraceptives or having an intrauterine device. Participants must agree to contraception use for at least 12 weeks post-infusion; after 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples with viral vector DNA below the limit of detection.
8. Willing to abstain from consumption of alcohol for at least the first 52 weeks following BMN 270 infusion.

9.3.2 Exclusion Criteria

Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study:

1. Any evidence of active infection, including COVID-19, or any immunosuppressive disorder except for HIV infection. HIV-positive patients who meet all other eligibility criteria may be included if they have a CD4 count > 200/mm3 and an undetectable viral load (unquantifiable viral load as defined as less than the limit of quantification by the testing laboratory's assay is permitted) while receiving an antiretroviral therapy (ART) regimen that does not contain efavirenz or another potentially hepatotoxic ART.
2. Significant liver dysfunction with any of the following abnormal laboratory results:

- ALT (alanine aminotransferase) > 1.25x ULN;
- AST (aspartate aminotransferase) > 1.25x ULN;
- GGT (gamma-glutamyltransferase) > 1.25x ULN;
- Total bilirubin > 1.25x ULN;
- Alkaline phosphatase > 1.25x ULN; or
- INR (international normalized ratio) ≥ 1.4

Subjects whose laboratory assessments fall outside of these ranges may undergo repeat testing of the entire liver test panel within the same Screening window and, if eligibility criteria are met on retest, may be enrolled after confirmation by the Medical Monitor.

3. Most recent, prior FibroScan or liver biopsy showing significant fibrosis of 3 or 4 as rated on a scale of 0-4 on the Batts-Ludwig (Batts 1995) or METAVIR (Bedossa 1996) scoring systems, or an equivalent grade of fibrosis if an alternative scale is used.
4. Evidence of any bleeding disorder not related to hemophilia A
5. Platelet count of < 100 x 10^9/L

6. Creatinine ≥ 1.5 mg/dL
7. Liver cirrhosis or other clinically significant liver disease of any etiology as assessed by liver ultrasound/FibroScan.
8. Chronic or active hepatitis B as evidenced by positive serology testing (hepatitis B surface antigen [HBsAg], hepatitis B surface antibody [HBsAb], and hepatitis B core antibody [HBcAb]) and confirmatory HBV DNA testing. Refer to the Centers for Disease Control (CDC) table for the interpretation of serological test results in the Laboratory Manual.
9. Active Hepatitis C as evidenced by detectable HCV RNA, or currently on antiviral therapy
10. Active malignancy, except non-melanoma skin cancer
11. History of hepatic malignancy
12. History of arterial or venous thromboembolic events (eg, deep vein thrombosis, non-hemorrhagic stroke, pulmonary embolism, myocardial infarction, arterial embolus), with the exception of catheter-associated thrombosis for which anti-thrombotic treatment is not currently ongoing.
13. Known inherited or acquired thrombophilia, including conditions associated with increased thromboembolic risk, such as atrial fibrillation.
14. A history of known inflammatory, connective tissue, or autoimmune disorders (eg, vasculitis).
15. Treatment with any Investigational Product within 30 days or 5 half-lives of the investigational product (whichever is longer) prior to the screening period. For subjects who have received a prior investigational product, all ongoing adverse events (AEs) experienced while receiving that investigational product must have resolved prior to screening for this study
16. Any condition that, in the opinion of the investigator or Sponsor would prevent the patient from fully complying with the requirements of the study (including corticosteroid treatment and/or use of alternative immunosuppressive agents outlined in the protocol) and/or would impact or interfere with evaluation and interpretation of subject safety or efficacy result.
17. Prior treatment with any vector or gene transfer agent
18. Major surgery planned in the 52-week period following the infusion with BMN 270
19. Use of systemic immunosuppressive agents, not including corticosteroids, or live vaccines within 30 days before the BMN 270 infusion
20. Concurrent enrollment in another clinical study, unless it is an observational (non-interventional) clinical study that does not interfere with the requirements of the current protocol or have the potential to impact the evaluation of safety and efficacy of BMN 270 and with prior consultation with the Medical Monitor

21. Known allergy or hypersensitivity to investigational product formulation
22. Unwilling to receive blood or blood products for treatment of an adverse event and/or a bleed

9.3.3 Removal of Subjects from Treatment or Assessment

Subjects may withdraw their consent to participate in the study at any time without prejudice. The Investigator must withdraw from the study any subject who requests to be withdrawn. Such subjects will always be asked about the reason(s) for withdrawal. The Investigator will discuss with the subject appropriate procedures for withdrawal from the study. The Investigator should ask the subject's consent to perform the procedures listed under the early termination visit. Should a subject withdraw from the study, all efforts will be made to complete and report the observations as thoroughly as possible up to the date of the withdrawal.

A subject's participation in the study may be discontinued at any time at the discretion of BioMarin or of the Investigator and in accordance with his/her clinical judgment. When possible, the tests and evaluations listed for the termination visit should be carried out and every effort will be made to gather follow-up safety data if possible.

BioMarin must be notified of all subject withdrawals as soon as possible. BioMarin also reserves the right to discontinue the study at any time for either clinical or administrative reasons and to discontinue participation by an individual Investigator or site for poor enrollment or noncompliance.

Reasons for which the Investigator or BioMarin may withdraw a subject from the study include, but are not limited to, the following:

- Subject requires medication or medical procedure prohibited by the protocol
- Subject was erroneously enrolled into the study or does not meet entry criteria and not yet been dosed with BMN 270; subjects who do not meet entry criteria but who erroneously receive BMN 270 should remain in the study for safety monitoring
- Subject is lost to follow-up

If a subject fails to return for scheduled visits, a documented effort must be made to determine the reason. If the subject cannot be reached by telephone, a certified letter should be sent to the subject requesting contact with the Investigator. This information should be recorded in the study records.

Subjects may be considered lost to follow-up if the subject has missed 3 consecutive visits in the study and has failed to communicate a reason for this to the site. In addition, the site has

documented at least 4 attempted contacts by key research personnel to reach the subject without success in the following manner:

- 2 attempts by telephone or email (if possible); then
- If telephone/email contacts are unsuccessful, 2 attempts must be made by certified letter or by appropriate local process.

Where communication has been made by phone, this should be documented in the subject source notes.

The Investigator or designee must explain to each subject, before enrollment into the study, that for evaluation of study results, the subject's protected health information obtained during the study may be shared with the study Sponsor, regulatory agencies, and IRB/EC It is the Investigator's (or designee's) responsibility to obtain written permission to use protected health information per country-specific regulations, such as the Health Insurance Portability and Accountability Act (HIPAA) in the US, from each subject. If permission to use protected health information is withdrawn, it is the Investigator's responsibility to obtain a written request, to ensure that no further data will be collected from the subject and the subject will be removed from the study.

9.3.3.1 Study Safety Evaluation Criteria

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC evaluation of safety and efficacy in treated subjects with the following triggers for pausing further enrollment:

- any related SAE;
- an related AE with a severity > Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

Additionally, the DMC should consider whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction. Following a temporary halt of enrolment, the DMC may approve resumption of enrolment in either cohort at a later date at its discretion based on further analysis of accumulating data.

If a Grade 4 or Grade 5 adverse event assessed as related to study drug occurs in a study subject, further enrollment into the trial will be halted until a full safety review of the data by the DMC has taken place. Relevant reporting and discussion with the Sponsor and the DMC will take place before resumption of dosing.

If any of the following events occur in a subject in the study who has received BMN 270 infusion, further enrollment into the trial will be temporarily put on hold pending prompt evaluation by the DMC.

1. Liver dysfunction (criteria do not apply to ALT elevations with an extra-hepatic etiology):
    - ALT >5x ULN, for more than 2 weeks
    - ALT >3x ULN and (total bilirubin >2x ULN **or** INR >1.5)
    - ALT >3x ULN with signs and symptoms of liver dysfunction
2. The occurrence of an AE of hepatic failure.
3. The detection of high titer neutralizing antibodies (>5 BU) to hFVIII following BMN 270 infusion in two subjects.
4. The occurrence of any cancer (except non-melanoma skin cancer) at any point after BMN 270 infusion.
5. The occurrence of a thromboembolic event with FVIII activity > 150 IU/dL in one subject.

If any of the following events occurs in a subject in the study who has received BMN 270 infusion, a prompt evaluation by the DMC will be required. Further enrollment into the trial will continue while DMC evaluation is ongoing, unless deemed otherwise by the DMC.

1. Acute hypersensitivity assessed as related to BMN 270
2. The detection of high titer neutralizing antibodies (>5 BU) to hFVIII following BMN 270 infusion in one subject
3. Occurrence of a thromboembolic event

9.3.4 Subject Identification and Replacement of Subjects

Each subject will be assigned a unique subject identifier. This unique identifier will be on all eCRF pages. In legal jurisdictions where use of initials is not permitted, a substitute identifier will be used.

Subjects who withdraw from the study after receiving BMN 270 will not be replaced.

9.3.5 Duration of Subject Participation

The duration of participation for each subject will be approximately 264 weeks. This includes 4 weeks of screening, 1 day of BMN 270 infusion, 26 weeks of Post-Infusion Follow-Up, and 234 weeks of Long-Term Follow-Up.

9.4 Treatments

9.4.1 Treatments Administered

BioMarin and/or its designee will provide the study site with a supply of IP sufficient for the completion of the study. BioMarin is responsible for shipping study drug to clinical sites.

9.4.2 Identity of Investigational Product

9.4.2.1 Product Characteristics and Labeling

BMN 270 is a sterile, clear, colorless-to-pale yellow solution for IV infusion and is supplied in a 10 mL Crystal Zenith® (CZ) vial. Each CZ vial contains 8.5 mL (extractable volume 8 mL) of AAV5-hFVIII-SQ at a concentration of 2E13 vector genomes per mL in a pH 7.4 phosphate buffer.

The study drug is labelled according to the particulars approved by the relevant regulatory agencies.

9.4.3 Storage

At the study site, all IP must be stored under the conditions specified in the Pharmacy Manual in a secure area accessible only to the designated pharmacists and clinical site personnel. All IP must be stored and inventoried and the inventories must be carefully and accurately documented according to applicable state, federal and local regulations, ICH GCP, and study procedures.

9.4.4 Directions for Administration

On the day of infusion, the subject will come to the infusion site, where a physical examination will be performed by the Investigator or designee. If the subject is found to have an active acute illness at the time of planned infusion, then the infusion should be deferred until the illness has resolved; screening procedures may require repetition if outside the specified window. An IV catheter or butterfly needle will be inserted into a suitable peripheral vein (eg, the median cubital vein) and flushed with saline. FVIII replacement therapy will not be given since venipuncture is a minimally invasive procedure in these individuals under ordinary conditions.

BMN 270 will be prepared and infused as a pure solution over a dose-dependent time. Prepared drug will be kept at room temperature prior to administration. An electric syringe pump will be used to infuse through an in-line, low protein binding 0.22 micron filter. BMN 270 will be infused through the catheter using an appropriate infusion pump at an initial rate of 1 mL/min. The infusion rate should be increased every 30 minutes by 1 mL/min

up to a maximum of 4 mL/min, provided that the subject's clinical condition permits such an increase. Of note, the IP has been shown to be stable at room temperature for approximately 10 hours following completion of product thaw. Vital signs (pulse, blood pressure, respiration rate and temperature) should be monitored at 15 minute (±5 minutes) intervals throughout the period of the infusion.

As with any infused biological product, there is a potential risk of acute, systemic hypersensitivity reactions (including anaphylaxis) with BMN 270. Dosing will be administered at a qualified infusion site, with appropriate resuscitation equipment and medication available and easily accessible.

Clinical staff administering BMN 270 should be trained appropriately in recognizing and managing the signs and symptoms associated with potential hypersensitivity, anaphylactic, and anaphylactoid reactions. Additionally, the Investigator should be familiar with Sampson's criteria for defining anaphylaxis (Sampson 2006; Appendix 1).

Should symptoms of potential hypersensitivity occur, the infusion may be slowed or halted at the Investigator's discretion, with consideration of the subject's clinical condition. If the infusion is halted, it should only be restarted if the Investigator considers it safe and appropriate to do so. Antihistamines, anti-pyretic, and/or corticosteroid administration is permitted prior to restarting an interrupted infusion by an infusion-related reaction. At the restart, the infusion rate may be adjusted (ie, to a slower rate [minimum of 1 mL/min], with the rate increased every 30 minutes by 1 mL/min up to a maximum rate of 4 mL/min, if the subject's clinical condition permits such an increase) with careful monitoring of the subject. In the event of an infusion rate reaction with more than one dosing interruption, the infusion rate would not go beyond 1 mL/min.

In case of hypersensitivity reactions, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine profiling, as well as possible additional exploratory testing) and samples for IgE and cytokine profiling (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine profiling. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.

Following completion of the infusion, vital signs will be monitored hourly (± 5 minutes). If the vital signs are stable the catheter will be removed 8 hours after the infusion. Hemostasis at the puncture site will be established by applying pressure according to standard protocol for infusing FVIII concentrates. Subjects will remain in the clinic for at least 24 hours to observe for any immediate toxicity of the procedure; in-patient observation can be extended beyond 24 hours if needed per Investigator discretion. After 24 hours, subjects will be discharged from the clinic unless toxicity has been observed in which case the stay in the clinic may be extended or the subject may transfer to a separate facility based on the evaluation and judgment of the Principal Investigator after consultation with the Medical Monitor.

Prior to discharging subjects from the clinic, the Investigator or designee should instruct subjects how to recognize signs and symptoms of potential (delayed) hypersensitivity reactions and anaphylaxis, and to contact a medical practitioner or seek emergency care in case of such an event.

9.4.5 Method of Assigning Subjects to Treatment Groups

Subjects who meet all eligibility criteria (refer to Section 9.3.1 and Section 9.3.2) may be enrolled into the study. Approval by the Medical Monitor will be required prior to enrollment of each study subject. Upon their enrollment into the study, subjects will be assigned a unique subject number.

9.4.6 Selection of Dose Used in the Study

Data from an ongoing first in human study (270-201) indicates that following single escalated doses of BMN 270 (6E12, 2E13, 4E13, 6E13 vg/kg), dose-related increases in FVIII activity were observed, with concurrent improvements in bleeding episodes and exogenous FVIII utilization, particularly at the 4E13 and 6E13 vg/kg dose levels. At all dose levels, BMN 270 is considered to be well-tolerated with mild increases in ALT as the most common adverse event. Please refer to the IB for detailed efficacy and safety data. The 6E13 vg/kg dose has been selected for this study to maximize the likelihood of transduction in the face of pre-existing AAV5 antibodies.

9.4.6.1 Selection of Timing of Dose for Each Subject

9.4.7 Blinding

This is an open-label study.

9.4.8 Prior and Concomitant Medications

All prescription and over-the-counter medications (including dietary and herbal supplements) taken by a subject for 30 days before Screening will be recorded on the designated eCRF. For HIV-positive patients, prior to enrollment, the Medical Monitor will review the patient's ART regimen to assess that it does not contain efavirenz or another potentially hepatotoxic ART. The Investigator may prescribe additional medications, deemed necessary to provide adequate prophylactic or supportive care, during the study, as long as the prescribed medication is not prohibited by the protocol. In the event of an emergency, any needed medications may be prescribed without prior approval, but the Medical Monitor must be notified of the use of any contraindicated medications immediately thereafter. Any concomitant medications added or discontinued during the study should be recorded on the eCRF. Medications should, whenever possible, not be recorded in the electronic database with a frequency of as needed (PRN).

The following medications are prohibited starting 30 days before Screening and through the end of the study, and the Sponsor must be notified if a subject receives any of these during the study:

- Any investigational therapy other than BMN 270
- Emicizumab
- Fitusiran
- Concizumab
- Efavirenz

Subjects who begin antiretroviral therapy following BMN 270 infusion and during their participation in 270-203 must have their antiretroviral medication regimen approved by the Investigator and Medical Monitor prior to starting antiretroviral treatment.

The following medications should be avoided, starting 30 days prior to and for at least 52 weeks after BMN 270 infusion and minimized throughout the remaining duration of the study.

- Alcohol
- Herbal and natural remedies and dietary supplements
- Medications which may be hepatotoxic, including isotretinoin and dextroamphetamine/amphetamine
- Medications which may reduce or increase the plasma concentration of corticosteroids

Subjects should be counseled to avoid starting potentially hepatotoxic therapies and to inform the Investigator of any new medications prescribed by other physicians. Investigators should carefully consider both the mechanism of action and potential hepatotoxicity of any new medication prior to initiation. If a potentially concerning new medication is started, Investigators should closely monitor both FVIII activity and ALT levels (eg, weekly to every 2 weeks for the first month) in order to determine if any detrimental effects on the efficacy or safety of BMN 270 have occurred. If co-medications are required during the course of the study, where possible, please check the National Center for Biotechnology Information LiverTox website for potential hepatotoxicity issues prior to prescribing (NCBI, 2020).

Vaccines should also be avoided during this period, but in particular during the first 26 weeks unless clinically indicated.

It is preferable for SARS-CoV-2 vaccination to occur prior to BMN 270 infusion. If a two-step SARS-CoV-2 vaccine is being used, sites should consider using the flexible re-screen option to allow subjects to receive both doses at least 14 days prior to treatment with BMN 270. If a live-virus SARS-CoV-2 vaccine is being used, subjects should wait at least 30 days after vaccination to receive a BMN 270 infusion. Investigators should use clinical judgment, taking into consideration local factors, individual risk factors, and the benefit/risk related to timing of vaccine administration. Administration of SARS-CoV-2 vaccine after BMN 270 infusion may occur after consultation between Investigator and Medical Monitor.

The following medications should be avoided during oral corticosteroid therapy:

- Vaccines
- NSAIDs

9.4.8.1 Concomitant Hemophilia Treatments

Subjects on prophylactic FVIII therapy will discontinue their regular treatment regimen starting 4 weeks after the day of infusion and switch to an episodic (or “on-demand”) schedule. FVIII replacement therapy can always be taken as needed by the subject for treatment of an acute bleeding episode; the subject must carefully record his treatment and bleeding episodes in his diary. Prophylactic FVIII can be used on a case-by-case basis and in consultation with the Medical Monitor to prevent bleeding in extenuating circumstances (eg, peri-operative).

In addition, information on FVIII usage and bleeding episodes by medical history will be collected from subjects for the 12-month period immediately preceding study enrollment.

9.4.8.2 Corticosteroid and/or Immunosuppressive Agent Treatment of Elevated Hepatic Transaminases

Refer to corticosteroid prescription guidelines for recommended monitoring for, and management of, potential side effects of corticosteroids, including guidance on medications that should be avoided during corticosteroid treatment.

9.4.8.2.1 Prophylactic Corticosteroids

All subjects will be started on prophylactic corticosteroids starting on the day of infusion (Day 1). Table 9.1.6 provides an example of the recommended prophylactic corticosteroid course, including taper and post-corticosteroid additional monitoring of FVIII activity, LTs, and hepatitis B/hepatitis C reactivation. Clinical judgment, weighing the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses. Discussions between the Investigator and Medical Monitor are advised for any questions or concerns.

9.4.8.2.2 Reactive Corticosteroids

Following initiation or completion of the prophylactic corticosteroid regimen, if ALT levels become increased (eg, > ULN or ≥ 1.5x baseline value) and alternative etiologies have been ruled out, prompt institution of newly administrated or an increased dose of reactive (ie, started in response to an ALT elevation) oral corticosteroids (prednisone or converted equivalent) should be considered after consultation with the Medical Monitor (refer to **Error! Unknown switch argument.**).

- Whenever possible, a confirmatory lab draw for ALT should be performed within 72 hours, along with FVIII activity, prior to initiating reactive oral corticosteroids.
- Newly administered corticosteroids or dose increases are not indicated if elevations in ALT are clearly not related to BMN 270 (eg, elevated ALT with concurrent increase in CPK due to intensive exercise) although this should be discussed with the Medical Monitor (in particular, for elevations occurring at least 52 weeks after the BMN 270 infusion).
- Alternative immunosuppressive agents may also be considered for use on a case-by-case basis and following consultation with the Medical Monitor (eg, if prolonged corticosteroid use is contraindicated).

