A Phase 2a, Randomized, Double-Blind, Placebo-Controlled Study of IMR-687 in Adult Patients with Sickle Cell Anemia (Homozygous HbSS or Sickle-β^0 Thalassemia)

Unique Protocol ID:	IMR-SCD-102
NCT Number:	NCT03401112
EudraCT Number:	2017-000653-39
Date of SAP:	22 December 2020

ADDENDUM TO STATISTICAL ANALYSIS PLAN

IMR-SCD-102

A Phase 2a, Randomized, Double-Blind, Placebo-Controlled Study of IMR-687 in Adult Patients with Sickle Cell Anemia (Homozygous HbSS or Sickle-β^0 Thalassemia)

AUTHOR: PPD

VERSION NUMBER: V2.0

DATE: 22DEC2020

SAP ADDENDUM SIGNATURE PAGE - AUTHOR

	Name	Signature	Date
Author:	PPD		22DEC2020
Position:			
Company:	Imara, Inc.		
Approved By:	PPD		22DEC2020
Position:			
Company:	Imara, Inc.		

SAP ADDENDUM

This SAP addendum is based on a Sponsor blinded review of the study subject data without treatment codes.

1. Imputation rules for summaries and figures of lab data:
   - GFR- GFR will only be included in data listings and will not be summarized in any tables or figures.

     NOTE: GFR records provided by TDL were calculated without adjusting for PPD race as noted in the COVAL variable of TDL's lab dataset. Therefore, all GFR records need to be adjusted for PPD race by multiplying by 1.212. There is one randomized subject PPD with a race of PPD This subject should be considered as of PPD race in the GFR calculation.
   - GGT- Non-missing data range from 5 to 488 IU/L. Only one non-missing record with <3. Will not impute this one record.
   - APTT – Non-missing data range from 19 to 172 seconds with one >150 and two >180. Of the two records with >180, one with 'query sample integrity, suggest repeat' and the other with 'suggest repeat to confirm'. Will not impute these three records.
   - Prothrombin Time (PT1) – Non-missing data range from 10 to 39 seconds with one >120. Will not impute this record.
   - Haptoglobin – Non-missing data range from 0.1 to 2.2 g/L with many records <0.1. Impute <0.1 with 0.05.
   - hsCRP – Non-missing data range from 0.3 to 91.6 mg/L with three records <0.3. Impute <0.3 with 0.15.

Lab Category	Lab Test	Unit	Normal Range	Result	Imputation
Chemistry	GFR	mL/min/1.73m^2		>90	NA
Chemistry	GFR	mL/min/1.73m^2		Non-missing	Adjust for PPD race; multiply by 1.212
Chemistry	GGT	IU/L	M: [10, 71] F: [6, 42]	<3	NA
Coagulation	APTT	seconds	[25, 37]	>150, >180	NA
Coagulation	Prothrombin Time	seconds	[10, 12]	>120	NA
PD	Haptoglobins	g/L	[0.3, 2.0]	<0.1	0.05
PD	hsCRP	mg/L	[0.0, 5.0]	<0.3	0.15

2. Lab data
   - If there are multiple records for a lab test on the same date with different time for a subject, the average of these records will be used as the records for that date.

- There are three lab records from subject PPD for "hemoglobin S and C" incorrectly attributed to "hemoglobin S". These lab records have dates of PPD 2019, PPD 2019 and PPD 2019 with the same value of 95.2% based on the attached memo to file. These should be corrected in the lab dataset.
- When there are non-missing "hemoglobin S" and "hemoglobin C" values based on a blood sample of a subject (PPD add these two values in creating a value in the ADaM dataset ADLB for "hemoglobin S and C" if it does not already exist.
- There is one subject PPD having creatinine measured using an enzymatic procedure (LBTESTCD='CRQ') for some samples. These will be treated as "standard" creatinine (LBTESTCD='CREA') records in the ADaM dataset ADLB.
- Hemolyzed labs (or labs beyond stability) for F Cells will be excluded from PD summary statistics but will be included in the data listings with a comment field flagging if hemolyzed or beyond stability.
- PK samples analyzed outside of the established freezer stability limits, hemolyzed, or received beyond stability will still be used for PK parameter analysis as well as summary tables and graphs. A comment field will be included in the PK concentration listing indicating which samples were affected and reason why.
- There are two sets of CRP data, one (hsCRP) from TDL's lab data and the other (CRP) from Myriad's PD-specific lab data. For the analysis of hsCRP, we will use the data from Myriad's PD-specific lab data.