Unless otherwise indicated, reactive corticosteroid treatment should be initiated at a dose of 60 mg/day. If the ALT level remains stable or declines after 2 weeks, consider gradual taper of corticosteroids: 40 mg/day for 4 weeks, 30 mg/day for 1 week, 20 mg/day for 1 week, and 10 mg/day for 1 week. Should a scenario arise in which a deviation from the minimum recommended dose and/or duration of reactive corticosteroids may be clinically indicated, a

discussion should take place between the Investigator and Medical Monitor regarding corticosteroid dose adjustments. Management of ALT elevations with reactive corticosteroids, including tapering of doses and managing worsening and/or recurrent ALT elevations, should be guided by the following (Table 9.4.8.2.2.1):

Table 9.4.8.2.2.1: Management of ALT Elevations with Reactive Corticosteroids

ALT ≥ 1.5x Baseline or > ULN	• Repeat LTs and FVIII within 24-72 hours • Continue to monitor LTs until ALT is stable or not increasing • Investigate for alternative etiologies (eg, concomitant medications, viral or autoimmune hepatitis, alcohol use, recreational drug use, special diets, strenuous exercise, prior and/or concurrent illnesses, exposure to environmental and/or industrial chemicals, etc.) • If no alternative etiology is found, initiate reactive corticosteroids with the following tapering schedule: 60 mg x 2 weeks; 40 mg x 3 weeks; 30 mg x 1 week; 20 mg x 1 week; 10 mg x 1 week upon consultation with the Medical Monitor • Consider evaluation with additional liver tests (including but not limited to ALT, AST, bilirubin, and alkaline phosphatase) • Consider obtaining other possibly relevant laboratory evaluations (albumin, PT/INR, CRP, etc.) • Consider obtaining complete blood count with differential to assess for eosinophilia • Consider obtaining PBMC, C3, C3a, Bb, and sC5b-9 to evaluate potential immune response (prior to starting reactive oral corticosteroids) • Continue to taper as long as subject's ALT is not increasing. Decisions regarding regimen modification may be made based upon Investigator judgement and discussion with the Medical Monitor • For any ALT elevations that begin after 52 weeks on study, please consult the Medical Monitor prior to initiating corticosteroids unless there is an imminent safety concern
Worsening ALT	If after 2 weeks ALT levels have worsened with corticosteroid dose of 60 mg/day, the following is recommended: • Investigate for alternative etiologies including labs noted above, if not previously checked • Increase corticosteroid dose up to a maximum of 1.2 mg/kg for no more than 2 weeks • For subjects who are refractory to the maximum dose of corticosteroids, or intolerant to use of corticosteroids, consider use of alternative immunosuppressants (tacrolimus or mycophenolate) • Consider gastroenterology and/or hepatology consult, abdominal workup, imaging (including MRI or ultrasound), and/or liver biopsy as appropriate Any concerns should be discussed between the Investigator and the Medical Monitor
Recurrent ALT elevations	If the subject has recurrent ALT elevations (≥ 1.5x Baseline or > ULN) and there are no safety concerns, the decision regarding management may be made at the discretion of the Investigator after discussion with the Medical Monitor

For any scenarios that are not accounted for in the above table, a discussion should take place between the Investigator and Medical Monitor regarding corticosteroid dose adjustments.

When ruling out alternative viral or autoimmune hepatitis as part of the elevated ALT workup, the following tests should be performed (Table 9.4.8.2.2.2):

Table 9.4.8.2.2.2: Viral and Autoimmune Hepatitis Testing

Viral Hepatitis Workup PCR Testing	Autoimmune Hepatitis Workup Testing
Hepatitis A	Smooth muscle antibody
Hepatitis B	Mitochondrial antibody
Hepatitis C	Liver/kidney microsomal antibodies
Hepatitis E	Antinuclear antibody (ANA) HEP-2
Cytomegalovirus (CMV)	
Epstein-Barr virus (EBV)	
Herpes simplex virus (HSV) 1 & 2	

After discontinuation of reactive oral corticosteroids, labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values.

Following completion of oral corticosteroids, if increased ALT levels (eg, > ULN or ≥1.5x baseline value) are reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor. Modification of the corticosteroid regimen may take into consideration possible confounders for the ALT elevation and impact on FVIII expression.

Management and monitoring of reactions to corticosteroids should be determined by the Investigator's clinical judgment in consultation with the Sponsor's Medical Monitor. This includes the contraindicated use of NSAIDs during corticosteroid treatment and specific monitoring not already covered by the SoA. The use of COX-2 inhibitors, while not contraindicated during corticosteroid treatment, should be limited, if possible. Practical management to prevent complications related to oral corticosteroid therapy may be undertaken at the discretion of the Investigator (eg, evaluation of glucose intolerance, hyperlipidemia). Alternative, non-steroidal systemic immunosuppressive agents may be used, following a discussion between the Investigator and the Medical Monitor, should corticosteroid use be deemed by an Investigator to be clinically ineffective, not tolerated, and/or contraindicated. Hepatitis B status and HCV viral load will be rechecked 6 weeks after the start of oral corticosteroid/immunosuppressive agent treatment and then 1 week and 13 weeks after the completion of oral corticosteroid/immunosuppressive agent treatment in

subjects with a history of hepatitis B or hepatitis C. All adverse events (including any adverse events suspected to be caused by or related to corticosteroid/immunosuppressive agent use) should be reported as outlined in Section 10 of the protocol.

Subjects on corticosteroids should receive appropriate counselling and support regarding side effects from the Investigator or the treating institution (eg, listings of side effects and when to notify carers, wallet card for emergencies if on steroids, etc.). Additional management, including the co prescription of additional medications to prevent complications related to corticosteroid therapy, may be undertaken at the discretion of the investigator, including, but not limited to, prophylaxis against the occurrence of gastric ulcers, osteoporosis, and infections. The above guidance should also be followed in the event that an alternative immunosuppressive agent is used, as applicable.

9.4.8.3 Monitoring of HIV-Positive Subjects

HIV-positive subjects may be enrolled in 270-203 if the subject is well controlled on an ART regimen that does not contain efavirenz or another potentially hepatotoxic ART, has a CD4 count > 200/mm^3, and has an undetectable viral load (unquantifiable viral load as defined as less than the limit of quantification by the testing laboratory's assay is permitted).

HIV-positive subjects were initially included in prior BMN 270 studies. However, after an HIV-positive subject in 270-302 developed markedly elevated liver enzyme levels after receiving 4E13 vg/kg of BMN 270, out of an abundance of caution for the long-term liver health of HIV-positive patients, further enrollment of HIV-positive subjects was suspended in 270-301 (Protocol Amendment 3) and 270-302 (Protocol Amendment 3). The subject in 270-302 referenced above was receiving efavirenz and lamivudine as part of his ART regimen. Following discussion with a liver advisory board and review of the accumulated 270-301 data, efavirenz and not lamivudine has been implicated as the most likely medication that interacted with BMN 270 and contributed to the 270-302 subject's elevated liver enzyme levels. Due to its hepatotoxicity, efavirenz is a prohibited medication in all BMN 270 studies.

The two HIV-positive subjects on stable, non-efavirenz-containing ART regimens who were enrolled in and dosed in 270-301 study prior to Amendment 3 have been monitored closely. Following BMN 270 infusion, these subjects continued their ART as prescribed and followed routine monitoring of CD4 count and viral load. Results from 270-301 show similar safety results for the two HIV-positive subjects compared to those who are HIV-negative. The Sponsor believes that HIV infection, in and of itself, is not a contraindication to receive BMN 270 and has therefore removed the exclusion of HIV-positive subjects.

Subjects should continue ART as prescribed and follow routine monitoring of CD4 count and viral load (US Dept Health Human Services 2019). Investigators will continue to monitor HIV-positive subjects per routine standard of care.

9.4.9 Treatment Compliance

Study drug will be administered to subjects at the dosing facility by a qualified health care professional. The quantity dispensed, returned, used, lost, etc. must be recorded on a dispensing log. Sites will be instructed to return or destroy all used and unused study drug containers.

9.5 Investigational Product Accountability

The Investigator or designee is responsible for maintaining accurate records (including dates and quantities) of IP(s) received and IP lost or accidentally or deliberately destroyed. The Investigator or designee must retain all unused or expired study supplies until the study monitor (on-site CRA) has confirmed the accountability data, if allowed by local SOPs.

9.5.1 Return and Disposition of Clinical Supplies

Unused study drug must be kept in a secure location for accountability and reconciliation by the study monitor. The Investigator or designee must provide an explanation for any destroyed or missing study drug or study materials (or must be referenced in their institution SOPs).

Unused study drug may be destroyed on site, per the site's standard operating procedures, but only after BioMarin has granted approval for drug destruction. The monitor must account for all study drug in a formal reconciliation process prior to study drug destruction. All study drug destroyed on site must be documented. Documentation must be provided to BioMarin or designee and retained in the Investigator study files. If a site is unable to destroy study drug appropriately, the site can return unused study drug to BioMarin upon request. The return of study drug or study drug materials must be accounted for on a Study Drug Return Form provided by BioMarin.

All study drug and related materials should be stored, inventoried, reconciled, and destroyed or returned according to applicable state and federal regulations and study procedures. For additional information, please refer to the Study Pharmacy Manual.

9.6 Dietary or Other Protocol Restrictions

There are no dietary or other protocol restrictions for this study. Alcohol should be avoided starting 30 days prior to and for at least the first 52 weeks of the study, and particularly

within 48 hours prior to lab work. Alcohol use should be minimized throughout the remaining duration of the study.

Subjects should be advised to abstain from any blood, organ, or sperm donation after BMN 270 infusion, until there is no further evidence of vector shedding from PCR analysis of samples.

9.7 Efficacy and Safety Variables

9.7.1 Efficacy and Safety Measurements Assessed

The SoA (Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5) describe the timing of required evaluations.

9.7.2 Efficacy Variables

9.7.2.1 FVIII Activity

Efficacy (response to treatment) will be defined as FVIII activity ≥ 5 IU/dL at Week 26 following BMN 270 infusion.

Values for FVIII activity will be excluded from analysis if obtained within 72 hours since the last infusion of exogenous FVIII protein concentrates. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; The 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy.

Note that fluctuations in FVIII activity after gene therapy are common, and more frequent monitoring of FVIII activity levels is not needed in the absence of a concurrent or recent ALT elevation or upon consultation between the Investigator and the Medical Monitor.

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 and inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 52, as applicable and at the discretion of the Investigator. Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.

Details on collecting FVIII activity samples are included in the Laboratory Manual.

9.7.2.2 Factor VIII Replacement Therapy/Bleeding Episodes

Additional efficacy variables are:

- Change of annualized utilization (IU/kg) of exogenous FVIII replacement therapy during Week 5 to Week 26 post BMN 270 infusion from the baseline utilization of exogenous FVIII replacement therapy.
- Change of ABRs requiring exogenous FVIII replacement treatment during Week 5 to Week 26 of the study post BMN 270 infusion from the baseline ABR.

During the study, subjects will be asked at each study visit to report the use of factor replacement therapy and the number of bleeding episodes since the previous visit. This information will be captured on the subject's diary or other subject records. Subjects will be encouraged to discuss any bleeding episodes with the Investigator and attempt to objectively assess any reported bleeds through use of ultrasound or non-invasive imaging.

Subjects are strongly encouraged to immediately consult the Investigator for guidance regarding exogenous FVIII administration for suspected bleeds or bleeding episodes within the first 6 weeks post BMN 270 infusion.

In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis.

9.7.2.3 Patient-Reported Outcome (PRO) Measures

The Haemo-QoL-A questionnaire is a validated hemophilia-specific health-related quality of life questionnaire for adults (Rentz 2008). It consists of 41 questions covering six domains (Physical Functioning, Role Functioning, Worry, Consequences of Bleeding, Emotional Impact and Treatment Concerns). Items are answered on a 6-point Likert-type scale, ranging from 0 (None of the time) to 5 (All of the time). Higher scores mean better health-related quality of life or less impairment for a particular subscale (Haemo-QoL Study Group 2017). Details regarding the Haemo-QoL-A assessment will be included in the Study Reference Manual.

The EQ-5D-5L instrument is a self-reported questionnaire designed to measure general health status (The EuroQol Group 1990; Brooks 1996). The EQ-5D-5L is composed of 2-parts: a descriptive system that assesses 5 levels of perceived problems (mobility, self-care, usual activities, pain/discomfort, and anxiety/depression) in 5 dimensions and the EQ visual analogue scale (EQ VAS) assessment for overall health. A sample copy of the EQ-5D-5L and additional information are provided in the Study Reference Manual.

The Haemophilia Activities List (HAL) measures the impact of hemophilia on self-perceived functional abilities in adults (van Genderen 2006). The instrument consists of multiple

domains including lying/sitting/kneeling/standing, leg and arm function, use of transportation, self-care, household tasks, and leisure activities where subjects are asked to rate their level of difficulty with activities of daily living on a 6-point Likert-type scale from 1 (Impossible) to 6 (Never). For some items, subjects are given the choice to answer 'Not applicable'. A sample copy of the HAL and additional information are provided in the Study Reference Manual.

The Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) instrument is designed to measure the effect of disease symptom severity on work productivity and classroom productivity (if applicable) (Recht 2014). The WPAI+CIQ:HS questionnaire yields scores related to work/classroom absenteeism, reduced on-the-job effectiveness, overall work/classroom impairment, and activity impairment. WPAI+CIQ:HS outcomes are expressed as impairment percentages, with higher numbers indicating greater impairment and less productivity (Reilly 2002). A sample copy of the WPAI+CIQ:HS and additional information are provided in the Study Reference Manual.

9.7.3 Immunogenicity

Immunogenicity assays will be performed on plasma and PBMCs. The assays will include detection of anti-AAV5 vector capsid and anti-FVIII total antibodies, as well as determination of neutralizing antibodies against FVIII (FVIII inhibitors) and against the AAV5 vector capsid (Transduction Inhibitors, TI). FVIII Inhibitors will be assessed using the Bethesda assay with Nijmegen modification. Any abnormality of the liver parameters will lead to a retrospective immunogenicity assessment to evaluate FVIII-and vector capsid-specific cellular immunogenicity. FVIII- and vector capsid-specific cellular immunity will be assessed by stimulated cytokine secretion using an ELISpot assay performed on collected PBMCs.

9.7.4 Pharmacodynamics

The FVIII protein concentration and activity level as measured by a validated immunoassay and by a validated FVIII activity assay, respectively, will be used for plasma profiles; FVIII protein and activity will be used to determine PD parameters.

9.7.5 Exploratory Assessments

A cytokine profiling assessment will be performed at Baseline and then at the timepoints listed in the Schedule of Activities.

In addition, blood samples will be collected from subjects at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5 to evaluate biochemical,

molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV5 gene transfer, and to develop assays used for these evaluations. Subject may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A.

All biomarker samples collected in this study may be used for exploratory biomarker research, including evaluation of additional biomarkers not specifically listed in the protocol. In addition, samples collected for other purposes in this study may be used for exploratory research once testing for the primary purpose has been completed.

9.7.6 Safety Variables

Safety in this study will be determined from evaluation of AEs, clinical laboratory assessments with a particular attention to the liver function, vital signs assessments, physical examinations, and immunogenicity.

9.7.6.1 Adverse Events

The determination, evaluation and reporting of AEs will be performed as outlined in Section 10.

9.7.6.2 Clinical Laboratory Assessments

The scheduled clinical laboratory tests are listed in Table 9.7.6.2.1. Refer to the Study Reference Manual for instructions on obtaining and shipping samples. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample.

Any abnormal test results determined to be clinically significant by the Investigator should be repeated (at the Investigator's discretion) until: (1) the cause of the abnormality is determined; (2) the value returns to baseline or to within normal limits; or (3) the Investigator determines that the abnormal value is no longer clinically significant.

All abnormal clinical laboratory results should be initialed and dated by an Investigator, along with a comment regarding whether or not the result is clinically significant. Each clinically significant laboratory result should be recorded as an adverse event.

The diagnosis, if known, associated with abnormalities in clinical laboratory tests that are considered clinically significant by the Investigator will be recorded on the AE eCRF.

Table 9.7.6.2.1: Clinical Laboratory Tests

Blood Chemistry	Hematology	Urine Tests	Coagulation Screen including:
Albumin	Hemoglobin	Appearance	APTT
BUN	Hematocrit	Color	PT/INR
Calcium	WBC count	pH	TT
Chloride	RBC count	Specific gravity	
Total cholesterol	Platelet count	Ketones	
CPK	Differential cell count	Protein	
Creatinine	RBC indices (MCV and MCH)	Glucose	
CRP		Bilirubin	Other Tests:
Glucose		Nitrite	ABO blood typing*
Phosphorus		Urobilinogen	
Potassium		Hemoglobin	
Total protein			
Sodium			
Uric Acid			

BUN, blood urea nitrogen; CPK, creatinine phosphokinase; CRP, C-reactive protein; PT, prothrombin time; APTT, activated partial thromboplastin time; RBC, red blood cell; WBC, white blood cell; TT, thrombin time; INR, international normalized ratio; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin.

*ABO blood typing assessment should be performed at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study.

In addition to scheduled clinical laboratory assessments, a fasting blood lipid panel (including triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) will be assessed at the BMN 270 infusion visit. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit.

In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine profiling, as well as possible additional exploratory testing) and samples for IgE and cytokine profiling (and possible additional exploratory testing) between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine profiling. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to

better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.

At applicable sites, certain study assessments may be performed by a mobile nursing (MN) professional at the patient's home or another suitable location, such as their school or office, to improve access and convenience for patients participating in the study. The Sponsor may select a healthcare company that will be responsible for providing MN services for participating sites (the MN vendor). The MN vendor is responsible for ensuring that all MN professionals are licensed, qualified, and in good standing, as per applicable regulations, and that appropriate background checks have been performed. If the investigator at a participating site determines that MN services are appropriate for a patient and the patient (or his legally authorized representative, if required) gives written informed consent to participate in MN visits, the MN network will communicate with the patient and the patient's site. Potential MN visits are designated in the Schedules of Events.

9.7.6.3 Malignancies

A liver ultrasound (and FibroScan, at the discretion of the Investigator) will be performed at Screening to screen for HCC. Thereafter, liver ultrasounds will be performed annually at each End of Year visit starting at Year 1 (Week 52) through the end of the study to screen for HCC. Additional liver ultrasounds may be performed prior to Week 52 and/or between the End of Year visits at the discretion of the Investigator.

Any development of a malignancy (except non-melanoma skin cancers) during the course of the study will be considered an EOSI (refer to Section 10.2.1) and is subject to expedited reporting. In addition, it is recommended that genomic analyses be performed on any malignancy (except non-melanoma skin cancers) diagnosed during the course of the study. The study site will coordinate sending samples from the malignancy for genomic analyses, if available.

9.7.6.4 Liver and Hepatitis Testing

Subjects will be screened for evidence of previous or active hepatitis B or hepatitis C infection at Screening; hepatitis B screening should include HBsAg, HBsAb, and HBcAb. Subjects with documented results showing an absence of active hepatitis B or hepatitis C infection (as measured by negative surface antigen or DNA for hepatitis B or negative RNA testing for hepatitis C) 30 days prior to providing signed informed consent do not need to repeat those tests during the screening period.

Evidence of ongoing hepatitis B or hepatitis C infection is exclusionary. Subjects with history of hepatitis B or hepatitis C infection prior to study entry will be tested for hepatitis B

and hepatitis C reactivation at Week 16. Subjects with a history of hepatitis B or hepatitis C will be asked for information about the treatments received as part of their medical history assessment at Screening.

Subjects with a previous history of hepatitis B or hepatitis C who receive oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.6.

A liver ultrasound, fasting FibroTest, and liver tests (LTs) during Screening will be performed to assess for clinically significant liver disease and HCC. A FibroScan may also be performed during Screening at the discretion of the Investigator. Fasting FibroTest results must be available prior to BMN 270 infusion.

Where a biopsy has been taken for safety-related reasons or was available from a past procedure, the Sponsor may request the biopsy information to help evaluate the impact of BMN 270 on the liver. The Sponsor may request that slides from a liver biopsy be made available for additional histopathological review.

Liver tests will be monitored on a regular basis; at each time point specified in the SoA, the following LTs should be assessed:

Table 9.7.6.4.1: Liver Tests

Liver Tests (LTs)			
Alkaline Phosphatase	AST (SGOT)	Total Bilirubin	LDH
ALT (SGPT)	Direct Bilirubin	GGT	

ALT, alanine aminotransferase; AST, aspartate aminotransferase; GGT, gamma-glutamyltransferase; LDH, lactate dehydrogenase; SGOT, serum glutamic-oxaloacetic transaminase; SGPT, serum glutamic-pyruvic transaminase

Elevated ALT levels should be evaluated according to plan outlined in Table 9.4.8.2.2.1 (note that these evaluations may indicate additional testing of LTs and FVIII levels at unscheduled visits; these unscheduled laboratory tests may be completed by a mobile nursing professional at sites where the use of MN services has been approved):

9.7.6.5 HIV Testing

HIV testing will be performed at Screening. Subjects with documented negative results within the last 30 days prior to screening do not need to be retested. Refer to Section 9.4.8.3 for guidance on monitoring of HIV-positive subjects.

9.7.6.6 Vital Signs, Physical Examinations and Other Observations Related to Safety

Vital signs will include seated systolic and diastolic blood pressure, heart rate, respiration rate, and temperature. Any clinically significant change in vital signs will be recorded as an AE.

Systolic blood pressure, diastolic blood pressure, heart rate, respiration rate, and temperature will be assessed at Screening, Baseline, and at the beginning of each visit during the Post-Infusion Follow-Up and Long-Term Follow-Up periods. On the day of the BMN 270 Infusion, vital signs will be monitored prior to infusion, during the infusion every 15 minutes (± 5 minutes), following the infusion hourly (± 5 minutes) for at least 8 hours during the subject's stay in the clinic. Any abnormal vital sign assessments should be repeated, and both values should be recorded in the eCRF.

A complete physical examination should be performed at Screening, Week 26, Week 52, and at the End of Year visits. A complete physical examination will include general appearance (head, eyes, ears, nose, and throat), cardiovascular, dermatologic, lymphatic, respiratory, gastrointestinal, genitourinary, musculoskeletal, and neurologic systems. The genitourinary examination may be deferred for visits after Year 1 unless the subject has genitourinary-related complaints.

At other visits, brief physical examinations may be performed at the discretion of the Investigator based on the subject's clinical condition. A brief physical examination will include general appearance, cardiovascular, dermatologic, respiratory, gastrointestinal, musculoskeletal, and neurologic assessments. Particular attention should be given to signs of bleeding, as well as assessing possible hemarthroses.

Height will be recorded at Screening only. Weight will be recorded at Screening and then every 4 weeks thereafter through Week 20, at Week 26, Week 32, Week 36, Week 44, and Week 52, and then at the second Q12W visit each year and at every End of Year visit during Years 2-5.

At visits where the MN services are used or shortened lab draw-only visits are conducted at the sites, the physical examination and vital signs assessments indicated in the Schedule of Activities will not be performed.

9.7.6.7 Vector Shedding

During the Post-Infusion Follow-Up period, subjects will undergo testing of various bodily samples to look for evidence of vector shedding for possible viral transmission. Bodily fluids will be tested by polymerase chain reaction (PCR). Fluids tested will include:

- Blood
- Saliva
- Semen
- Urine
- Stool

Vector shedding will also be extensively studied in the present clinical trial, at the time points indicated in Table 9.1.1, Table 9.1.2, Table 9.1.3, Table 9.1.4, and Table 9.1.5. Testing will continue until at least 3 consecutive results below the limit of detection are obtained. If a positive result is obtained in a matrix after 3 consecutive results below the limit of detection have already been recorded, testing in that matrix should restart and continue until an additional 3 consecutive results below the limit of detection have been obtained in order to confirm clearance.

Testing of semen will continue at least through Week 12, even if 3 consecutive results below the limit of detection have been recorded in that compartment prior to that time point. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 26 should continue to have PCR testing in semen at the timepoints designated in the SoA until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor). Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule (refer to Section 12.5.3) but who have not cleared vector shedding from a matrix must still provide samples for assessment until vector shedding has cleared: at Weeks 32, 36, 40, 44, 48, and 52 (during Year 1) and every 6 weeks (for semen samples) or every 12 weeks (for all uncleared matrices) during Years 2-5. Such subjects may provide samples on the designated study visit dates either at the sites or through use of a MN professional.

Samples may be fractionated prior to shedding analysis in order to better characterize the presence, structure, and location of vector DNA and/or vector capsid within each matrix. If needed, the fractionation may be performed with samples collected specifically for shedding analysis (saliva, blood, semen, urine, feces). Alternatively, the vector DNA characterization during shedding analysis may utilize already fractionated exploratory samples obtained from

the above biofluids, such as exploratory plasma samples, exploratory PBMC samples, and red blood cells recovered during PBMC/plasma isolations.

Fractionation of semen to collect purified sperm separately from non-sperm cells may be performed in parallel at any visit where semen samples are collected. The shedding analysis of a fractionated semen sample will only be performed if vector DNA was detected in the whole semen sample for the same visit. Fractionation of semen during shedding analysis may be stopped if purified sperm tested positive for vector DNA on at least three visits, or if purified sperm tested below the limit of detection for vector DNA on at least three consecutive visits.

Contraception use may need to be extended beyond 12 weeks in individual subjects based on observed vector shedding in semen. After 12 weeks, subjects may stop contraception use only if they have had 3 consecutive semen samples below the limit of detection (upon consultation between the Investigator and Medical Monitor).

Details for sample collection and storage are provided in the Laboratory Manual.

10 REPORTING ADVERSE EVENTS

10.1 Safety Parameters and Definitions

Safety assessments will consist of monitoring and recording protocol-defined AEs and SAEs; measurement of protocol-specified hematology, clinical chemistry, and urinalysis variables; measurement of protocol-specified vital signs; and other protocol-defined events of special interest that are deemed critical to the safety evaluation of the study drug.

10.1.1 Adverse Events

For this protocol, an adverse event (AE) is any untoward medical occurrence in a subject, temporally associated with the use of study intervention, whether or not considered related to the study intervention.

An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of study intervention.

Events meeting the AE definition include:

- Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition.
- New conditions detected or diagnosed after study intervention administration even though it may have been present before the start of the study.
- Signs, symptoms, or the clinical sequelae of a suspected drug-drug interaction.

Events not meeting the AE definition include:

- Any clinically significant abnormal laboratory findings or other abnormal safety assessments which are associated with the underlying disease, unless judged by the investigator to be more severe than expected for the subject's condition.
- The disease/disorder being studied or expected progression, signs, or symptoms of the disease/disorder being studied, unless more severe than expected for the subject's condition.
- Medical or surgical procedure (eg, endoscopy, appendectomy): the condition that leads to the procedure is the AE.
- Situations in which an untoward medical occurrence did not occur (social and/or convenience admission to a hospital).
- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

10.1.1.1 Bleeding and Suspected Bleeding Events

All bleeding events and suspected bleeding events, regardless of the need for exogenous FVIII therapy as treatment, should be captured in subject diaries and recorded on the designated bleeding eCRF. Bleeding events and suspected bleeding events should not be reported as adverse events, with the following exception:

- All bleeding events and suspected bleeding events which meet one or more of the criteria for being serious (refer to Section 10.1) should be reported as serious adverse events (whether or not they are bleeding events that are normal sequelae of hemophilia, and whether or not they required exogenous FVIII as treatment).

10.2 Serious Adverse Events

A serious adverse event (SAE) is any untoward medical occurrence that, at any dose:

- Results in death
- Is life-threatening

  Note: Life-threatening refers to an event in which the subject was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.

- Requires inpatient hospitalization or prolongation of existing hospitalization
    - Note: In general, hospitalization signifies that the subject has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization, the event is serious.

  Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE.
- Results in persistent or significant disability or incapacity
- Is a congenital anomaly or birth defect in the child or fetus of a subject exposed to IP prior to conception or during pregnancy
- Is an important medical event or reaction – that, based on medical judgment, may jeopardize the subject or require medical/surgical intervention to prevent one of the other outcomes listed above (eg, anaphylaxis)

10.2.1 Events of Special Interest (EOSI)

The following EOSI need to be reported to the Sponsor within 24 hours of site awareness, irrespective of seriousness, severity or causality:

- Elevation of ALT > ULN or ≥1.5x baseline value, regardless of whether that elevation triggers an initiation or modification of oral corticosteroid treatment
- Events potentially meeting the criteria for Hy's law (ALT or AST elevation ≥ 3x ULN plus total bilirubin ≥ 2x ULN)
- Thromboembolic event
- Systemic hypersensitivity, anaphylactic, or anaphylactoid reactions (refer to Appendix 1)
- Development of anti-FVIII inhibitory antibodies (inhibitors)
- Any new diagnosis of malignancy (except non-melanoma skin cancer)

10.3 Methods and Timing for Capturing and Assessing Safety Parameters

10.3.1 Adverse Event Reporting Period

The study AE reporting period is as follows: After informed consent but prior to initiation of study drug, only SAEs associated with any protocol-imposed interventions will be collected. After informed consent is obtained and following infusion of study drug, the reporting period for all non-serious AEs and SAEs begins and continues for approximately 5 years or until study discontinuation/termination, whichever is longer. The criteria for determining, and the reporting of SAEs is provided in Section 10.1.

10.3.2 Eliciting Adverse Events

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and non-leading verbal questioning of the subject is the preferred method to inquire about AE occurrences. The Investigator will record all relevant AE/SAE/EOSI information in the subject's medical record and AE Case Report Form (eCRF).

10.3.3 Assessment of Seriousness, Severity, and Causality

The Investigator responsible for the care of the subject or medically qualified designee will assess AEs for severity, relationship to study drug and/or corticosteroids and/or other immunosuppressive agents, and seriousness (refer to Section 10.2 for SAE definitions). These assessments must be made by a study clinician with the training and authority to make a diagnosis (eg, MD/DO, physician's assistant, nurse practitioner, or DDS).

10.3.3.1 Seriousness

The Investigator will assess if an AE should be classified as "serious" based on the seriousness criteria enumerated in Section 10.2. Seriousness serves as a guide for defining regulatory reporting obligations.

10.3.3.2 Severity

Severity (as in mild, moderate, or severe headache) is not equivalent to seriousness, which is based on subject/event outcome or action criteria usually associated with events that pose a threat to a subject's life or functioning. The Investigator will determine the severity of each AE, SAE and EOSI using the NCI CTCAE v4.03. Adverse events that do not have a corresponding CTCAE term will be assessed according to the general guidelines for grading used in the CTCAE v4.03 as stated in Table 10.3.3.2.1.

Table 10.3.3.2.1: Adverse Event Grading (Severity) Scale

Grade	Description	
1	Mild: asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated	
2	Moderate: minimal, local or noninvasive intervention indicated; limiting age-appropriate instrumental activities of daily living (ADL)[a]	
3	Severe or medically significant but not immediately life-threatening: hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL[b]	
4	Life threatening consequences; urgent intervention indicated	Grade 4 and 5 AEs should always be reported as SAEs
5	Death related to AE	

[a] Instrumental ADL refer to the following examples: preparing meals, shopping for groceries or clothes, using the telephone, managing money.

[b] Self-care ADL refer to the following examples: bathing, dressing and undressing, feeding oneself, using the toilet, taking medications, not bedridden.

10.3.3.3 Causality

The Investigator will determine the relationship of an AE to the study drug and/or corticosteroids and/or other immunosuppressive agents and will record it on the source documents and AE eCRF. To ensure consistency of causality assessments, Investigators should apply the guidance in Table 10.3.3.3.1.

Table 10.3.3.3.1: Causality Attribution Guidance

Relationship	Description
Not Related	• Exposure to the IP and/or corticosteroids and/or other immunosuppressive agents has not occurred **OR** • The administration of the IP and/or corticosteroids and/or other immunosuppressive agents and the occurrence of the AE are not reasonably related in time **OR** • The AE is considered likely to be related to an etiology other than the use of the IP and/or corticosteroids and/or other immunosuppressive agents; that is, there are no facts, evidence, or arguments to suggest a causal relationship to the IP and/or corticosteroids and/or other immunosuppressive agents.
Related	• The administration of the IP and/or corticosteroids and/or other immunosuppressive agents and the occurrence of the AE are reasonably related in time **AND** • The AE could not possibly be explained by factors or causes other than exposure to the IP and/or corticosteroids and/or other immunosuppressive agents **OR** • The administration of IP and/or corticosteroids and/or other immunosuppressive agents and the occurrence of the AE are reasonably related in time **AND** • The AE is more likely explained by exposure to the IP and/or corticosteroids and/or other immunosuppressive agents than by other factors or causes.

Factors suggestive of a causal relationship could include (but are not limited to):

- Plausible temporal relationship
- Absence of alternative explanations
- Rarity of event in a given patient or disease state
- Absence of event prior to study drug and/or corticosteroid and/or other immunosuppressive agent exposure
- Consistency with study product pharmacology
- Known relationship to underlying mechanism of study drug and/or corticosteroid and/or other immunosuppressive agent action

- Similarity to adverse reactions seen with related drug products
- Abatement of AE with discontinuation of study drug and/or corticosteroids and/or other immunosuppressive agents, and/or recurrence of AE with reintroduction of study drug and/or corticosteroids and/or other immunosuppressive agents

The Investigator's assessment of causality for individual AE reports is part of the study documentation process. Regardless of the Investigator's assessment of causality for individual AE reports, the Sponsor will promptly evaluate all reported SAEs against cumulative product experience to identify and expeditiously communicate possible new safety findings to Investigators and applicable regulatory authorities.

10.4 Procedures for Recording Adverse Events

10.4.1 Recording Adverse Events on a eCRF

Investigators should use precise medical terminology when recording AEs or SAEs on the AE eCRF. Avoid colloquialisms and abbreviations.

Record only one diagnosis, sign, or symptom per event field on the AE eCRF (eg, nausea and vomiting should not be recorded in the same entry, but as 2 separate entries).

In order to classify AEs and diseases, preferred terms will be assigned by the Sponsor to the original terms entered on the AE eCRF, using MedDRA (Medical Dictionary for Regulatory Activities) terminology.

10.4.1.1 Diagnosis versus Signs and Symptoms

The Investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE. Using accepted medical terminology, enter the diagnosis (if known). If not known, enter sign(s) and/or symptom(s). If a diagnosis subsequently becomes available, then this diagnosis should be entered on the AE (or SAE, as appropriate) eCRF, replacing the original entries where appropriate.

10.4.1.2 Persistent or Recurrent Adverse Events

A persistent AE (duration of adverse event > 7 days) is one that extends continuously, without resolution, between subject evaluation time points. Events that change in severity necessitate the recording of an additional AE. AEs that do not have a change in severity should be recorded only once on the eCRF.

A recurrent AE is one that occurs and resolves between subject evaluation time points, but then subsequently recurs. All recurrences of the AE should be recorded on the AE eCRF. For example, if a subject has an adverse event of ALT increased that subsequently resolves,

but the subject's ALT increases again, that should be reported as two adverse events – the initial ALT increase, and the second ALT increase.

10.4.1.3 Abnormal Laboratory Values

Laboratory test results (including any local FVIII activity or liver test results) will be recorded on the laboratory results pages of the eCRF, or appear on electronically produced laboratory reports submitted directly from the central laboratory, if applicable.

Any laboratory result abnormality fulfilling the criteria for a SAE or EOSI should be reported as such, and recorded in the AE eCRF unless associated with an AE that has already been reported.

Any laboratory result abnormality of CTCAE Grade 4 or 5 should be recorded as an SAE in the AE eCRF unless associated with an AE that has already been reported.

A clinical laboratory abnormality is considered clinically significant and should be documented as an AE if not refuted by a repeat test to confirm the abnormality and **any** one or more of the following conditions is met:

- Accompanied by clinical symptoms
- Requiring a change in concomitant therapy (eg, addition of, interruption of, discontinuation of, or any other change in a concomitant medication, therapy or treatment).
- The abnormality suggests a disease and/or organ toxicity
- The abnormality is of a degree that requires active management (eg, change of dose, discontinuation of study drug, more frequent follow-up assessments, further diagnostic investigation, etc.)

This applies to any protocol and non-protocol specified safety and efficacy laboratory result from tests performed after the first dose of study medication that falls outside the laboratory reference range and meets the clinical significance criteria.

This does not apply to any abnormal laboratory result that falls outside the laboratory reference range but that does not meet the clinical significance criteria (these will be analyzed and reported as laboratory abnormalities), those that are considered AEs of the type explicitly exempted by the protocol, or those which are a result of an AE that has already been reported.

For purposes of this study, laboratory tests showing a decreased level of FVIII activity should not be reported as adverse events unless there is an impact to clinical outcomes (eg, increased rate of bleeding, worsening of joint disease).

10.4.1.4 Pre-existing Conditions

A pre-existing condition is one that is present prior to administration of BMN 270. Such conditions should be recorded as medical history on the appropriate eCRF.

A pre-existing condition should be recorded as an AE or SAE during the study **only** if the frequency, intensity, or character of the condition worsens during the study period. It is important to convey the concept that a pre-existing condition has changed by including applicable language in the verbatim description of the event (eg, *more frequent* headaches).

10.4.1.5 General Physical Examination Findings

At screening, any clinically significant abnormality should be recorded as a pre-existing condition (refer to Section 10.4.1.4). During the study, any new clinically significant findings and/or abnormalities discovered on physical examination that meet the definition of an AE (or an SAE) must be recorded and documented as an AE or SAE on the AE eCRF.

10.4.1.6 Hospitalization, Prolonged Hospitalization, or Surgery

Any AE that results in hospitalization or prolonged hospitalization should be documented and reported as an SAE unless specifically instructed otherwise in this protocol (refer to Section 10.2).

There are some hospitalization scenarios that do not require reporting as an SAE when there is no occurrence of an AE. These scenarios include planned hospitalizations or prolonged hospitalizations to:

- Perform a protocol-mandated efficacy measurement
- Undergo a diagnostic or elective surgical procedure for a pre-existing medical condition that has not worsened
- Insert an in-dwelling IV catheter (such as a Port-a-Cath or other brand, if applicable) for administration of study drug or FVIII replacement therapy
- Receive scheduled therapy (study drug or otherwise) for the study indication

10.4.1.7 Deaths

All deaths that occur during the AE reporting period (refer to Section 10.3.1), regardless of attribution, will be recorded on the AE eCRF and expeditiously reported to the Sponsor as an SAE.

When recording a death, the event or condition that caused or contributed to the fatal outcome should be recorded as the single medical concept on the eCRF. If the cause of death

is unknown and cannot be ascertained at the time of reporting, record "Unexplained Death" or "Death of Unknown Cause" on the AE eCRF.

10.4.1.8 Pregnancy

Although not an AE per se, pregnancy in the partner of a subject taking trial medication should be reported expeditiously to the Sponsor to facilitate outcome monitoring by the Sponsor. Pregnancy in partner should be reported during the period up to 5 years after viral infusion.

Pregnancy in a partner should be reported within 24 hours of the site becoming aware of the pregnancy by entering the information on the Pregnancy eCRF and submitting to BPV within 24 hours of the site becoming aware of the event. The Investigator must make every effort to follow the subject's partner (with that partner's consent) through resolution of the pregnancy (delivery or termination) and to report the resolution on the Pregnancy Follow-up eCRF. In the event of pregnancy in the partner of a study subject, the Investigator should make every reasonable attempt to obtain the woman's consent for release of protected health information.