3. Exposure data
   - When the end date of starting dose is the same as the start date of escalated dose for a subject the former will be set to be the one day earlier. The above data convention is to be carried out programmatically to the exposure data in calculating drug exposure and compliance. This applies to the following patients: PPD
   - Subject PPD had first dose on PPD 2019 and dose increase on PPD 2019, both with end date of PPD 2020. The end date for the starting dose should be changed to PPD 2019 based on the attached memo to file.
4. CM data
   - The ATC class is not available in the dataset. Pain medications including opioids will be identified by Imara's medical review with an EXCEL file to be merged with CM data.
5. ImaraGo App data (from Mobile Programming)
   - Baseline (week prior to Day 1) ImaraGo data was not collected; therefore, daily data will be summarized by mean values of each week and not change from baseline.
6. PD analyses

In analyses of PD parameters, baseline is defined as the average of non-missing values from the screening and baseline visit. If either baseline or screening value is missing, the remaining value will be used as the baseline value. The end-of-treatment endpoint is Week 25 for populations A, A1 and B1 and Week 17 for population B. The primary PD endpoint analyses for change from baseline will be based on the primary baseline and end-of-treatment endpoint.

- Sensitivity PD endpoint analyses for change from baseline will be based on the baseline and end-of treatment endpoint. The average of non-missing values from Week 21 and Week 25 for populations A, A1 and B1 and the average of non-missing value from Week 13 and Week 17 for population B will be derived for PD parameters in ADaM dataset ADPD for potential ad hoc PD analyses.

7. PK analyses
   - AUC_{0-24} is missing from the list of primary PK parameters for the HU DDI assessment. Tau = 24 hr for HU. This should be added and at least attempted in the DDI assessment (as agreed upon by email on the day of SAP finalization). AUCtau should be included in box plot assessments.
   - In addition, the HU DDI assessment should be at the end of the study vs. lead-in PK evaluation; Day 29 (first day after change in IMR-687 dose to 100 mg). DDI assessment is not useful because IMR-687 is not expected to be at steady state just after a dose change.
   - There may be an outlier analysis to handle terminal 24 hour spikes in IMR-687 PK levels. If reported 24hr concentrations are >10-fold higher than expected, these outliers will be excluded, facilitating t1/2, AUCtau and Rac assessments as feasible.
   - When a subject's PK concentration-time profile is all BLQ on a full PK day, they should be excluded from summary statistics and corresponding graphs for that day. Subject's (BLQ) data is still included in the individual listings.
8. PD/PK analysis population: patients/timepoints to exclude:

Subject ID	Population	Exclude PD/PK Analysis Data post-date (MM/DD/YYYY)	Reason
PPD	A	PPD 2018	RBC transfusion received
	B	2018	RBC transfusion received
	A	2018	RBC transfusion received
	A1	2020	Did not dose escalate per the protocol
	A1	2020*	RBC transfusion received
	A	2018	RBC transfusion received
	B	2019	Did not dose escalate per the protocol
	A	Exclude subject- all timepoints	Did not dose escalated per exposure data
	A	PPD 2018	RBC transfusion received
	B	2018	RBC transfusion received
	B	Exclude subject- all timepoints	HU baseline/lead-in PKs missing
	A	PPD 2019	RBC transfusion received
	A1	2019	RBC transfusion received
	B	2019	RBC transfusion received
	A1	2019	RBC transfusion received
	B1	Exclude subject – all timepoints	Protocol violation of exclusion criteria due to pretreatment RBC transfusion

* Based on VOC data, the transfusion occurred post Day 1 but between PPD 2020 and PPD 2020.

Therefore, the earliest time to exclude data is PPD 2020.