Abortion, whether therapeutic or spontaneous, should always be classified as an SAE (as the Sponsor considers these to be medically significant), recorded on the AE eCRF, and expeditiously reported to the Sponsor as an SAE.

10.5 Reporting Requirements

10.5.1 Expedited Reporting Requirements

All SAEs and EOSI that occur during the course of the AE Reporting Period (refer to Section 10.3.1), whether or not considered related to study drug, must be reported by entering the information in the AE eCRF and submitting to BPV within 24 hours of the site becoming aware of the event. Investigators should not wait to collect information that fully documents the event before notifying BPV of an SAE. BioMarin may be required to report certain SAEs to regulatory authorities within 7 calendar days of being notified about the event; therefore, it is important that Investigators submit any information requested by BioMarin as soon as it becomes available. IND safety reports will be submitted within 7 calendar days for unexpected fatal or life-threatening unexpected suspected adverse reactions (SUSARs) and within 15 calendar days for other non-life-threatening SUSARs.

The Sponsor is responsible for identifying, preparing and reporting all SUSARs to the relevant competent authorities, ethics committees and Investigators in accordance with the requirements identified in the Clinical Trials Regulations.

If the EDC is unavailable, all SAEs should be reported to BPV by completing the SAE Report Form and faxing or emailing the completed form to BPV within 24 hours of the site becoming aware of the event. Once the EDC is available, the information should be entered in the AE eCRF.

10.5.2 Institutional Review Board or Independent Ethics Committee Reporting Requirements

Reporting of SAEs to the Independent Ethics Committee (EC) or Institutional Review Board (IRB) will be done in compliance with the standard operating procedures and policies of the IRB/EC and with applicable regulatory requirements. Adequate documentation must be obtained by BioMarin showing that the IRB/EC was properly and promptly notified as required.

10.6 Follow-up of Subjects after Adverse Events

After the initial AE/SAE/EOSI report, the Investigator is required to proactively follow each subject at subsequent visits/contacts. All AEs/SAEs/EOSI will be followed until resolution, stabilization, the event is otherwise explained, or the subject is lost to follow-up. Resolution of AEs/SAEs/EOSI (with dates) should be documented on the AE eCRF and submitted to BioMarin Pharmacovigilance and in the subject's medical record to facilitate source data verification.

For some SAEs and EOSI, the Sponsor may follow up by telephone, fax, electronic mail, and/or a monitoring visit to obtain additional case details (eg, hospital discharge summary, consultant report, or autopsy report) deemed necessary to appropriately evaluate the SAE or EOSI report.

10.7 Post-Study Adverse Events

At the last scheduled visit, the Investigator should instruct each subject to report, to the Investigator and/or to BPV directly, any subsequent SAEs that the subject's personal physician(s) believes might be related to prior study drug.

The Investigator should notify the study Sponsor of any death or SAE occurring at any time after a subject has discontinued or terminated study participation, if the Investigator believes that the death or SAE may have been related to prior study drug. The Sponsor should also be notified if the Investigator should become aware of the development of cancer or of a congenital anomaly in a subsequently conceived offspring of a subject that participated in this study.

10.8 Urgent Safety Measures

The regulations governing clinical trials state that the Sponsor and Investigator are required to take appropriate urgent safety measures to protect subjects against any immediate hazards that may affect the safety of subjects, and that the appropriate regulatory bodies should be notified according to their respective regulations. According to the European Union (EU) Clinical Trial Directive 2001/20/EC, "…in the light of the circumstances, notably the occurrence of any new event relating to the conduct of the trial or the development of the investigational medicinal product where that new event is likely to affect the safety of the patients, the Sponsor and the Investigator shall take appropriate urgent safety measures to protect the patients against any immediate hazard. The Sponsor shall forthwith inform the competent authorities of those new events and the measures taken and shall ensure that the IRB/EC is notified at the same time."

The reporting period for these events which may require the implementation of urgent safety measures is the period from the time of signing of the ICF through the completion of the last study visit or at the Early Termination Visit (ETV). Investigators are required to report any events which may require the implementation of urgent safety measures to BioMarin within 24 hours of becoming aware of the event.

Examples of situations that may require urgent safety measures include discovery of the following:

- Lack of study scientific value, or detrimental study conduct or management
- Discovery that the quality or safety of the IP does not meet established safety requirements

10.9 BioMarin Pharmacovigilance Contact Information

Contact information for BioMarin Pharmacovigilance is as follows:

BioMarin Pharmaceutical Inc.

Address	105 Digital Drive Novato, CA 94949
Phone:	PI
Fax:	PI
E-mail:	drugsafety@bmrn.com

The Investigator is encouraged to discuss with the medical monitor any AEs for which the issue of seriousness is unclear or questioned. Contact information for the medical monitor is as follows:

Name:	PI , MD
Address:	BioMarin (UK) Ltd. 10 Bloomsbury Way London WC1A 2SL
Phone	PI (office) PI (mobile)
E-mail:	PI

11 APPROPRIATENESS OF MEASUREMENTS

The measures of efficacy to be used in this study are standard, ie, widely used and generally recognized as reliable, accurate, and relevant (able to discriminate between effective and ineffective agents). The measures of safety used in this study are routine clinical and laboratory procedures.

The chromogenic substrate FVIII assay and the one-stage clotting FVIII assay are both validated and utilize CE marked reagents.

12 STUDY PROCEDURES

12.1 Prestudy

An ICF must be signed and dated by the subject (or the subject's legally authorized representative), the Investigator or designee and witness (if required) before any study-related procedures are performed.

12.2 Screening Visit(s)

Screening assessments should be performed within 42 days of BMN 270 infusion, while baseline assessments will take place within 7 days prior to BMN 270 infusion (Day 1). Should the screening visit occur within 30 days of the drug infusion, physical examination, vital signs, blood chemistry, LTs, hematology, urine tests, and coagulation tests do not need to be repeated at Baseline.

During the first part of the Screening Period (Day -42 to Day -29), testing for AAV5 TAb titers using the ARUP screening assay may be performed, so that subjects can verify their AAV5 TAb status. Subjects who agree to participate in this activity (or their legally authorized representative) will be asked to sign a separate ICF documenting this decision. Subjects who do not agree will have the ARUP AAV5 TAb screening assay performed along with other assessments during the regular Screening period.

The following procedures will be performed during the Screening Period (Day -28 to Day -1):

- Demographics (age, sex, race, ethnicity)
- Full medical history, including hemophilia A history, Hepatitis B, Hepatitis C, and HIV. Subjects with a history of hepatitis B, hepatitis C, or HIV will be asked for information about the treatments received. Any prior pharmacokinetics information obtained while the subject was receiving prophylactic or episodic FVIII therapy prior to the study should also be collected.
- Complete Physical Examination
- Height and weight
- Vital Signs (systolic and diastolic blood pressure, heart rate, respiration rate, and temperature)
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information) for the previous 12 months
- Distribution of subject diaries and training in diary completion

- Electrocardiogram
- Liver ultrasound to screen for hepatocellular carcinoma and clinically significant liver disease (FibroScan can be performed additionally at the discretion of the Investigator)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay
    - hFVIII coagulation activity exploratory assay (collected but not tested prior to enrollment)
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
    - hFVIII total antibody assay (collected but not tested prior to enrollment)
    - hFVIII antigen assay (collected but not tested prior to enrollment)
- Blood sample for ARUP AAV5 TAb assay
    - Subjects who underwent the ARUP AAV5 TAb assay test earlier in the Screening period with the standalone consent do not need to repeat the test as part of regular Screening.
- Screen for Hepatitis B, Hepatitis C, and HIV if required (subjects with documented negative results 30 days prior to informed consent being obtained do not need to be retested)
    - Hepatitis B screening should include HBsAg, HBsAb, and HBcAb
- SARS-CoV-2 screening (local or central testing)
    - SARS-CoV-2 RT-PCR testing is required during Screening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing. A SARS-CoV-2 vaccine is recommended for subjects if indicated and if available (refer to Section 9.4.8 for vaccine guidance).
- Fasting FibroTest
    - Subjects will fast for at least 8 hours prior to sampling on the day of the FibroTest Screening visit. Fasting FibroTest results must be available prior to BMN 270 infusion.
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.4.1)

- Blood samples for Biomarker testing (may include HLA genotyping and FVIII genotyping status)

12.2.1 "Smart Rescreening" Visit

Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once.

If a patient has to be screened again because the original assessments have fallen out of the 28 + 14 day period allowed for Screening (refer to Section 12.2), then only the following assessments need to be performed (rather than the full list indicated in Section 12.2) for the patient to be successfully re-screened for the study:

- Vital signs
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- hFVIII Assays (only the hFVIII inhibitor level (Bethesda assay with Nijmegen modification))
- AAV5 TAb assay (ARUP)
- SARS-CoV-2 screening (local or central testing)
    - SARS-CoV-2 RT-PCR testing is required during smart rescreening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing. A SARS-CoV-2 vaccine is recommended for subjects if indicated and if available (refer to Section 9.4.8 for vaccine guidance).
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.4.1)

12.3 Baseline Visit

Baseline values will be recorded from 1 to 7 days prior to the infusion visit. The following procedures will be performed during the Baseline Period:

- Brief physical examination
- Vital signs

- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay
    - hFVIII coagulation activity exploratory assay
    - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
    - hFVIII total antibody assay
    - hFVIII antigen assay
- Von Willebrand Factor Antigen (VWF:Ag)
- Blood sample for AAV5 Total Antibody assay
    - Baseline sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
    - ABO blood typing assessment should be performed at Baseline, or at another regularly scheduled visit prior to the end of the subject's participation in the study.
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.4.1)
- PBMC collection for CTL baseline
- Blood sample for AAV5 TI assay
- PCR of vector DNA in blood, saliva, urine, semen, and stools
- Exploratory biomarker assessments
- Cytokine profiling
- Hypersensitivity blood assessments
- Haemo-QoL-A assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.4 Treatment Visit/BMN 270 Infusion Visit (Day 1)

There will be one infusion visit for each subject. Subjects will remain in the clinic for at least 24 hours for the BMN 270 Infusion Visit. The following procedures will be performed during the BMN 270 Infusion Visit:

- Brief physical examination (pre-infusion)
- Start prophylactic corticosteroids (pre-infusion)
- Assessment of Adverse Events and Concomitant Medications (pre-infusion)
- AAV5 TAb assay (ARUP) (sample collected pre-infusion for analysis)
- Blood sample for AAV5 TI assay (sample collected pre-infusion for analysis)
- Fasting blood sample for future exploratory analysis (sample collected pre-infusion)
- Fasting lipid panel (blood triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) (sample collected pre-infusion)
    - Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit.
- BMN 270 Infusion
- Complement panel and exploratory cytokine profiling
    - Complement panel should include C3, C3a, C4, Bb, and sC5b-9 and should be collected within 2 hours post-BMN 270 infusion.
- Vital Signs
    - Vital signs will be recorded prior to BMN 270 infusion and then every 15 minutes (± 5 minutes) during BMN 270 infusion. Following infusion, vital signs will be monitored every 1 hour (± 5 minutes) for at least 8 hours during the subject's stay in the clinic.
- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Collection of samples for PCR testing should occur between 2 and 24 hours after the BMN 270 infusion has been completed

In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine profiling, as well as possible additional exploratory testing) and samples for IgE and cytokine profiling (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine profiling . In-patient observation can

be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.

Subjects should also start prophylactic corticosteroids, prior to BMN 270 infusion, on Study Day 1 (refer to Table 9.1.6 for a possible prophylactic corticosteroid regimen, and to Section 9.4.8.2 further discussion). Clinical judgment, weighting the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses. Discussions between the Investigator and Medical Monitor are advised for any questions or concerns.

12.5 BMN 270 Infusion Follow-Up Visits

At applicable sites, certain study assessments may be performed by a mobile nursing (MN) professional at the patient's home or another suitable location, such as their school or office, to improve access and convenience for patients participating in the study. The Sponsor may select a healthcare company that will be responsible for providing MN services for participating sites (the MN vendor). The MN vendor is responsible for ensuring that all MN professionals are licensed, qualified, and in good standing, as per applicable regulations, and that appropriate background checks have been performed. If the investigator at a participating site determines that MN services are appropriate for a patient and the patient (or his legally authorized representative, if required) gives written informed consent to participate in MN visits, the MN network will communicate with the patient and the patient’s site. Potential MN visits are designated in the Schedules of Events.

12.5.1 Week 1

During Week 1, the subject will be assessed on Study Day 2, Study Day 4, and Study Day 8.

12.5.1.1 Week 1, Study Day 2

On Study Day 2, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.4.1)

- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Separate collection on Day 2 is not required if vector shedding samples were collected on Day 1 post-BMN 270 infusion; if collection on Day 2 is needed, it must occur within 24 hours of the time of the BMN 270 infusion.
- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection
- Complement panel
    - Complement panel should include C3, C3a, C4, Bb, and sC5b-9.
- Cytokine profiling

12.5.1.2 Week 1, Study Day 4

On Study Day 4, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.4.1)
- Complement panel
    - Complement panel should include C3, C3a, C4, Bb, and sC5b-9.
- PCR of vector DNA in blood, saliva, urine, semen, and stools

12.5.1.3 Week 1, Study Day 8

On Study Day 8, the following assessments will be performed:

- Brief physical examination
- Assessment of Adverse Events and Concomitant Medications
- Vital Signs
- Liver Tests (refer to Table 9.7.6.4.1)
- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay

- Blood sample for AAV5 TI assay
- Samples for FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level
    - hFVIII antigen assay
- Complement panel
    - Complement panel should include C3, C3a, C4, Bb, and sC5b-9.
- PCR of vector DNA in blood, saliva, urine, semen, and stools
- Cytokine profiling

12.5.2 Weeks 2-26

After Week 1 (Day 8), subjects will return to the study site once a week (± 48 hours) during Weeks 2-26.

12.5.2.1 Once per week (Weeks 2 through 26)

The following procedures will be performed once per week from Weeks 2 through 26:

- Brief physical examination (complete physical examination at Week 26)
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, physical examination will not be performed.
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
    - For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.
- Vital Signs
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, vital signs will not be performed.
- Liver Tests (refer to Table 9.7.6.4.1)
    - LT assessment may be checked more frequently when ALT values are > ULN or ≥ 1.5x baseline value or based upon discussion between the Medical Monitor and the Investigator.

- Samples for FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level
    - hFVIII antigen assay

12.5.2.2 Every Other Week (Weeks 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, and 26)

The following procedures will be performed every other week (at Weeks 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22, 24, and 26):

- Blood sample for AAV5 TAb Assay
    - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- PBMC collection

12.5.2.3 Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26

The following procedure will be performed at Weeks 2, 3, 4, 6, 8, 12, 16, 20, 24, and 26:

- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Collection to occur until at least 3 consecutive results below the limit of detection are obtained. Semen samples should continue to be collected and tested through Week 12, even if 3 consecutive results below the limit of detection in that compartment have been recorded prior to that time point.

12.5.2.4 Weeks 2, 4, 6, 8, 10, and 12

The following procedure will be performed at Weeks 2, 4, 6, 8, 10, and 12:

- Complement panel
    - Complement panel should include C3, C3a, C4, Bb, and sC5b-9.

12.5.2.5 Weeks 2, 4, 6, 8, 10, 12, 16, 20, 24, and 26

The following procedure will be performed at Weeks 2, 4, 6, 8, 10, 12, 16, 20, 24 and 26:

- hFVIII total antibody assay

12.5.2.6 Weeks 2, 4, 6, 8, 14, 20, and 26

The following procedure will be performed at Weeks 2, 4, 6, 8, 14, 20, and 26:

- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)

12.5.2.7 Weeks 2, 6, 10, 14, 18, 22, and 26

The following procedure will be performed at Weeks 2, 6, 10, 14, 18, 22, and 26:

- Exploratory biomarker assessments

12.5.2.8 Weeks 2, 6, 10, 14, 20, 24, and 26

The following procedure will be performed at Weeks 2, 6, 10, 14, 20, 24, and 26:

- Cytokine profiling

12.5.2.9 Weeks 4, 8, 12, 16, 20, and 26

The following procedures will be performed at Weeks 4, 8, 12, 16, 20, and 26:

- Weight
- VWF:Ag

12.5.2.10 Weeks 12 and 26

The following procedures will be performed at Weeks 12 and 26:

- Urine tests (to be performed locally)
- Haemo-QoL-A assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire

12.5.2.11 Week 16

The following procedure will be performed at Week 16:

- Testing for reactivation of hepatitis B and hepatitis C (only in subjects with evidence of prior exposure to hepatitis B and/or hepatitis C)
    - Subjects who receive reactive oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.6.

12.5.3 Post-Infusion Follow-Up – Weeks 27-52

During Weeks 27-36, subjects will return to the study site weekly (± 48 hours). During Weeks 37-52, subjects will return to the study site every 2 weeks (Week 38, 40, 42, 44, 46, 48, 50, and 52) (± 1 week). At these visits, the following procedures will be completed:

12.5.3.1 Every Visit

At every visit (Weeks 27-36, 38, 40, 42, 44, 46, 48, 50, and 52), the following procedures will be performed:

- Physical examination
    - Brief physical examination should be performed at all weeks except Week 52, when a complete physical examination should be performed
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, physical examination will not be performed.
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
    - For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.
- Vital Signs
    - For visits where a MN service is being used or a lab draw-only site visit is conducted, vital signs will not be performed.
- Liver Tests (refer to Table 9.7.6.4.1)
    - LT assessment may be checked more frequently when ALT values are > ULN or ≥ 1.5x baseline value or based upon discussion between the Medical Monitor and the Investigator.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
        - FVIII coagulation activity exploratory assay
        - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
        - FVIII antigen assay

12.5.3.2 Weeks 28, 30, 32, 34, 36, 40, 44, 48, and 52

At Weeks 28, 30, 32, 34, 36, 40, 44, 48, and 52, the following procedures will be performed:

- AAV5 TAb Assay
- AAV5 TI Assay

12.5.3.3 Weeks 28, 30, 32, 34, 36, 44, and 52

At Weeks 28, 30, 32, 34, 36, 44, and 52, the following procedure will be performed:

- PBMC collection

12.5.3.4 Weeks 32, 36, 44, and 52

At Weeks 32, 36, 44, and 52, the following procedures will be performed:

- Weight
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- FVIII antibody titer
- Cytokine profiling

12.5.3.5 Weeks 32, 36, 40, 44, 48, and 52

At Weeks 32, 36, 40, 44, 48, and 52, the following procedures will be performed:

- Exploratory biomarker assessments
- PCR of vector DNA in blood, saliva, urine, semen, and stools
    - Sample testing to occur until at least 3 consecutive sample below the limit of detection results have been obtained. Subjects who have not had 3 consecutive semen samples below the limit of detection by Week 52 should continue to have PCR testing of semen every 6 weeks until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).
    - Subjects considered to be treatment failures must continue to provide samples for PCR assessment at these timepoints until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints)

12.5.3.6 Week 36 and 52

At Weeks 36 and 52, the following procedures will be performed:

- Urine Tests (refer to Table 9.7.6.2.1)
- VWF:Ag

12.5.3.7 Week 52

At Week 52, the following procedures will be performed:

- Liver ultrasound
    - Additional liver ultrasounds may be performed prior to Week 52 at the discretion of the Investigator.
- Haemo-QoL-A assessment
- EQ-5D-5L
- HAL
- WPAI+CIQ:HS

12.6 Post-Infusion Follow-Up – Years 2-5

Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 and inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 52 of the study by attending only the Q12W and End of Year visits during Years 2-5.

Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from semen must still provide semen samples for assessment every 6 weeks during Years 2-5 until vector shedding has cleared (but do not have to perform other scheduled assessments at those study visits). Subjects who are not attending the Q6W visits during Years 2-5 may receive a scheduled monthly follow-up telephone call from the site to ask about adverse events, concomitant medications, and bleeding events/FVIII replacement usage.