NOTES:
RBC transfusion received: PK/PD data from the first transfusion start date forward will be excluded from PK/PD analysis. PD/PK listings will include all data and will include a flag to identify timepoints that were excluded from PD/PK analyses due to "RBC transfusion received".
HU baseline/lead-in PKs missing: When all samples for the HU baseline/lead-in PKs are missing (preventing the establishment of a stable dose of HU; protocol deviation), the subject should be excluded from PD/PK analyses. These subjects will also be excluded from the HU-IMR-687 drug interaction assessment by default. PD/PK listings will include all data and will include a flag to identify patients that were excluded from PD/PK analyses due to "HU baseline/lead-in PKs missing".
No dose escalation per protocol: PK/PD data from the date when the dose escalation was planned to occur per protocol forward will be excluded from PK/PD analysis. PD/PK listings will include all data and will include a flag to identify timepoints that were excluded from PD/PK analyses due to "no dose escalation".

Appendix: Notes to file

Imara, Inc.
116 Huntington Avenue, 6th Floor
Boston, MA 02116 USA
www.imaratx.com

PPD

www.imaratx.com

December 11, 2020

Memo-to-File

Re: Incorrect Attribution of Hemoglobin S and C Values to Hemoglobin S Values

Protocol: IMR-SCD-102

This memo-to-file serves to document that incorrect attribution of hemoglobin S and C values to hemoglobin S values in the lab data received from TDL.

There are three lab records from subject PPD for "hemoglobin S and C" incorrectly attributed to "hemoglobin S". These lab records have dates of PPD 19, PPD 19 and PPD 20 have same value of 95.2%. We will correct these in the lab dataset.

When there are non-missing hemoglobin S and non-missing hemoglobin C values based on a blood sample of a subject, we will add these two values in creating a value for hemoglobin S and C if it is not already existed in the lab dataset.

PPD
Imara, Inc.
PPD

Advancing New Sickle Cell Therapies

IMARA Inc.
116 Huntington Avenue, 6th Floor
Boston, MA 02116 USA

PPD

www.imaratx.com

Note to File

Date:	11-Dec-2020
From:	Imara, PPD
To:	eCRF
Protocol:	**IMR-SCD-102**
Study Title:	**A Phase 2a, Randomised, Double-Blind, Placebo-Controlled Study of IMR-687 in Adult Patients with Sickle Cell Anaemia (Homozygous HbSS or Sickle-β0 Thalassemia)**
Site:	PPD
Subject:	Change of the end date of the IP starting dose for subject PPD
Re:	Change in end date
Details:	This NTF is written to clarify the change of the end date of the IP starting dose for subject PPD For subject PPD (in B1 population) having the end date of the IP starting dose being the same as the end date of IP escalated dose, the end date of the IP starting dose will be changed to one day earlier than the start of IP escalated dose. Subject PPD had first IP starting dose on PPD 2019 and dose increase (escalation dose) on PPD 2019, both with end date of PPD 2020. The end date for the starting dose should be PPD 2019. The start and stop dates of IP doses will be: IP starting dose: PPD 2019 - PPD 2019 IP increase/escalation dose: PPD 2019 - PPD 2020

PPD (PI)
Name (Printed)

PPD

PPD

Name (Signature)

17 DEC 2020
Date

Note to file_v1.0_03Dec2020

ADDENDUM TO STATISTICAL ANALYSIS PLAN

IMR-SCD-102

A Phase 2a, Randomized, Double-Blind, Placebo-Controlled Study of IMR-687 in Adult Patients with Sickle Cell Anemia (Homozygous HbSS or Sickle-β^0 Thalassemia)

AUTHOR: PPD

VERSION NUMBER: V1.0

DATE: 05DEC2020

SAP ADDENDUM SIGNATURE PAGE - AUTHOR

	Name	Signature	Date
Author:	PPD		05DEC2020
Position:			
Company:	Imara, Inc.		
Approved By:	PPD		05DEC2020
Position:			
Company:	Imara, Inc.		

SAP ADDENDUM

This SAP addendum is based on a Sponsor blinded review of the study subject data without treatment codes.

1. Imputation rules for summaries and figures of lab data:
   - GFR- GFR will only be included in data listings and will not be summarized in any tables or figures.