During Years 2-5 of Post-Infusion Follow-up, the following procedures will be completed:

12.6.1 Years 2-5 – Every 6 Weeks (not required for treatment failure)

During Years 2-5, every 6 weeks (± 2 weeks), the following procedures will be performed:

- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
    - For visits where a MN service is being used, the service will contact the subject via e-mail or phone call to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use.

- Liver Tests (refer to Table 9.7.6.4.1)
    - LT assessment may be checked more frequently when ALT values are > ULN or ≥ 1.5x baseline value or based upon discussion between the Medical Monitor and the Investigator.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
        - If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.
- PCR of vector DNA in semen (if required)
    - Sample testing during Years 2-5 is not required if at least 3 consecutive samples are clear by the end of Year 1. Subjects who have not had 3 consecutive semen samples below the limit of detection by the end of Year 1 should continue to have PCR testing of semen every 6 weeks during Years 2-5 until 3 consecutive samples below the limit of detection are documented (or upon consultation between the Investigator and Medical Monitor).
        - Subjects who meet the definition of treatment failure and wish to follow an abbreviated schedule but who have not cleared vector shedding from semen must still provide semen samples for assessment every 6 weeks during Years 2-5 until vector shedding has cleared (in such a case, these subjects do not need to perform other assessments except for PCR sample delivery scheduled at these timepoints).

12.6.2 Years 2-5 – Every 12 Weeks and End of Year Visits (required for all subjects)

During Years 2-5, subjects will be asked to return to the study site for visits at the following study weeks (±2 weeks):

- Year 2 – Week 64, Week 76, Week 88, Week 104
- Year 3 – Week 116, Week 128, Week 140, Week 156
- Year 4 – Week 168, Week 180, Week 192, Week 208
- Year 5 – Week 220, Week 232, Week 244, Week 260

For each of these years, the last study visit listed (Week 104, Week 156, Week 208, and Week 260) will serve as an End of Year visit.

At the every 12 week and End of Year visits, the following procedures will be performed:

- Brief physical examination (at every 12 week visits) or complete physical examination (at End of Year visits; genitourinary examination may be deferred)
- Weight (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Assessment of Adverse Events and Concomitant Medications (including review of subject diary for bleeding and FVIII use)
- Liver Tests (refer to Table 9.7.6.4.1)
    - LT assessment may be checked more frequently when ALT values are > ULN or ≥ 1.5x baseline value or based upon discussion between the Medical Monitor and the Investigator.
- FVIII Assays
    - FVIII activity level (chromogenic substrate FVIII assay)
    - FVIII activity level (one-stage clotting FVIII assay)
    - FVIII coagulation activity exploratory assay
    - Bethesda assay (with Nijmegen modification) for FVIII inhibitor level
        - If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result.
- FVIII antigen assay
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1) (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Urine Tests (refer to Table 9.7.6.2.1) (at Week 76, Week 128, Week 180, Week 232, and End of Year visits only)
- Vital Signs
- AAV5 TAb Assay (at Week 64, Week 76, Week 88, Week 104, then at End of Year visit for Years 3-5)
- AAV5 TI Assay (at End of Year visit for Years 2-5 only)
- FVIII antibody titer (at Week 64, Week 76, Week 88, Week 104, then at End of Year visit for Years 3-5)
- Haemo-QoL-A assessment (at End of Year visits only)
- EQ-5D-5L (at End of Year visits only)
- HAL (at End of Year visits only)
- WPAI+CIQ:HS (at End of Year visits only)

- Exploratory biomarker assessments
- Cytokine profiling
- PBMC collection (at Week 64, Week 76, Week 88, Week 104, Week 128, Week 156, Week 180, Week 208, Week 232, and Week 260)
- VWF:Ag
- Liver ultrasound (at End of Year visits only)
    - Additional liver ultrasounds may be performed at interim time points (ie, between the End of Year visits) at the discretion of the Investigator.
- PCR of vector DNA in blood, saliva, urine, semen, and stools (if required)
    - Sample testing during Years 2-5 is not required if at least 3 consecutive samples are below the limit of detection during the Post-Infusion Follow-Up period in Weeks 1-52.

12.7 Early Termination Visit

The Early Termination visit will occur on the date the subject withdraws from the study, even if the date does not correspond to a protocol-specific visit.

If a subject leaves the study prior to the Week 260 visit, the subject will be asked to return to the study site and complete an Early Termination visit. At the Early Termination visit, as many of the following assessments as possible should be done:

- Complete physical examination
- Weight
- Assessment of Adverse Events and Concomitant Medications
- Documentation of bleeding episodes and FVIII usage (by either subject or clinical information)
- Vital signs
- Blood chemistry, hematology, and coagulation tests (refer to Table 9.7.6.2.1)
- Urine Tests (refer to Table 9.7.6.2.1)
- Liver Tests (refer to Table 9.7.6.4.1)
- Samples for hFVIII Assays
    - Baseline FVIII activity – chromogenic substrate FVIII assay
    - Baseline FVIII activity level – one-stage clotting FVIII assay
    - hFVIII coagulation activity exploratory assay

  - hFVIII inhibitors (Bethesda assay with Nijmegen modification)
  - hFVIII antigen assay
- hFVIII total antibody assay
- Exploratory biomarker assessments
- Blood sample for AAV5 Total Antibody assay
  - Sample will be tested with a AAV5 TAb post-dose immunogenicity monitoring assay
- Blood sample for AAV5 TI assay
- Cytokine profiling
- PBMC collection
- VWF:Ag
- Liver ultrasound
- Haemo-QoL-A assessment
- EQ-5D-5L
- Hemophilia Activities List (HAL)
- Work Productivity and Activity Impairment plus Classroom Impairment Questions: Hemophilia Specific (WPAI+CIQ:HS) questionnaire
- PCR of vector DNA in blood, saliva, urine, semen, and stool
  - Sample testing at the Early Termination Visit is not required if at least 3 consecutive samples are clear during the period of the subject's participation in the study.

12.8 End of Study

The study will end after the last subject yet to complete the last Long-Term Follow-Up visit (Week 260) does so, has transferred to another BMN 270 study, is withdrawn from the study, or discontinues from the study. BioMarin reserves the right to discontinue the study any time for clinical or administrative reasons and to discontinue participation of an individual Investigator or site for clinical or administrative reasons, including, but not limited to, poor enrollment or noncompliance with procedures of the protocol or GCP. In addition, the study may be terminated if, in the opinion of BioMarin, the safety of the study subjects may be compromised.

13 DATA QUALITY ASSURANCE

BioMarin personnel or designees will visit the study site prior to initiation of the study to review with the site personnel information about the IP, protocol and other regulatory document requirements, any applicable randomization procedures, source document requirements, CRFs, monitoring requirements, and procedures for reporting AEs, including SAEs.

At visits during and after the study, a CRA will monitor the site for compliance with regulatory documentation, with a focus on accurate and complete recording of data on CRFs from source documents, adherence to protocol, randomization (if applicable), SAE reporting and drug accountability records.

Sites will enter study data into eCRFs into the study EDC system. Data Quality Control will be performed by BioMarin Clinical Data Management or designee through implementation of quality control checks specified in the study data management plan and edit check specifications.

14 STATISTICAL METHODS AND DETERMINATION OF SAMPLE SIZE

14.1 Statistical and Analytical Plans

The statistical analysis plan (SAP) will provide additional details on the planned statistical analysis. Unless otherwise stated, all analyses will be performed using SAS.

14.1.1 Interim Analyses

No interim analysis is planned.

14.1.2 Procedures for Accounting for Missing, Unused and Spurious Data

Because the completeness of the data affects the integrity and accuracy of the final study analysis, every effort should be made to ensure complete, accurate, and timely data collection and, therefore, avoid missing data.

Sensitivity analyses will be conducted to assess the impact of missing data on the primary efficacy endpoint analysis. Additional details regarding the handling of missing data will be provided in the SAP.

14.2 Efficacy Analysis

The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using longitudinal methods, such as mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at ≥ 5 IU/dL at 26 weeks following study treatment. We can only assess the true steady state of FVIII produced from BMN 270 after a minimum of 72 hours (or 5x the known half-life of the FVIII replacement therapy administered) has elapsed since the last infusion of FVIII concentrates.

14.3 Safety Analysis

The Medical Dictionary for Regulatory Activities terminology (MedDRA) will be used by the Sponsor to assign system organ class and preferred term classification to events and diseases, based on the original terms entered on the eCRF.

All AEs will be coded using the current version of MedDRA. The incidence of AEs will be summarized by system organ class, preferred term, relationship to study drug, and severity. A by-subject listing will be provided for those subjects who experience a serious AE (SAE), including death, or experience an AE associated with early withdrawal from the study or study drug.

Clinical laboratory data will be summarized by the type of laboratory test. For each clinical laboratory test, descriptive statistics will be provided on Baseline as well as all subsequent visits. Descriptive statistics for physical examination results and vital signs will also be provided.

Analysis of neutralizing antibody response and other immunological parameters as well as vector shedding will be primarily descriptive and involve both inter-subject and intra-subject comparisons across doses.

Detailed statistical methods will be provided in the SAP.

14.4 Determination of Sample Size

The sample size is based upon clinical considerations and is sufficient to detect a strong clinical efficacy signal. Approximately 10 subjects may be dosed in the study.

14.5 Analysis Populations

The efficacy analysis set will be comprised of all subjects who have received the BMN 270 infusion.

The safety population will consist of all subjects who receive BMN 270 infusion during the study.

14.6 Changes in the Conduct of the Study or Planned Analyses

Only BioMarin may modify the protocol. Any change in study conduct considered necessary by the Investigator will be made only after consultation with BioMarin, who will then issue a formal protocol amendment to implement the change. The only exception is when an Investigator considers that a subject's safety is compromised without immediate action. In these circumstances, immediate approval of the chairman of the IRB/EC must be sought, and the Investigator should inform BioMarin and the full IRB/EC within 2 working days after the emergency occurs.

With the exception of minor administrative or typographical changes, the IRB/EC must review and approve all protocol amendments. Protocol amendments that have an impact on subject risk or the study objectives, or require revision of the ICF, must receive approval from the IRB/EC prior to their implementation.

When a protocol amendment substantially alters the study design or the potential risks or burden to subjects, the ICF will be amended and approved by BioMarin and the IRB/EC, and all active subjects (or their legally authorized representative, if required) must again provide informed consent.

15 DATA MONITORING COMMITTEE

The Data Monitoring Committee (DMC) will consist of experts in clinical trials, statistics, and hemophilia. The DMC will review available safety and efficacy data during the study on an ongoing basis to determine, based on emerging data and the benefit/risk profile, whether further enrollment should continue.

Duties of the DMC include:

- Conducting an ongoing review of individual subject safety and efficacy data during the study;
- Recommending whether to proceed with enrollment of subjects at a different gating schedules based on emerging data from 270-203 and the overall risk/benefit analysis of BMN 270;
- If applicable, considering whether to restrict dosing to subjects with anti-AAV5 titers below levels that appear to be causing clinically significant inhibition of transduction.
- Making other recommendations on the conduct and reporting of the trial based on their evaluation of clinical data including institution of any pause or stopping stages.

Guidance on proceeding from Cohort 1 to Cohort 2 and completion of each cohort will be based on DMC determination of safety and efficacy in treated subjects with the following triggers for pausing further enrollment:

- any related SAE;
- any related AE with a severity > Grade 3; or
- FVIII activity < 5% in at least 2/3 subjects after a minimum of 6 weeks post-BMN 270 infusion.

16 COSTS, COMPENSATION, AND SUBJECT INJURY

BioMarin will pay the full costs of the study-related tests, procedures, and treatments set forth in the protocol. In addition, after IRB/EC approval, BioMarin may reimburse the reasonable cost of travel for study-related visits in accordance with BioMarin's travel and reimbursement policy.

The Investigator should contact BioMarin immediately upon notification that a study subject has been injured by the study drug or by procedures performed as part of the study. Any subject who experiences a study-related injury should be instructed by the Investigator to seek immediate medical treatment at a pre-specified medical institution if possible, or at the closest medical treatment facility if necessary. The subject should be given the name of a person to contact to seek further information about, and assistance with, treatment for study-related injuries. BioMarin or the institution may pay for reasonable and necessary medical services to treat the injuries caused by the study drug or study procedures. In some jurisdictions, BioMarin is obligated by law to pay for study-related injuries. If this is the case, BioMarin will comply with the law. BioMarin will not pay for any hospitalizations, tests, or treatments for medical problems of any sort related solely to the study subject's primary disease or any concurrent disease that are unrelated to this study.

17 CASE REPORT FORMS AND SOURCE DOCUMENTS

Electronic case report forms will be provided for each subject. The Investigator must review and electronically sign the completed eCRF casebook to verify its accuracy.

eCRFs must be completed using a web–based application developed and validated. Study site personnel will be trained on the application and will enter the clinical data from source documentation. Unless explicitly allowed in the eCRF instructions, blank data fields are not acceptable.

In the event of an entry error, or if new information becomes available, the value will be corrected by deselecting the erroneous response and then selecting or entering the factual response. In compliance with ICH GCP Guidelines and 21 CFR Part 11, the system will require the personnel making the correction to enter a reason for changing the value. The documented audit trail will include the reason for the change, the original value, the new value, the time of the correction and the identity of the operator.

BioMarin's policy is that study data on the eCRFs must be verifiable to the source data, which necessitates access to all original recordings, laboratory reports, and subject records. In addition, all source data should be attributable (signed and dated). The Investigator must therefore agree to allow direct access to all source data. Subjects (or their legally authorized representative) must also allow access to their medical records, and subjects (or their legally authorized representative) will be informed of this and will confirm their agreement when giving informed consent. If direct source document verification of study data by the site monitor is prohibited by institutional policy or local law, then the Investigator must make available facilities and/or personnel to allow GCP-compliant source verification to occur. Examples of such methods include certified copies of records which have study data visible but sensitive information redacted, or other GCP-compliant means agreed between the Investigator and the Sponsor.

A site monitor designated by BioMarin will compare the eCRFs with the original source documents at the study site and evaluate them for completeness and accuracy before designating them as "Source Data Verified" (SDV). If an error is discovered at any time or a clarification is needed, the site monitor, or designee, will create an electronic query on the associated field. Site personnel will then answer the query by either correcting the data or responding to the query. The site monitor will then review the response and determine either to close the query or re–query the site if the response does not fully address the question. This process is repeated until all open queries have been answered and closed.

Before a subject's eCRF casebook can be locked, data fields must be source data verified and all queries closed. Refer to the Study Monitoring Plan for details on which fields must be source data verified. The Data Manager, or designee, will set the status of the forms, visits, and the entire casebook to Locked. The Investigator will then electronically sign the casebook, specifying that the information on the eCRFs is accurate and complete. Upon completion of the CSR, an electronic copy of each site's casebooks will be copied to a compact disk (CD) and sent to each site for retention with other study documents.

18 STUDY MONITORING AND AUDITING

Qualified individuals designated by BioMarin will monitor all aspects of the study according to GCP and standard operating procedures for compliance with applicable government regulations. The Investigator agrees to allow these monitors direct access to the clinical supplies, dispensing, and storage areas and to the clinical files, including original medical records, of the study subjects, and, if requested, agrees to assist the monitors. The Investigator and staff are responsible for being present or available for consultation during routinely scheduled site visits conducted by BioMarin or its designees. When in person site monitoring or source data verification cannot be conducted, remote site monitoring and/or source data verification will be conducted where allowed by country and local health authorities and ECs/IRBs.

Members of BioMarin's GCP Compliance Department or designees may conduct an audit of a clinical site at any time before, during, or after completion of the study. The Investigator will be informed if an audit is to take place and advised as to the scope of the audit. Representatives of the FDA or other Regulatory Agencies may also conduct an audit of the study. If informed of such an inspection, the Investigator should notify BioMarin immediately. The Investigator will ensure that the auditors have access to the clinical supplies, study site facilities, original source documentation, and all study files.

19 RETENTION OF RECORDS

The Investigator must retain all study records required by BioMarin and by the applicable regulations in a secure and safe facility. The Investigator must consult a BioMarin representative before disposal of any study records, and must notify BioMarin of any change in the location, disposition or custody of the study files. The Investigator /institution must take measures to prevent accidental or premature destruction of essential documents, that is, documents that individually and collectively permit evaluation of the conduct of a study and the quality of the data produced, including paper copies of study records (eg, subject charts) as well as any original source documents that are electronic as required by applicable regulatory requirements.

All study records must be retained for at least 2 years after the last approval of a marketing application in the US or an ICH region and until (1) there are no pending or contemplated marketing applications in the U.S. or an ICH region or (2) at least 2 years have elapsed since the formal discontinuation of clinical development of the IP. The Investigator /institution should retain subject identifiers for at least 15 years after the completion or discontinuation of the study. Subject files and other source data must be kept for the maximum period of time permitted by the hospital, institution or private practice, but not less than 15 years. These documents should be retained for a longer period, however, if required by the applicable regulatory requirements or by a BioMarin agreement. BioMarin must be notified and will assist with retention should Investigator /institution be unable to continue maintenance of subject files for the full 15 years. It is the responsibility of BioMarin to inform the Investigator /institution as to when these documents no longer need to be retained.

20 USE OF INFORMATION AND PUBLICATION

BioMarin recognizes the importance of communicating medical study data and therefore encourages the publication of these data in reputable, peer-reviewed scientific journals and at seminars or conferences. The details of the processes of producing and reviewing reports, manuscripts, and presentations based on the data from this study will be described in the Clinical Trial Agreement between BioMarin and the Investigator/Institution. Consideration for authorship of all publications will be based on compliance with the Uniform Requirements for Manuscripts Submitted to Biomedical Journals ("Uniform Requirements") of the International Committee of Medical Journal Editors (ICMJE) (http://www.icmje.org/ethical_1author.html) and good publication practices (GPP).

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22 INVESTIGATOR RESPONSIBILITIES

22.1 Conduct of Study and Protection of Human Subjects

In accordance with FDA Form 1572 and/or principles of ICH E6 GCP, the Investigator will ensure that:

- He or she will conduct the study in accordance with the relevant, current protocol and will only make changes in a protocol after notifying the sponsor, except when necessary to protect the safety, rights, or welfare of subjects.
- He or she will personally conduct or supervise the study.
- He or she will inform any potential subjects, or any persons used as controls, that the drugs are being used for investigational purposes, and he or she will ensure that the requirements relating to obtaining informed consent in 21 CFR Part 50 and/or ICH E6 Sections 2.9 and 4.8 are met. As well, he or she will ensure that IRB/EC review and approval in 21 CFR Part 56 and/or ICH E6 Section 2.6 are met.
- He or she will report to the sponsor adverse experiences that occur in the course of the investigation in accordance with 21 CFR 312.64 and/or ICH E6 Section 4.11.
- He or she has read and understands the information in the Investigator's Brochure, including potential risks and side effects of the drug.
- His or her staff and all persons who assist in the conduct of the study are informed about their obligations in meeting the above commitments
- Adequate and accurate records in accordance with 21 CFR 312.62 and/or ICH E6 Section 4.9 are kept, and those records are available for inspection in accordance with 21 CFR 312.68 and/or ICH E6 Section 4.9.7.
- The IRB/EC complies with the requirements of 21 CFR Part 56, ICH E6 Section 3.0, and other applicable regulations, and conducts initial and ongoing reviews and approvals of the study. He or she will also ensure that any change in research activity and all problems involving risks to human subjects or others are reported to the IRB/EC. Additionally, he or she will not make any changes in the research without IRB/EC approval, except where necessary to eliminate apparent immediate hazards to human subjects.
- He or she agrees to comply with all other requirements regarding the obligations of clinical Investigators and all other pertinent requirements in 21 CFR Part 312 and/or ICH E6.

23 SIGNATURE PAGE

Protocol Title: A Phase 1/2 Safety, Tolerability, and Efficacy Study of BMN 270, an Adeno-Associated Virus Vector–Mediated Gene Transfer of Human Factor VIII in Hemophilia A Patients with Residual FVIII Levels ≤ 1 IU/dL and Pre-existing Antibodies Against AAV5

Protocol Number: 270-203 Amendment 4

I have read the foregoing protocol and agree to conduct this study as outlined. I agree to conduct the study in compliance with all applicable regulations and guidelines, including E6R2 ICH, as stated in the protocol, and other information supplied to me.