     NOTE: GFR records provided by TDL were calculated without adjusting for PPD race as noted in the COVAL variable of TDL's lab dataset. Therefore, all GFR records need to be adjusted for PPD race by multiplying by 1.212. There is one randomized subject PPD with a race of PPD This subject should be considered as of PPD race in the GFR calculation.
   - GGT- Non-missing data range from 5 to 488 IU/L. Only one non-missing record with <3. Will not impute this one record.
   - APTT – Non-missing data range from 19 to 172 seconds with one >150 and two >180. Of the two records with >180, one with 'query sample integrity, suggest repeat' and the other with 'suggest repeat to confirm'. Will not impute these three records.
   - Prothrombin Time (PT1) – Non-missing data range from 10 to 39 seconds with one >120. Will not impute this record.
   - Haptoglobin – Non-missing data range from 0.1 to 2.2 g/L with many records <0.1. Impute <0.1 with 0.05.
   - hsCRP – Non-missing data range from 0.3 to 91.6 mg/L with three records <0.3. Impute <0.3 with 0.15.

Lab Category	Lab Test	Unit	Normal Range	Result	Imputation
Chemistry	GFR	mL/min/1.73m^2		>90	NA
Chemistry	GFR	mL/min/1.73m^2		Non-missing	Adjust for PPD race; multiply by 1.212
Chemistry	GGT	IU/L	M: [10, 71] F: [6, 42]	<3	NA
Coagulation	APTT	seconds	[25, 37]	>150, >180	NA
Coagulation	Prothrombin Time	seconds	[10, 12]	>120	NA
PD	Haptoglobins	g/L	[0.3, 2.0]	<0.1	0.05
PD	hsCRP	mg/L	[0.0, 5.0]	<0.3	0.15

2. Lab data
   - When there are non-missing "hemoglobin S" and "hemoglobin C" values based on a blood sample of a subject (PPD add these two values in creating a value in the ADaM dataset ADLB for "hemoglobin S and C" if it does not already exist.

- There is one subject PPD having creatinine measured using an enzymatic procedure (LBTESTCD='CRQ') for some samples. These will be treated as "standard" creatinine (LBTESTCD='CREA') records in the ADaM dataset ADLB.
- Hemolyzed labs (or labs beyond stability) for F Cells will be excluded from PD summary statistics but will be included in the data listings with a comment field flagging if hemolyzed or beyond stability.
- PK samples analyzed outside of the established freezer stability limits, hemolyzed, or received beyond stability will still be used for PK parameter analysis as well as summary tables and graphs. A comment field will be included in the PK concentration listing indicating which samples were affected and reason why.
- There are two sets of CRP data, one (hsCRP) from TDL's lab data and the other (CRP) from Myriad's PD-specific lab data. For the analysis of hsCRP, we will use the data from Myriad's PD-specific lab data.

3. Exposure data
   - When the end date of starting dose is the same as the start date of escalated dose for a subject the former will be set to be the one day earlier. The above data convention is to be carried out programmatically to the exposure data. This applies to the following patients: PPD
   - For subjects (PPD both in B1 population) having the end date of starting dose being the same as the end date of escalated dose, the end date of starting dose will be changed to one day earlier than the start of escalated dose.
     - Subject PPD had first dose on PPD 2019 and dose increase on PPD 2019, both with end date of PPD 2020. The end date for the starting dose should be changed to PPD 2019 based on the attached memo to file.
     - Subject PPD had first dose on PPD 2020, dose escalation on PPD 2020 both with end date of PPD 2020. The end date for the starting dose should be changed to PPD 2020 based on the attached memo to file.
4. CM data
   - The ATC class is not available in the dataset. Pain medications including opioids will be identified by Imara's medical review with an EXCEL file to be merged with CM data.
5. ImaraGo App data (from Mobile Programming)
   - Baseline (week prior to Day 1) ImaraGo data was not collected; therefore, daily data will be summarized by mean values of each week and not change from baseline.
6. PD analyses
   - In analyses of PD parameters, baseline is defined as the average of non-missing values from the screening and baseline visit. If either baseline or screening value is missing, the remaining value will be used as the baseline value. The end-of-treatment endpoint is Week 25 for populations A, A1 and B1 and Week 17 for population B. The primary PD endpoint analyses for change from baseline will be based on the primary baseline and end-of-treatment endpoint.