Investigator Signature Date

Printed name: _____

Accepted for the Sponsor:

Medical Monitor Signature Date

Printed name: PI , MD, PI , Clinical Sciences_____

24 APPENDIX 1: SAMPSON'S ANAPHYLAXIS CRITERIA

According to the National Institute of Allergy and Infectious Disease/Food Allergy and Anaphylaxis Network (NIAID/FAAN) Second Symposium on the definition and management of anaphylaxis, anaphylaxis is highly likely when any one of the following 3 criteria are fulfilled:

1. Acute onset of an illness (minutes to several hours) with involvement of the skin, mucosal tissue, or both (eg, generalized hives, pruritus or flushing, swollen lips-tongue-uvula)

13. AND AT LEAST ONE OF THE FOLLOWING

a. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced peak expiratory flow [PEF], hypoxemia)

b. Reduced blood pressure (BP) or associated symptoms of end-organ dysfunction (eg, hypotonia [collapse], syncope, incontinence)

14. Two or more of the following that occur rapidly after exposure *to a likely allergen for that patient* (minutes to several hours):

a. Involvement of the skin-mucosal tissue (eg, generalized hives, itch-flush, swollen lips-tongue-uvula)

b. Respiratory compromise (eg, dyspnea, wheeze-bronchospasm, stridor, reduced PEF, hypoxemia)

c. Reduced BP or associated symptoms (eg, hypotonia [collapse], syncope, incontinence)

d. Persistent gastrointestinal symptoms (eg, crampy abdominal pain, vomiting)

15. Reduced BP after exposure to *known allergen for that patient* (minutes to several hours):

a. Infants and children: low systolic blood pressure (age specific) or greater than 30% decrease is systolic BP

b. Adults: systolic BP of less than 90 mmHg or greater than 30% decrease from that person's baseline.

Source: Sampson 2006.

25 PROTOCOL AMENDMENT TEXT REVISIONS

The following table summarizes the revisions made to Protocol Amendment 4 and relates the changes to the appropriate rationale (refer to pages 2-5). Added text is indicated by underlined font and deleted text is indicated by ~~strikethrough~~ font.

Section No./Title	Revision	Rationale
2/Synopsis (Study Sites)	Approximately 5-~~6~~14 sites globally	12
2/Synopsis (Study Rationale)	Treatment of severe HA presently mainly consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2-~~-~~3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half-~~-~~life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1-4 with prophylactic treatment in a recently published retrospective observational study and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with episodic (on-~~-~~demand-~~-~~only) therapy in the same studies, respectively. Continued repeated bleeding events compound debilitating multiple-joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death. … BMN 270 is being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-~~-~~201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. ~~Three~~An additional study has been undertaken at the 6E13 vg/kg dose (270-301 in subjects with severe HA), as well as a study in subjects receiving the 4E13 vg/kg dose (270-302). Four-year results from 270-~~-~~201 and one-year results from 270-301 have demonstrated that following gene transfer, mean and median FVIII activity levels above 15% (15 IU/dL), as measured by a chromogenic substrate assay, are achievable and sustained following a single infusion of 6E13 vg/kg of BMN 270, with administration of prophylactic corticosteroids and an acceptable safety profile ~~(Pasi, 2019). In addition, an interim analysis of clinical study 270-301, an ongoing phase 3 study designed to assess the efficacy and safety of BMN 270 at a dose of 6E13 vg/kg and administration of therapeutic corticosteroids (ie, in response to ALT elevations), demonstrated FVIII activity levels that were also well above 15%, albeit lower than what was observed for the 6E13 vg/kg cohort in 270-201 at 26 weeks (Pasi, 2019).~~. Preliminary results from optional liver biopsies (in subjects receiving lower doses of BMN 270 in 270-201) confirm dose-dependent pan-lobular and otherwise healthy liver transduction at 2.7-4.1 years. Subjects receiving 6E13 vg/kg in 270-201 received a different corticosteroid regimen than subjects in 270-301; in 270-201, subjects were scheduled to start corticosteroids by Week 3 (either before Week 3, in response to an alanine aminotransferase (ALT) elevation, or at Week 3 otherwise, per protocol), whereas in 270-301 subjects received corticosteroids only in response	6, 12

Section No./Title	Revision	Rationale
	to an ALT elevation. Possibly as a result of this difference, subjects receiving 6E13 vg/kg in 270-201 started corticosteroids at an earlier date in reference to the date of BMN 270 infusion and showed later onset of first ALT elevations when compared with subjects in 270-301. Recently published data from 270-201 and recent analysis of 270-301 data suggest that corticosteroids may have assisted in rescue or protection of FVIII activity levels during elevations of ALT and in resolution of elevated ALT levels in some subjects. Subjects enrolled and infused in 270-201 screened negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. Subjects enrolled and infused in 270-301 also screened negative for AAV5 antibodies. These pre-existing vector-specific antibodies are thought to arise from previous exposure to AAV, with seroprevalence varying across human populations and across studies. AAV5 has consistently been shown to have the lowest seroprevalence among the common AAV serotypes.	
2/Synopsis (Study Design and Plan)	This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to the AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects may be enrolled at 5-~~6~~14 sites in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. … Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from ~~a minimum of~~ 1-6 weeks post-infusion) reviewed. FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response, based on data from 270-201. ~~Up to~~Between 1 and 6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness). … Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed. Additional blood samples will be collected (tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine ~~bead array~~profiling, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine ~~bead array~~profiling (and possible additional exploratory testing) will be collected between 8-~~-~~24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine ~~bead array~~profiling. …	8, 12

Section No./Title	Revision	Rationale
	Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as ~~either~~ failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 ~~or~~and inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor or Sponsor-designated Data Monitor, follow an abbreviated visit schedule after Week 52 by attending only the Q12W and End of Year visits during Years 2-5.	
2/Synopsis (Inclusion Criteria)	Individuals eligible to participate in this study must meet all of the following criteria: 5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures. If the subject is unable to provide consent, a legally authorized representative may provide written informed consent.	11
2/Synopsis (Exclusion Criteria)	Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study: 1. Any evidence of active infection, including COVID-19, or any immunosuppressive disorder~~, including HIV infection~~ except for HIV infection. HIV-positive patients who meet all other eligibility criteria may be included if they have a CD4 count > 200/mm3 and an undetectable viral load (unquantifiable viral load as defined as less than the limit of quantification by the testing laboratory's assay is permitted) while receiving an antiretroviral therapy (ART) regimen that does not contain efavirenz or another potentially hepatotoxic ART. 7. Liver cirrhosis or other clinically significant liver disease of any etiology as assessed by liver ultrasound/FibroScan.	4, 12
2/Synopsis (Statistical Methods)	Approximately 10 subjects may be dosed in the study. The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using longitudinal methods, such as a mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at ≥ 5 IU/dL at 26 weeks following study treatment. ~~FVIII activity will be assessed weekly during the study period.~~	12
5.2/Ethical Conduct of Study	Specifically, this study is based on adequately performed laboratory and animal experimentation. The study will be conducted under a protocol reviewed and approved by an IRB/EC and will be conducted by scientifically and medically qualified persons. The benefits of the study are in proportion to the risks. The rights and welfare of the subjects will be respected and the investigators conducting the study do not find the hazards to outweigh the potential benefits. Each subject (or his legally authorized representative, if required) will provide written, informed consent before any study-related tests or evaluations are performed.	11
6/Investigators and Study Administrative Structure	During administration of informed consent, expectations regarding participation in the study should be made clear to subjects (and their legally authorized representatives, if required). Subjects who are not willing and/or are not able to comply with all aspects of the study should not be encouraged to participate.	11

Section No./Title	Revision	Rationale
7/Introduction	Hemophilia A (HA) is an X-linked recessive bleeding disorder that affects approximately 1 in 5,000 males (~~Nathwani, 1992~~Iorio 2019). … Gene therapy offers the potential of disease-modifying therapy by continuous endogenous production of human FVIII (hFVIII) following a single administration of vector. Hemophilia A is well-suited for this approach because its clinical manifestations are attributable to the lack of a single gene product (FVIII) that circulates in low amounts (100–200 ng/ml) in the plasma. Tightly regulated control of gene expression is not essential, and a modest increase in the level of FVIII (a plasma level of 2 ng/ml protein leads to a 1% expression) can ameliorate the severe phenotype (Srivastava~~, 2013~~ 2020); thus, the therapeutic goal for gene therapy is a modest increase in hFVIII.	12
7.2/Ongoing Clinical Studies	Ongoing clinical studies for BMN 270 include: • ~~270-203, a phase 1/2 study in patients with severe HA who have anti-AAV5 antibody titers~~ • 270-205, a phase 1/2 study in patients with severe HA who have active or prior FVIII inhibitors • 270-303, a phase 3 study in patients with severe HA who received BMN 270 at the 6E13 vg/kg dose level along with prophylactic corticosteroids A comprehensive review of safety, efficacy, and immunogenicity results ~~from 270-201 and 270-301~~ as of the latest data cut is contained in the IB supplied by BioMarin. Investigators are to review this document prior to initiating this study.	12
7.3/Study Rationale	Treatment of severe HA presently mainly consists of intravenous injection of plasma-derived or recombinant human FVIII protein (rhFVIII) concentrates, both as prophylaxis 2–3 times per week or at the time of a bleed, to prevent or control bleeding episodes, respectively. The half–life for FVIII (12 to 18 hours for most approved products) necessitates frequent infusions. While infusion of FVIII is a major advance in the treatment of HA, severe HA patients continue to have bleeding events on prophylactic therapy; median ABRs were 1–4 with prophylactic treatment in a recently published retrospective observational study (Berntorp~~,~~ 2017) and between 1-2 in 6 prospective FVIII interventional studies. Median ABRs were 4.5-18 and between 20-60 with episodic (on-demand-only) therapy in the same studies, respectively. Continued repeated bleeding events compound debilitating multiple–joint arthropathies as well as the risk of catastrophic events such as intracranial hemorrhage and death. … BMN 270 is being evaluated in clinical study 270-201, an ongoing first-in-human, phase 1/2 dose escalation study in subjects with severe HA designed to assess the safety and efficacy of BMN 270 at various dose levels (6E12 vg/kg, 2E13 vg/kg, 4E13 vg/kg, 6E13 vg/kg). Specifically, 270-201 explores the relationship of vector dose to the augmentation of residual FVIII activity and whether these levels are sufficient to alter the clinical phenotype. ~~Three~~An additional study has been undertaken at	6, 12

Section No./Title	Revision	Rationale
	the 6E13 vg/kg dose (270-301 in subjects with severe HA), as well as a study in subjects receiving the 4E13 vg/kg dose (270-302). Four-year results from 270-~~-~~201 and one-year results from 270-301 have demonstrated that following gene transfer, mean and median FVIII activity above 15% (15 IU/dL), as measured by a chromogenic substrate assay, are achievable and sustained following a single infusion of 6E13 vg/kg of BMN 270, with administration of prophylactic corticosteroids and an acceptable safety profile ~~(Pasi, 2019). In addition, an interim analysis of clinical study 270-301, an ongoing phase 3 study designed to assess the efficacy and safety of BMN 270 at a dose of 6E13 vg/kg and administration of on-demand corticosteroids (ie, in response to ALT elevations), demonstrated FVIII activity levels that were also well above 15% at 26 weeks, albeit lower than what was observed for the 6E13 vg/kg cohort in 270-201 (Pasi, 2019). For additional information on preliminary data in 270-201, refer to the current version of the Investigator's Brochure.~~. Preliminary results from optional liver biopsies (in subjects receiving lower doses of BMN 270 in 270-201) confirm dose-dependent pan-lobular and otherwise healthy liver transduction at 2.7-4.1 years. Subjects receiving 6E13 vg/kg in 270-201 received a different corticosteroid regimen than subjects in 270-301; in 270-201, subjects were scheduled to start corticosteroids by Week 3 (either before Week 3, in response to an alanine aminotransferase (ALT) elevation, or at Week 3 otherwise, per protocol), whereas in 270-301 subjects received corticosteroids only in response to an ALT elevation. Possibly as a result of this difference, subjects receiving 6E13 vg/kg in 270-201 started corticosteroids at an earlier date in reference to the date of BMN 270 infusion and showed later onset of first ALT elevations when compared with subjects in 270-301. Recently published data from 270-201 and recent analysis of 270-301 data suggest that corticosteroids may have assisted in rescue or protection of FVIII activity levels during elevations of ALT and in resolution of elevated ALT levels in some subjects. Subjects enrolled and infused in 270-201 were screened and negative for AAV5 antibodies, as the prevailing expectation has been that vector-specific antibodies could interfere with receptor binding and effectively neutralize efficient transduction of the target cell population. As this was the first FVIII gene therapy with AAV5 in humans, this criterion was adopted out of an abundance of caution. Subjects enrolled and infused in 270-301 also screened negative for AAV5 antibodies.	
7.4/Summary of Risks and Benefits	Transient ALT elevation (Grade 1 to 3 in severity) has been observed in most subjects administered BMN 270 shortly after dosing, with no evidence for major impacts upon liver function; no events meeting the Hy's Law criteria have been identified. Liver function has remained stable over time. ~~ALT elevations have been reported as events of interest in 13 subjects in 270-201, 1 subject in 270-302, and 91 subjects in 270-301. Although the majority of events have been Grade 1 or Grade 2 in severity, 11 subjects (1 in 270-302 and 10 in 270-301) had a reported Grade 3 ALT elevation. Only one serious event of ALT increased has~~	12

Section No./Title	Revision	Rationale
	~~been reported by investigators (in addition to one event that BioMarin conservatively assessed as serious based on the details of the case).~~Across the 6E13 vg/kg cohort of 270-201 and 270-301, subjects enrolled in 270-201 developed ALT elevation about 5.5 weeks later than subjects in 270-301, generally once the first course of corticosteroids was being tapered, and experienced lower peak elevations in ALT (75.7 U/L) than subjects in 270-301 (112.5 U/L). The difference in the ALT profile seen between the 6E13 vg/kg subjects in 270-201 and the subjects in 270-301 could be attributed to the difference in the protocol-specified corticosteroid regimens in place in those studies, including the early use of corticosteroids (ie, by Week 3 post-BMN 270 infusion). While the majority of ALT elevations responded rapidly to corticosteroids, given current interest in the field of AAV gene therapy for the use of non-steroidal approaches to managing or preventing ALT elevations, alternate non-steroidal systemic immunosuppressive agents have also been used to manage hepatic reactions where corticosteroids have proven to be ineffective or where high doses/and or prolonged exposure to corticosteroids have led to unwanted side effects. Overall, the literature and clinical experience with BMN 270 suggests that transient elevations in liver enzymes are expected following AAV-based gene therapy for the treatment for hemophilia A or B without any long-term concerns of hepatic injury (Manno~~,~~ 2006; Nathwani~~,~~ 2011; George~~,~~ 2016; Miesbach~~,~~ 2016; Pasi~~, 2017~~ 2020). Short-lived infusion reactions associated with one-time BMN 270 administration have included symptoms such as nausea, maculopapular rash, urticaria, diarrhea, watery eyes, rigors, chills, myalgia, fever, tachycardia and hypotension emerging within 24 hours of receiving BMN 270. Most infusion-related reactions were Grade 1 or Grade 2 in severity, and all events resolved, typically within 48 hours following medical management. ~~Three of these~~While some cases required temporary interruption of the infusion, followed by re-initiation at a slower rate~~.~~, all subjects completed their infusions. The reactions with onset during or within approximately 5 hours after the end of infusion responded to treatment with systemic antihistamines and/or corticosteroids, where administered. Infusion-related reactions were effectively mitigated by managing infusion rate and medications. No subjects have experienced thromboembolic events or developed inhibitors to FVIII following BMN 270 infusion. ~~At~~Subjects given the ~~highest~~6E13 vg/kg dose ~~tested~~ in 270-201 ~~(6E13 vg/kg), the majority of subjects~~and 270-301 have achieved mean FVIII ~~levels~~activity above ~~50~~40 IU/dL at 49-52 weeks post-infusion~~. Subjects in that cohort also reported markedly decreased bleeding compared with pre-study rates and the ability to discontinue prophylactic FVIII infusions. Subjects at all dose levels continue to be followed.~~ ~~In 270-301, an interim analysis has shown increased FVIII activity in the majority of subjects to mild HA or normal levels at 26 weeks post-infusion, also,~~ with markedly decreased bleeding compared with pre-study rates and the ability to discontinue	

Section No./Title	Revision	Rationale
	prophylactic FVIII infusions. ~~All subjects who will be included in the final analysis have been dosed with 6E13 vg/kg and~~Subjects at all dose levels continue to be followed.	
9.1/Overall Study Design and Plan	This is a Phase 1/2, single-arm, open-label study in severe HA patients (FVIII ≤ 1 IU/dL), with a history of at least 150 exposure days to FVIII concentrates/cryoprecipitates, with pre-existing antibodies to AAV5 vector capsid as measured by total antibody [TAb] assay. Approximately 10 subjects may be enrolled at 5-~~6~~14 sites globally in 2 cohorts (5 subjects in each cohort) and will receive a single dose of 6E13 vg/kg BMN 270 as an IV infusion. Subjects in Cohort 1 will have a Screening AAV5 TAb ≤ 500, while subjects in Cohort 2 will have a Screening AAV5 TAb > 500. Choosing a titer cutoff of 500 reflects the observed distribution of existing titer data with the BioMarin titer assay seen to date. … Subjects will be dosed sequentially in Cohort 1 or Cohort 2, based on the results of their Screening AAV5 TAb titers. Subjects in Cohort 2 can be dosed after a minimum of 3 and a maximum of 5 subjects have been dosed in Cohort 1 and had their safety and efficacy data (from ~~a minimum of~~ 1-6 weeks post-infusion) reviewed. Based on data from 270-201, FVIII activity ≥ 5% after six weeks post-infusion is expected to be the earliest differentiating time point for the majority of subjects dosed with 6E13 vg/kg who later achieved normal FVIII activity levels, compared with the one subject who had a slightly lower response. ~~Up to~~Between 1 and 6 weeks post-infusion will provide an appropriate timeframe to evaluate the development of any potential delayed hypersensitivity reaction (eg, serum sickness). … Dosing will be administered at a qualified infusion site, and subjects will be monitored for at least 24 hours post-infusion for any immediate hypersensitivity or adverse drug reaction. In case of suspected hypersensitivity or adverse drug reaction, safety assessments, in addition to physical examination and vital signs, will be performed. Additional blood samples will be collected (tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine ~~bead array~~profiling, as well as possible additional exploratory testing) within 1 hour of hypersensitivity reaction, and samples for IgE and cytokine ~~bead array~~profiling (and possible additional exploratory testing) will be collected between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine ~~bead array~~profiling. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. Any safety signal may trigger a review of the data and possible additional immunogenicity studies or other diagnostics deemed necessary to assess cellular immune responses using collected peripheral blood mononuclear cells (PBMCs). The duration of observation may be extended and additional blood samples collected at the discretion of the Investigator and Medical Monitor.	8, 12

Section No./Title	Revision	Rationale
	… In subjects who experience recurrent bleeding episodes, the Investigator and Medical Monitor will discuss whether to resume prior FVIII prophylaxis. Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as ~~either~~ failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 ~~or~~and inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor or Sponsor-designated Data Monitor, follow an abbreviated visit schedule after Week 52 by attending only the Q12W and End of Year visits during Years 2-5.	
Figure 9.1.1/270-203 Dosing Schedule	Figure 9.1.1 has been updated consistent with changes made elsewhere in the protocol.	12
Table 9.1.1 through Table 9.1.5 – Schedules of Activities	Tables 9.1.1 through 9.1.5 have been updated consistent with changes made elsewhere in the protocol and in the table footnotes (detailed below).	1, 2, 5, 7, 8, 9, 12
Table 9.1.1 – footnotes	[ec]All patients must have a liver ultrasound performed during the Screening period to screen for significant liver disease and hepatocellular carcinoma. A FibroScan may also be performed at the discretion of the Investigator. ~~g COVID 19~~[h]SARS-CoV-2 RT-PCR testing is required during Screening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing. A SARS-CoV-2 vaccine is recommended for subjects if indicated and if available. If a two-step SARS-CoV-2 vaccine is being used, sites should consider using the flexible re-screen option to allow subjects to receive both doses at least 14 days prior to treatment with BMN 270 (or at least 30 days prior to treatment with BMN 270 for any live-virus vaccines). It is preferable for SARS-CoV-2 vaccination to occur prior to BMN 270 infusion. Investigators should use clinical judgment, taking into consideration local factors, individual risk factors, and benefit/risk related to timing of vaccine administration. ~~j~~[ik] Blood samples will be collected from subjects to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for those evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for ~~TGA assay and~~ any ~~other~~ exploratory assessments) will be performed only as deemed necessary by the Sponsor. [kl]Complement panel should include C3, C3a, C4, Bb, and sC5b-9 and should be collected within 2 hours post-infusion. While exploratory samples for cytokine profiling will be collected at the time points indicated above, testing of these samples will be performed only as deemed necessary by the Sponsor.	1, 5, 7, 12

Section No./Title	Revision	Rationale
	[m] Smart rescreening should only be performed if a subject has been determined to be eligible for the study and is unable to complete the Baseline assessments and Infusion prior to the closing of the original Screening window (28 + 14 days). ~~COVID-19~~SARS-CoV-2 RT-PCR testing is required as part of smart rescreening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing. Subjects who undergo smart rescreening must complete the rescreening assessments and receive the infusion within 90 days of signing the original consent. Subjects who do not complete dosing within 90 days will be required to re-consent and undergo all screening procedures. Subjects may not undergo smart rescreening more than once. ~~[m]~~[o] With the exception of the collection of samples for PCR vector DNA analysis and the collection of the complement panel/exploratory cytokine profiling sample, assessments on the day of infusion must be performed prior to the infusion. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit. On the day of the BMN 270 Infusion, vital signs will be monitored prior to the infusion, during the infusion every 15 minutes (± 5 minutes), and following the infusion hourly (± 5 minutes) for at least 8 hours during the subject's stay in the clinic. Shedding samples for PCR of vector DNA analysis (blood, saliva, urine, semen, stool) should be collected between 2 and 24 hours after the infusion has been completed. ~~[e] Complement panel should include C3, C3a, C4, Bb, and sC5b-9 and should be collected within 2 hours post-infusion.~~ [pg]In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination, vital signs will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine ~~bead array~~profiling, as well as possible additional exploratory testing) and samples for IgE and cytokine ~~bead array~~profiling (and possible additional exploratory testing) between 8-24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine ~~bead array~~profiling. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions. In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor.	
Table 9.1.2 – footnotes	[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.4.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs assessment is weekly, but may be ~~monitored~~checked more ~~or less~~ frequently ~~(and in particular~~ when ALT values are > ULN or ≥ 1.5x baseline value~~)~~ or based ~~on~~upon discussion between the Medical Monitor and the Investigator ~~and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.~~. Subjects with ALT > ULN	6, 7, 11, 12

Section No./Title	Revision	Rationale
	or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: ~~(1)~~ the ALT has increased to above ULN~~; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level~~; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. ~~If FVIII levels and/or~~If ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample. [c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen assay. Chromogenic Nijmegen-Bethesda Assay for hFVIII inhibitor level will be tested as deemed necessary by the Sponsor. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72--hour interval without FVIII use is achieved; the 72--hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism. [e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for ~~TGA assay and~~ any ~~other~~ exploratory assessments) will be performed only as deemed necessary by the Sponsor. [g] The scheduled visits at Week 5, Week 7, Week 9, Week 11, Week 13, and Week 15 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject [or his legally authorized representative, if required] has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes,	

Section No./Title	Revision	Rationale
	and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits. [h] Complement panel should include C3, C3a, C4, Bb, and sC5b-9. While exploratory samples for cytokine profiling will be collected at the time points indicated above, testing of these samples will be performed only as deemed necessary by the Sponsor.	
Table 9.1.3 - footnotes	[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.4.1 for liver tests. Urine tests will be performed locally and centrally; all other laboratory assessments will be performed at the central laboratory. LTs assessment may be ~~monitored~~checked more ~~or less~~ frequently ~~(and in particular~~ when ALT values are > ULN or ≥ 1.5x baseline value~~)~~ or based ~~on~~upon discussion between the Medical Monitor and the Investigator ~~and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN~~. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: ~~(1)~~ the ALT has increased to above ULN~~; (2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level;~~;; or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample [c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII antigen assay. Chromogenic Nijmegen-Bethesda Assay for hFVIII inhibitor level will be tested as deemed necessary by the Sponsor. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism.	6, 7, 11, 12

Section No./Title	Revision	Rationale
	[e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for ~~TGA assay and~~ any ~~other~~ exploratory assessments) will be performed only as deemed necessary by the Sponsor. [f] The scheduled visits at Week 17, Week 19, Week 21, Week 23, Week 25, Week 27, Week 29, and Week 31 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject [or his legally authorized representative, if required] has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits. [g] While exploratory samples for cytokine profiling will be collected at the time points indicated above, testing of these samples will be performed only as deemed necessary by the Sponsor.	
Table 9.1.4 – footnotes	[b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.4.1 for liver tests. LTs assessment may be ~~monitored~~checked more ~~or less~~ frequently ~~(and in particular~~ when ALT values are > ULN or ≥ 1.5x baseline value~~)~~ or based ~~on~~upon discussion between the Medical Monitor and the Investigator ~~and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN.~~. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: ~~(1)~~ the ALT has increased to above ULN or increased by > 10 U/L from prior assessment; ~~(2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level;~~ or after a discussion between the Medical Monitor and the Investigator based on a review of subject data. If FVIII levels and/or ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. In case of a safety concern, additional unscheduled laboratory tests may be performed locally (at the Investigator's discretion) in order to facilitate timely and appropriate clinical management decisions; where possible, a	2, 6, 7, 11, 12

Section No./Title	Revision	Rationale
	matched sample for testing at the central laboratory should be collected (using the Unscheduled Visit lab kit) at the same time as the local unscheduled sample. [c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, coagulation exploratory assay, and hFVIII ~~protein assay.~~antigen assay. Chromogenic Nijmegen-Bethesda Assay for hFVIII inhibitor level will be tested as deemed necessary by the Sponsor. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72~~-~~-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism. [d] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for ~~TGA assay and~~ any ~~other~~ exploratory assessments) will be performed only as deemed necessary by the Sponsor. [e] The scheduled visits at Week 33, Week 35, Week 38, Week 42, Week 46, and Week 50 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject [or his legally authorized representative, if required] has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits. [f] While exploratory samples for cytokine profiling will be collected at the time points indicated above, testing of these samples will be performed only as deemed necessary by the Sponsor. [g] Additional liver ultrasounds may be performed prior to Week 52 at the discretion of the Investigator.	

Section No./Title	Revision	Rationale
Table 9.1.5 – footnotes	ETV: Early Termination Visit [a] ~~Brief~~Complete physical examination should be performed at ~~all~~the End of Year visits ~~during Years 2-5~~(genitourinary examination may be deferred); brief physical examination may be performed at Q12W visits. Weight should be recorded at the second Q12W visit each year and at every End of Year visit during Years 2~~-~~-5. [b] Refer to Table 9.7.6.2.1 for laboratory assessments to be included, and to Table 9.7.6.4.1 for liver tests. ~~LTs~~LT assessment may be ~~monitored~~checked more ~~or less~~ frequently ~~(and in particular~~ when ALT values are > ULN or ≥ 1.5x baseline value~~)~~ or based ~~on~~upon discussion between the Medical Monitor and the Investigator ~~and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN~~. Subjects with ALT > ULN or ≥ 1.5x baseline value during the study and either an alternative etiology considered or for further evaluation of the lab abnormality may undergo additional evaluations (eg, imaging studies including MRI or ultrasound, additional blood tests, and/or liver biopsy) at the discretion of the Investigator. Additional testing of FVIII and LTs during any study week may be performed if: (1) the ALT has increased to above ULN or increased by > 10 U/L from prior assessment; ~~(2) Increases in ALT values from prior assessment are accompanied by declines in FVIII activity level; or~~or (2) after a discussion between the Medical Monitor and the Investigator based on a review of subject data. ~~If FVIII levels and/or~~If ALT values are stable over the course of several weeks, assessment of FVIII activity and ALT levels may be adjusted based on discussion between the Medical Monitor and the Investigator. During Years 2-5 of the Post-Infusion Follow-Up period, urine tests and blood, chemistry, and coagulation tests should be performed at the second Q12W visit each year and at every End of Year visit. [c] Includes FVIII activity level (chromogenic substrate FVIII assay and one-stage clotting FVIII assay), Bethesda assay (with Nijmegen modification) for hFVIII inhibitor level, and coagulation exploratory assay~~, and~~. Chromogenic Nijmegen-Bethesda Assay for hFVIII ~~protein assay.~~inhibitor level will be tested as deemed necessary by the Sponsor. If a subject has used FVIII within 72 hours of a measurement day, all efforts should be made to obtain FVIII activity measurements when a 72~~-~~-hour interval without FVIII use is achieved; the 72-hour wash-out period is only intended for subjects who have achieved FVIII ≥ 5 IU/dL at 16 weeks after BMN 270 infusion and who have not resumed FVIII replacement therapy. If FVIII levels are elevated or stable over the course of several weeks, assessment of FVIII activity may be adjusted based on discussion between the Medical Monitor and the Investigator. A D-dimer may be considered under circumstances in which FVIII activity levels are above the normal physiological range and/or if there is a concern for a venous thromboembolism. If a subject tests positive in the Bethesda assay (with Nijmegen modification) during Years 2-5, a fresh sample should be collected within 4 weeks of the initial visit that had a positive result. [e] Blood samples will be collected to evaluate biochemical, molecular, cellular, and genetic/genomic aspects relevant to hemophilia A, coagulation, and/or AAV gene transfer, and to develop assays used for these evaluations. Subjects may choose	2, 6, 7, 8, 11, 12

Section No./Title	Revision	Rationale
	to opt out of the exploratory genetic/genomic research being done to study or try to discover genes that are not yet known to be associated with hemophilia A. While exploratory samples will be collected at the time points indicated above, testing of these samples (including those for ~~TGA assay and~~ any ~~other~~ exploratory assessments) will be performed only as deemed necessary by the Sponsor. [eh]The scheduled Q6W visits during Years 2-5 may be performed by a mobile nursing (MN) professional at the subject's home or another suitable location (if the subject [or his legally authorized representative, if required] has given written informed consent to participate in MN visits), or at the site as a shortened lab draw-only visit. The MN service or site will collect blood samples at these visits. For site lab draw-only visits, sites will contact the subject via phone call or e-mail to collect information regarding adverse events, changes to concomitant medications, bleeding episodes, and FVIII use. For MN visits, the service will collect this information. The physical examination and vital signs assessments listed in the Schedule of Activities will not be performed at these MN or lab draw-only visits. [i] While exploratory samples for cytokine profiling will be collected at the time points indicated above, testing of these samples will be performed only as deemed necessary by the Sponsor. [j] Additional liver ultrasounds may be performed at interim timepoints (ie, between the End of Year visits) at the discretion of the Investigator. [k] PBMC collection should occur at each Q12W visit during Year 2, then at every other Q12W visit during Years 3-5, as well as at all End of Year Visits for Years 2-5.	
Table 9.1.6 – footnotes	[b] Following initiation or completion of corticosteroid regimen, if a recurrence of ALT values ≥ ULN or ≥ 2x baseline value is reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor. Modification of the corticosteroid regimen may take into consideration timing of ALT elevation (prior to or after Week 52), as well as possible confounders for the ALT elevation~~, relationship between increases in ALT and FVIII activity, ALT/FVIII levels post-corticosteroid initiation,~~ and adverse events related to corticosteroid dosing. Guidance for tapering oral corticosteroid dosing can be found in Section 9.4.8.2, although a discussion between the PI and Medical Monitor should take place prior to tapering the corticosteroid dose.	6, 12
9.2/Discussion of Study Design	Study 270-203 is designed to be a Phase 1/2, single-arm, open-label study in hemophilia A patients with residual FVIII levels ≤ 1 IU/dL and pre-existing antibodies to the AAV5 vector capsid. Hemophilia A patients who provide written informed consent (or who have a legally authorized representative provide consent, if required) and meet the entry criteria will be eligible to enroll in the study.	11
9.3.1/Inclusion Criteria	Individuals eligible to participate in this study must meet all of the following criteria:	11

Section No./Title	Revision	Rationale
	5. Willing and able to provide written, signed informed consent after the nature of the study has been explained and prior to any study-related procedures. If the subject is unable to provide consent, a legally authorized representative may provide written informed consent.	
9.3.2/Exclusion Criteria	Individuals who meet any of the following exclusion criteria will not be eligible to participate in the study: 1. Any evidence of active infection, including COVID-19, or any immunosuppressive disorder~~, including HIV infection~~ except for HIV infection. HIV-positive patients who meet all other eligibility criteria may be included if they have a CD4 count > 200/mm3 and an undetectable viral load (unquantifiable viral load as defined as less than the limit of quantification by the testing laboratory's assay is permitted) while receiving an antiretroviral therapy (ART) regimen that does not contain efavirenz or another potentially hepatotoxic ART. 7. Liver cirrhosis or other clinically significant liver disease of any etiology as assessed by liver ultrasound/FibroScan.	4, 12
9.4.4/Directions for Administration	In case of hypersensitivity reactions, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine ~~bead array~~profiling, as well as possible additional exploratory testing) and samples for IgE and cytokine ~~bead array~~profiling (and possible additional exploratory testing) between 8-~~-~~24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine ~~bead array~~profiling.	12
9.4.8/Prior and Concomitant Medications	All prescription and over-the-counter medications (including dietary and herbal supplements) taken by a subject for 30 days before Screening will be recorded on the designated eCRF. For HIV-positive patients, prior to enrollment, the Medical Monitor will review the patient's ART regimen to assess that it does not contain efavirenz or another potentially hepatotoxic ART. The following medications are prohibited starting 30 days before Screening and through the end of the study, and the Sponsor must be notified if a subject receives any of these during the study: • Any investigational therapy other than BMN 270 … It is preferable for SARS-CoV-2 vaccination to occur prior to BMN 270 infusion. If a two-step SARS-CoV-2 vaccine is being used, sites should consider using the flexible re-screen option to allow subjects to receive both doses at least 14 days prior to treatment with BMN 270. If a live-virus SARS-CoV-2 vaccine is being used, subjects should wait at least 30 days after vaccination to receive a BMN 270 infusion. Investigators should use clinical judgment, taking into consideration local factors, individual risk factors, and the benefit/risk related to timing of vaccine administration. Administration of SARS-CoV-2 vaccine after BMN 270 infusion may occur after consultation between Investigator and Medical Monitor.	4, 5, 12

Section No./Title	Revision	Rationale
9.4.8.2.1/Prophylactic Corticosteroids	All subjects will be started on prophylactic corticosteroids starting on the day of infusion (Day 1). Table 9.1.6 provides an example of ~~a possible~~ the recommended prophylactic corticosteroid course, including taper and post-corticosteroid additional monitoring of FVIII activity, LTs, and hepatitis B/hepatitis C reactivation. Clinical judgment, ~~weighting~~weighing the potential risks and benefits of corticosteroid treatment, should always be exercised when considering adjustment of corticosteroid doses. Discussions between the Investigator and Medical Monitor are advised for any questions or concerns.	12
9.4.8.2.2/Reactive Corticosteroids	Following initiation or completion of the prophylactic corticosteroid regimen, if ALT levels become increased (eg, > ULN or ≥ 1.5x baseline value) and alternative etiologies have been ruled out, prompt institution of ~~therapeutic or on-demand~~newly administrated or an increased dose of reactive (ie, started in response to an ALT elevation) oral corticosteroids (prednisone or converted equivalent) should be considered after consultation with the Medical Monitor (refer to Table 9.~~7.6.3~~4.8.2.2.1). • Whenever possible, a confirmatory lab draw for ALT should be performed within 72 hours, along with FVIII activity, prior to initiating reactive oral corticosteroids. • ~~Corticosteroids may be delayed~~Newly administered corticosteroids or dose increases are not indicated if elevations in ALT are clearly not related to BMN 270 (eg, elevated ~~in~~ ALT with concurrent increase in CPK due to intensive exercise) although this should be discussed with the Medical Monitor (in particular, for elevations occurring at least 52 weeks after the BMN 270 infusion). • Alternative immunosuppressive agents may also be considered for use on a case-by-case basis and following consultation with the Medical Monitor (eg, if prolonged corticosteroid use is contraindicated). ~~Therapeutic~~Unless otherwise indicated, reactive corticosteroid treatment should be initiated at a dose of 60 mg/day. ~~At minimum,~~If the ~~recommended duration~~ ALT level remains stable or declines after 2 weeks, consider gradual taper of ~~therapeutic~~ corticosteroids ~~is 60 mg/day for 3 weeks,~~ : 40 mg/day for 4 weeks, ~~and~~ 30 mg/day for ~~4 weeks, followed by a gradual taper thereafter.~~1 week, 20 mg/day for 1 week, and 10 mg/day for 1 week. Should a scenario arise in which a deviation from the minimum recommended dose and/or duration of ~~therapeutic~~reactive corticosteroids may be clinically indicated, a discussion should take place between the Investigator and Medical Monitor regarding corticosteroid dose adjustments. ~~Tapering~~Management of ~~corticosteroid dosages~~ALT elevations with reactive corticosteroids, including tapering of doses and managing worsening and/or recurrent ALT elevations, should be guided by the following (Table 9.4.8.2.2.1): … When ruling out alternative viral or autoimmune hepatitis as part of the elevated ALT workup, the following tests should be performed (Table 9.4.8.2.2.2):	6, 12

Section No./Title	Revision	Rationale
	… After discontinuation of reactive oral corticosteroids, labs for ALT and FVIII levels will be measured once a week for 4 weeks to ensure stability in values. Following ~~initiation or~~ completion of ~~therapeutic~~ oral corticosteroids, if increased ALT levels (eg, > ULN or ≥1.5x baseline value) are reported, corticosteroid management decisions will be based on discussions between the Investigator and Medical Monitor. Modification of the corticosteroid regimen may take into consideration possible confounders for the ALT elevation and impact on FVIII expression.	
Table 9.4.8.2.2.1/ Management of ALT Elevations with Corticosteroids	Table 9.4.8.2.2.1 has been relocated from Section 9.7.6.3, and updated consistent with changes made elsewhere in the protocol.	6, 12
Table 9.4.8.2.2.2/ Viral and Autoimmune Hepatitis Testing	Table 9.4.8.2.2.2 has been relocated from Section 9.7.6.3.	6
9.4.8.3/Monitoring of HIV-Positive Subjects	HIV-positive subjects may be enrolled in 270-203 if the subject is well controlled on an ART regimen that does not contain efavirenz or another potentially hepatotoxic ART, has a CD4 count > 200/mm^3, and has an undetectable viral load (unquantifiable viral load as defined as less than the limit of quantification by the testing laboratory's assay is permitted). HIV-positive subjects were initially included in prior BMN 270 studies. However, after an HIV-positive subject in 270-302 developed markedly elevated liver enzyme levels after receiving 4E13 vg/kg of BMN 270, out of an abundance of caution for the long-term liver health of HIV-positive patients, further enrollment of HIV-positive subjects was suspended in 270-301 (Protocol Amendment 3) and 270-302 (Protocol Amendment 3). The subject in 270-302 referenced above was receiving efavirenz and lamivudine as part of his ART regimen. Following discussion with a liver advisory board and review of the accumulated 270-301 data, efavirenz and not lamivudine has been implicated as the most likely medication that interacted with BMN 270 and contributed to the 270-302 subject's elevated liver enzyme levels. Due to its hepatotoxicity, efavirenz is a prohibited medication in all BMN 270 studies. The two HIV-positive subjects on stable, non-efavirenz-containing ART regimens who were enrolled in and dosed in 270-301 study prior to Amendment 3 have been monitored closely. Following BMN 270 infusion, these subjects continued their ART as prescribed and followed routine monitoring of CD4 count and viral load. Results from 270-301 show similar safety results for	4