- Sensitivity PD endpoint analyses for change from baseline will be based on the baseline and end-of treatment endpoint. The average of non-missing values from Week 21 and Week 25 for populations A, A1 and B1 and the average of non-missing value from Week 13 and Week 17 for population B will be derived for PD parameters in ADaM dataset ADPD for potential ad hoc PD analyses.

7. PK analyses
   - AUC_{0-24} is missing from the list of primary PK parameters for the HU DDI assessment. Tau = 24 hr for HU. This should be added and at least attempted in the DDI assessment (as agreed upon by email on the day of SAP finalization). AUCtau should be included in box plot assessments.
   - In addition, the HU DDI assessment should be at the end of the study vs. lead-in PK evaluation; Day 29 (first day after change in IMR-687 dose to 100 mg). DDI assessment is not useful because IMR-687 is not expected to be at steady state just after a dose change.
   - There may be an outlier analysis to handle terminal 24 hour spikes in IMR-687 PK levels. If reported 24hr concentrations are >10-fold higher than expected, these outliers will be excluded, facilitating t1/2, AUCtau and Rac assessments as feasible.
   - When a subject's PK concentration-time profile is all BLQ on a full PK day, they should be excluded from summary statistics and corresponding graphs for that day. Subject's (BLQ) data is still included in the individual listings.
8. PD/PK analysis population: patients/timepoints to exclude:

Subject ID	Population	Exclude PD/PK Analysis Data post-date (MM/DD/YYYY)	Reason
PPD	A	PPD /2018	RBC transfusion received
	B	/2018	RBC transfusion received
	A	/2018	RBC transfusion received
	A1	/2020	Did not dose escalate per the protocol
	A1	2020*	RBC transfusion received
	A	/2018	RBC transfusion received
	B	/2019	Did not dose escalate per the protocol
	A	/2018	RBC transfusion received
	B	/2018	RBC transfusion received
	B	Exclude subject- all timepoints	HU baseline/lead-in PKs missing
	A	PPD 2019	RBC transfusion received
	A1	2019	RBC transfusion received
	B	2019	RBC transfusion received
	A1	2019	RBC transfusion received
	B1	Exclude subject – all timepoints	Protocol violation of exclusion criteria due to pretreatment RBC transfusion

* Based on VOC data, the transfusion occurred post Day 1 but between PPD 2020 and PPD 2020. Therefore, the earliest time to exclude data is PPD /2020.

NOTES:

RBC transfusion received: PK/PD data from the first transfusion start date forward will be excluded from PK/PD analysis. PD/PK listings will include all data and will include a flag to identify timepoints that were excluded from PD/PK analyses due to "RBC transfusion received".

HU baseline/lead-in PKs missing: When all samples for the HU baseline/lead-in PKs are missing (preventing the establishment of a stable dose of HU; protocol deviation), the subject should be excluded from PD/PK analyses. These subjects will also be excluded from the HU-IMR-687 drug interaction assessment by default. PD/PK listings will include all data and will include a flag to identify patients that were excluded from PD/PK analyses due to "HU baseline/lead-in PKs missing".

No dose escalation per protocol: PK/PD data from the date when the dose escalation was planned to occur per protocol forward will be excluded from PK/PD analysis. PD/PK listings will include all data and will include a flag to identify timepoints that were excluded from PD/PK analyses due to "no dose escalation".