Section No./Title	Revision	Rationale
	the two HIV-positive subjects compared to those who are HIV-negative. The Sponsor believes that HIV infection, in and of itself, is not a contraindication to receive BMN 270 and has therefore removed the exclusion of HIV-positive subjects. Subjects should continue ART as prescribed and follow routine monitoring of CD4 count and viral load (US Dept Health Human Services 2019). Investigators will continue to monitor HIV-positive subjects per routine standard of care.	
9.7.2.1/FVIII Activity	~~In the event of an FVIII activity level decline during the study:~~ ~~• If FVIII activity has declined at least 20% from the peak but less than 35% and has declined for at least 2 consecutive assessments, FVIII activity and LTs should be repeated every 7 days until FVIII activity is stable or increasing~~ ~~• If FVIII activity has declined >35% from the peak and has declined for at least 2 consecutive assessments, FVIII activity and LTs should be repeated every 72 hours until FVIII activity is stable or increasing.~~ Note that fluctuations in FVIII activity after gene therapy are common, and ~~if no clear trend indicating a decline in~~ more frequent monitoring of FVIII activity levels is ~~observed, then this additional testing may be deferred (~~not needed in the absence of a concurrent or recent ALT elevation or upon consultation between the Investigator and the Medical Monitor~~) until either a more clear trend of decline has been demonstrated or until the FVIII activity levels stabilize or increase.~~. Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as ~~either~~ failure to achieve FVIII activity ≥ 5 IU/dL by either Week 26 or Week 52 ~~or~~and inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 52, as applicable and at the discretion of the Investigator.	8, 12
9.7.2.2/FVIII Replacement Therapy/ Bleeding Episodes	During the study, subjects will be asked at each study visit to report the use of factor replacement therapy and the number of bleeding episodes since the previous visit. This information will be captured on the subject's diary or other subject records. Subjects will be encouraged to discuss any bleeding episodes with the Investigator and attempt to objectively assess any reported bleeds through use of ultrasound or non-invasive imaging.	12
9.7.5/Exploratory Assessments	A cytokine ~~bead array assay~~profiling assessment will be performed at Baseline and then at the timepoints listed in the Schedule of Activities.	12

Section No./Title	Revision	Rationale
9.7.6.2/Clinical Laboratory Assessments	In addition to scheduled clinical laboratory assessments, a fasting blood lipid panel (including triglycerides, total cholesterol, HDL cholesterol, and LDL cholesterol) ~~and fasting FibroTest~~ will be assessed at the BMN 270 infusion visit. Subjects will fast for at least 8 hours prior to pre-infusion laboratory sampling on the day of the infusion visit. In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine ~~bead array~~profiling, as well as possible additional exploratory testing) and samples for IgE and cytokine ~~bead array~~profiling (and possible additional exploratory testing) between 8-24 hours after the reaction. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine ~~bead array~~profiling. Exploratory biomarker samples at baseline and at post-infusion study visits may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes. At applicable sites, certain study assessments may be performed by a mobile nursing (MN) professional at the patient's home or another suitable location, such as their school or office, to improve access and convenience for patients participating in the study. The Sponsor may select a healthcare company that will be responsible for providing MN services for participating sites (the MN vendor). The MN vendor is responsible for ensuring that all MN professionals are licensed, qualified, and in good standing, as per applicable regulations, and that appropriate background checks have been performed. If the investigator at a participating site determines that MN services are appropriate for a patient and the patient (or his legally authorized representative, if required) gives written informed consent to participate in MN visits, the MN network will communicate with the patient and the patient's site. Potential MN visits are designated in the Schedules of Events.	1, 11, 12
9.7.6.3/Malignancies	A liver ultrasound (and FibroScan, at the discretion of the Investigator) will be performed at Screening to screen for HCC. Thereafter, liver ultrasounds will be performed annually at each End of Year visit starting at Year 1 (Week 52) through the end of the study to screen for HCC. Additional liver ultrasounds may be performed prior to Week 52 and/or between the End of Year visits at the discretion of the Investigator. Any development of a malignancy (except non-melanoma skin cancers) during the course of the study will be considered an EOSI (refer to Section 10.2.1) and is subject to expedited reporting. In addition, it is recommended that genomic analyses be performed on any malignancy (except non-melanoma skin cancers) diagnosed during the course of the study. The study site will coordinate sending samples from the malignancy for genomic analyses, if available.	1, 2, 3

Section No./Title	Revision	Rationale
9.7.6.4/Liver and Hepatitis Testing	Subjects with a previous history of hepatitis B or hepatitis C who receive ~~therapeutic~~ oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.6. A liver ultrasound~~/FibroScan~~, fasting FibroTest, and liver tests (LTs) during Screening will ~~identify any~~be performed to assess for clinically significant ~~hepatic dysfunction~~liver disease and HCC. A FibroScan may also be performed during Screening at the discretion of the Investigator. Fasting FibroTest results must be available prior to BMN 270 infusion. … Elevated ALT levels should be evaluated according to ~~the following~~ plan outlined in Table 9.4.8.2.2.1 (note that these evaluations may indicate additional testing of LTs and FVIII levels at unscheduled visits; these unscheduled laboratory tests may be completed by a mobile nursing professional at sites where the use of MN services has been approved).	1, 6, 12
9.7.6.5/HIV Testing	HIV testing will be performed at Screening. Subjects with documented negative results within the last 30 days prior to screening do not need to be retested. Refer to Section 9.4.8.3 for guidance on monitoring of HIV-positive subjects.	4
9.7.6.6/Vital Signs, Physical Exams, and Other Safety	A complete physical examination should be performed at Screening, Week 26, Week 52, and at the End of Year visits. A complete physical examination will include general appearance (head, eyes, ears, nose, and throat), cardiovascular, dermatologic, lymphatic, respiratory, gastrointestinal, genitourinary, musculoskeletal, and neurologic systems. The genitourinary examination may be deferred for visits after Year 1 unless the subject has genitourinary-related complaints.	12
10.2.1/EOSI	The following EOSI need to be reported to the Sponsor within 24 hours of site awareness, irrespective of seriousness, severity or causality: • Any new diagnosis of malignancy (except non-melanoma skin cancer)	3
10.8/Urgent Safety Measures	The reporting period for these events which may require the implementation of urgent safety measures is the period from the time of signing of the ICF through the completion of the last study visit or at the Early Termination Visit (ETV). Investigators are required to report any events which may require the implementation of urgent safety measures to BioMarin within 24 hours of becoming aware of the event.	12
10.9/Contact Information	Contact information for the medical monitor is as follows: ~~Nina Mitchell, MA MB BChir MSc~~Thomas Machnig, MD	10

Section No./Title	Revision	Rationale
	+44 (0) 207 420 ~~3414~~3405 (office) +~~44~~49 (0) ~~7761 659 535~~1522/1825323 (mobile) ~~nina.mitchell~~thomas.machnig@bmrn.com	
12.1/Prestudy	An ICF must be signed and dated by the subject~~,~~ (or the subject's legally authorized representative), the Investigator or designee and witness (if required) before any study-related procedures are performed.	11
12.2/Screening	During the first part of the Screening Period (Day -42 to Day -29), testing for AAV5 TAb titers using the ARUP screening assay may be performed, so that subjects can verify their AAV5 TAb status. Subjects who agree to participate in this activity (or their legally authorized representative) will be asked to sign a separate ICF documenting this decision. Subjects who do not agree will have the ARUP AAV5 TAb screening assay performed along with other assessments during the regular Screening period. The following procedures will be performed during the Screening Period (Day -28 to Day ~~–~~1): • Full medical history, including hemophilia A history, Hepatitis B, Hepatitis C, and HIV. Subjects with a history of hepatitis B ~~or~~, hepatitis C, or HIV will be asked for information about the treatments received. Any prior pharmacokinetics information obtained while the subject was receiving prophylactic or ~~on-demand~~episodic FVIII therapy prior to the study should also be collected. • ~~Liver Ultrasound/FibroScan~~ • Liver ultrasound to screen for hepatocellular carcinoma and clinically significant liver disease (FibroScan can be performed additionally at the discretion of the Investigator) • ~~Screen for COVID-19~~SARS-CoV-2 screening (local or central testing) o ~~COVID-19~~SARS-CoV-2 RT-PCR testing is required during Screening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing. A SARS-CoV-2 vaccine is recommended for subjects if indicated and if available (refer to Section 9.4.8 for vaccine guidance). • Fasting FibroTest o Subjects will fast for at least 8 hours prior to sampling on the day of the FibroTest Screening visit. Fasting FibroTest results must be available prior to BMN 270 infusion.	1, 4, 5, 6, 11, 12

Section No./Title	Revision	Rationale
12.2.1/Smart Rescreening	If a patient has to be screened again because the original assessments have fallen out of the 28 + 14 day period allowed for Screening (refer to Section 12.2), then only the following assessments need to be performed (rather than the full list indicated in Section 12.2) for the patient to be successfully re-screened for the study: • ~~Screen for COVID-19~~SARS-CoV-2 screening (local or central testing) o ~~COVID-19~~SARS-CoV-2 RT-PCR testing is required during smart rescreening, and all subjects must have at least one negative test result prior to dosing. If the test is performed locally an additional 2nd test is recommended, but negative results from the 2nd test must be received prior to dosing. A SARS-CoV-2 vaccine is recommended for subjects if indicated and if available (refer to Section 9.4.8 for vaccine guidance)	5
12.3/Baseline	The following procedures will be performed during the Baseline Period: • ~~Thrombin Generation Assay~~ • Cytokine ~~bead array assay~~profiling	7, 12
12.4/Infusion Visit	The following procedures will be performed during the BMN 270 Infusion Visit: • Brief physical examination (pre-infusion) • Assessment of Adverse Events and Concomitant Medications (pre-infusion) • ~~Fasting FibroTest~~ • Complement panel and exploratory cytokine profiling o Complement panel should include C3, C3a, C4, Bb, and sC5b-9 and should be collected within 2 hours post-BMN 270 infusion. In case of hypersensitivity or adverse drug reaction, safety assessment, including physical examination and vital signs, will be done and additional blood samples will be collected within 1 hour of the hypersensitivity reaction (eg, tryptase, C3, C3a, C4, Bb, sC5b-9, and cytokine ~~bead array~~profiling, as well as possible additional exploratory testing) and samples for IgE and cytokine ~~bead array~~profiling (and possible additional exploratory testing) between 8–24 hours after the reaction, if possible. In addition, a blood sample should be taken 1 week after the hypersensitivity reaction for assessment of the cytokine ~~bead array~~profiling . In-patient observation can be extended and additional blood samples can be collected if deemed necessary at the discretion of the Investigator and Medical Monitor. Exploratory biomarker samples at baseline and at post-infusion study visits	1, 12

Section No./Title	Revision	Rationale
	may also be used to assess changes in these biomarkers to better elucidate the mechanisms of infusion-related hypersensitivity reactions" as described in the rationale of changes.	
12.5/Infusion Follow-Up Visits	If the investigator at a participating site determines that MN services are appropriate for a patient and the patient (or his legally authorized representative, if required) gives written informed consent to participate in MN visits, the MN network will communicate with the patient and the patient's site.	11
12.5.1.1/Day 2	On Study Day 2, the following assessments will be performed: • Cytokine ~~bead array assay~~profiling	12
12.5.1.3/Day 8	On Study Day 8, the following assessments will be performed: • Cytokine ~~bead array assay~~profiling	12
12.5.2.1/Once per Week, Weeks 2-26	The following procedures will be performed once per week from Weeks 1 through 26: • Liver Tests (refer to Table 9.7.6.~~3~~4.1) o ~~LTs~~LT assessment may be ~~monitored~~checked more ~~or less~~ frequently ~~(and in particular~~ when ALT values are > ULN or ≥ 1.5x baseline value~~)~~ or based ~~on~~upon discussion between the Medical Monitor and the Investigator ~~and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN~~.	6
12.5.2.8/Weeks 2, 6, 10, 14, 20, 24, 26	The following procedure will be performed at Weeks 2, 6, 10, 14, 20, 24, and 26: • Cytokine ~~bead array assay~~profiling	12
12.5.2.11/Week 16	The following procedure will be performed at Week 16: • Testing for reactivation of hepatitis B and hepatitis C (only in subjects with evidence of prior exposure to hepatitis B and/or hepatitis C) o Subjects who receive ~~therapeutic~~reactive oral corticosteroids prior to Week 16 do not need to complete the Week 16 reactivation assessment; instead, they will be tested for hepatitis B and hepatitis C reactivation at the time points listed in Table 9.1.6.	6
~~12.5.2.12/Weeks 20, 24, 26~~	~~The following procedure will be performed at Weeks 20, 24, and 26:~~ ~~• Thrombin Generation Assay~~	7

Section No./Title	Revision	Rationale
12.5.3.1/Every Visit Weeks 27-52	At every visit (Weeks 27-36, 38, 40, 42, 44, 46, 48, 50, and 52), the following procedures will be performed: • Liver Tests (refer to Table 9.7.6.~~3~~4.1) o ~~LTs~~LT assessment may be ~~monitored~~checked more ~~or less~~ frequently ~~(and in particular~~ when ALT values are > ULN or ≥ 1.5x baseline value~~)~~ or based ~~on~~upon discussion between the Medical Monitor and the Investigator~~ and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN~~.	6
12.5.3.4/Weeks 32, 36, 44, 52	At Weeks 32, 36, 44, and 52, the following procedures will be performed: • Cytokine ~~bead array assay~~profiling	12
12.5.3.5/Weeks 32, 36, 40, 44, 48, 52	At Weeks 32, 36, 40, 44, 48, and 52, the following procedures will be performed: • ~~TGA Assay~~	7
12.5.3.7/Week 52	At Week 52, the following procedures will be performed: • Liver ultrasound o Additional liver ultrasounds may be performed prior to Week 52 at the discretion of the Investigator.	2
12.6/Years 2-5	Subjects who do not respond to BMN 270 treatment (ie, treatment failure, manifesting as ~~either~~ failure to achieve FVIII activity ≥ 5 IU/dL by Week 52 ~~or~~and inability to maintain independence from prophylactic FVIII replacement therapy due to joint bleeding episodes, in the judgment of the Investigator) may, at the Investigator's discretion and after discussion with the Medical Monitor, follow an abbreviated visit schedule after Week 52 of the study by attending only the Q12W and End of Year visits during Years 2-5.	8
12.6.1/Years 2-5, Every 6 Weeks	During Years 2-5, every 6 weeks (± 2 weeks), the following procedures will be performed: • Liver Tests (refer to Table 9.7.6.~~3~~4.1) o ~~LTs~~LT assessment may be ~~monitored~~checked more ~~or less~~ frequently ~~(and in particular~~ when ALT values are > ULN or ≥ 1.5x baseline value~~)~~ or based ~~on~~upon discussion between the Medical Monitor and the Investigator~~ and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN~~. • FVIII Assays o ~~FVIII protein assay~~	6, 9, 12

Section No./Title	Revision	Rationale
12.6.2/Years 2-5, Every 12 Weeks and End of Year Visits	At the every 12 week and End of Year visits, the following procedures will be performed: • Brief physical examination (at every 12 week visits) or complete physical examination (at End of Year visits; genitourinary examination may be deferred) • Liver Tests (refer to Table 9.7.6.~~3~~4.1) o ~~LTs~~LT assessment may be ~~monitored~~checked more ~~or less~~ frequently ~~(and in particular~~ when ALT values are > ULN or ≥ 1.5x baseline value~~)~~ or based ~~on~~upon discussion between the Medical Monitor and the Investigator ~~and review of subject data, but LTs will be monitored at least twice weekly during periods when a subject's ALT is ≥ 3x ULN~~. • AAV5 TAb Assay (at Week 64, Week 76, Week 88, Week 104, then at End of Year visit for Years 3-5) • AAV5 TI Assay (at End of Year visit for Years 2-5 only) • FVIII antibody titer (at Week 64, Week 76, Week 88, Week 104, then at End of Year visit for Years 3-5) • ~~PBMC collection~~ • Cytokine profiling • PBMC collection (at Week 64, Week 76, Week 88, Week 104, Week 128, Week 156, Week 180, Week 208, Week 232, and Week 260) • ~~TGA Assay~~ • Liver ultrasound (at End of Year visits only) o Additional liver ultrasounds may be performed at interim time points (ie, between the End of Year visits) at the discretion of the Investigator.	2, 6, 7, 9, 12
12.7/ETV	At the Early Termination visit, as many of the following assessments as possible should be done: • Cytokine ~~bead array assay~~profiling • PBMC collection ~~for CTL baseline~~ • ~~Thrombin Generation Assay~~ • Liver ultrasound	2, 7, 12
14.2/Efficacy Analysis	The subjects will be followed in a longitudinal manner, and all the relevant information will be collected. The analysis of the data will be descriptive in nature, but data collected in a longitudinal manner may be analyzed using longitudinal methods, such	12

Section No./Title	Revision	Rationale
	as mixed effect model, which take into account the correlation among the observations taken at various time points within a subject. Efficacy is a secondary endpoint, defined as biologically active FVIII at ≥ 5 IU/dL at 26 weeks following study treatment. ~~FVIII activity will be assessed weekly during the study period.~~	
14.6/Changes in Study Conduct	When a protocol amendment substantially alters the study design or the potential risks or burden to subjects, the ICF will be amended and approved by BioMarin and the IRB/EC, and all active subjects (or their legally authorized representative, if required) must again provide informed consent.	11
17/CRFs and Source Documents	BioMarin's policy is that study data on the eCRFs must be verifiable to the source data, which necessitates access to all original recordings, laboratory reports, and subject records. In addition, all source data should be attributable (signed and dated). The Investigator must therefore agree to allow direct access to all source data. Subjects (or their legally authorized representative) must also allow access to their medical records, and subjects (or their legally authorized representative) will be informed of this and will confirm their agreement when giving informed consent.	11
18/Study Monitoring	Qualified individuals designated by BioMarin will monitor all aspects of the study according to GCP and standard operating procedures for compliance with applicable government regulations. The Investigator agrees to allow these monitors direct access to the clinical supplies, dispensing, and storage areas and to the clinical files, including original medical records, of the study subjects, and, if requested, agrees to assist the monitors. The Investigator and staff are responsible for being present or available for consultation during routinely scheduled site visits conducted by BioMarin or its designees. When in person site monitoring or source data verification cannot be conducted, remote site monitoring and/or source data verification will be conducted where allowed by country and local health authorities and ECs/IRBs.	12
21/References	Haemo-QoL Study Group. Scoring Manual. Available at: http://haemoqol.de/scoring/ manual. Last accessed ~~28 July 2017~~17 May 2021. National Center for Biotechnology Information (NCBI). LiverTox: Clinical and Research Information on Drug-Induced Liver Injury. Available at: https://livertox.nih.gov (last accessed ~~20 August 2020~~17 May 2021). ~~Pasi KJ, Rangarajan S, Mitchell N, Lester W et al. First-in-human Evidence of Durable Therapeutic Efficacy and Safety of Durable Therapy Over Three-years with Valoctocogene Roxaparvovec for Severe Haemophilia A (BMN 270-201 Study). Res Pract Thromb Haemost. 2019;3(S2):2.~~ Reilly, M. WPAI Scoring. 2002. Available at: http://www.reillyassociates.net/ WPAI_Scoring.html. Last accessed ~~28 July 2017~~17 May 2021.	4, 12

Section No./Title	Revision	Rationale
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