Sponsor:	Imara, Inc.		
Protocol:	IMR-SCD-102		
Document Version No.:	1.0	**Document Date:**	3 NOV 2020

TABLE OF CONTENTS

Sponsor:	Imara, Inc.		
Protocol:	IMR-SCD-102		
Document Version No.:	1.0	**Document Date:**	3-NOV-2020

Protocol IMR-SCD-102

A Phase 2a, Randomized, Double-Blind, Placebo-Controlled Study of IMR-687 in Adult Patients with Sickle Cell Anaemia (Homozygous HbSS or Sickle-β^0 Thalassemia)

Protocol Number: (Version Date)	United States: 008 United Kingdom: 011 (28 June 2019)
Name of Test Drug:	IMR-687
Phase:	2a
Methodology:	Randomized, Double-blind, Placebo-controlled
Sponsor:	Imara, Inc. 116 Huntington Ave., 6th Floor Boston, MA 02116
Sponsor Representative:	PPD PPD Imara PPD PPD
Document Date:	3 November 2020
Document Version:	1.0

Sponsor:	Imara, Inc.		
Protocol:	IMR-SCD-102		
Document Version No.:	1.0	**Document Date:**	3-NOV-2020

SIGNATURE PAGE

Protocol Title:	A Phase 2a, Randomised, Double-Blind, Placebo-Controlled Study of IMR-687 in Adult Patients with Sickle Cell Anaemia (Homozygous HbSS or Sickle-β^0 Thalassemia)
Sponsor:	Imara, Inc. 116 Huntington Ave., 6th Floor Boston, MA 02116
Protocol Number:	IMR-SCD-102
Document Date/Version:	3 November 2020 (Version 1.0)

Cytel, Inc. Author:

PPD

Cytel, Inc.
675 Massachusetts Avenue
Cambridge, MA 02139

Signature:_____

Date:_____

Sponsor Approval

By signing this document, I acknowledge that I have read the document and approve of the planned statistical analyses described herein. I agree that the planned statistical analyses are appropriate for this study, are in accordance with the study objectives, and are consistent with the statistical methodology described in the protocol, clinical development plan, and all applicable regulatory guidance's and guidelines. I have discussed any questions I have regarding the contents of this document with the biostatistical author. I also understand that any subsequent changes to the planned statistical analyses, as described herein, may have a regulatory impact and/or result in timeline adjustments. All changes to the planned analyses will be described in the clinical study report (CSR).

Sponsor Signatories:

PPD

Imara, Inc.
116 Huntington Ave, 6th Floor
Boston, MA 02116

Signature:_____

Date:_____

PPD

Imara, Inc.
116 Huntington Ave, 6th Floor
Boston, MA 02116

Signature:_____

Date:_____

Sponsor:	Imara, Inc.		
Protocol:	IMR-SCD-102		
Document Version No.:	1.0	**Document Date:**	3-NOV-2020

LIST OF IN-TEXT TABLES

LIST OF IN-TEXT FIGURES

ABBREVIATIONS

Abbreviation	Definition
ADL	Activities of daily living
AE	Adverse events
ASCQ-Me	Adult Sickle Cell Quality-of-Life Measurement Information System
$AUC_{0\text{-}12}$	Area under the concentration versus time curve from time 0 to the 12 hr time point
$AUC_{0\text{-}24}$	Area under the concentration versus time curve from time 0 to the 24 hr time point
$AUC_{0\text{-}last}$	Area under the concentration versus time curve from time 0 to the last measurable time point
AUC_{inf}	Area under the concentration-time curve, from time 0 extrapolated to infinity
cGMP	Cyclic guanosine monophosphate
C_{max}	Maximum observed concentration
CTCAE	Common Terminology Criteria for Adverse Events
eCRF	Electronic case report form
CRO	Clinical research organization
CSR	Clinical study report
DDI	Drug-drug interactions
ECG	Electrocardiogram
FDA	Food and Drug Administration
HbF	Foetal haemoglobin
HbS	Sickle haemoglobin
HbSS	Homozygous sickle haemoglobin
HU	Hydroxyurea
IAP	Interim analysis plan
ICH	International Council for Harmonisation
ITT	Intent-to-treat
MedDRA	Medical Dictionary for Regulatory Activities
NO	Nitric oxide

Abbreviation	**Definition**
PD	Pharmacodynamic
PDF	Portable Document Format
PK	Pharmacokinetic
PT	Preferred term
QD	Once daily
RBC	Red blood cells
SAE	Serious adverse event
SAP	Statistical analysis plan
SCA	Sickle cell anaemia
SCD	Sickle cell disease
SI	International System of Units
SOA	Schedule of Assessments
SOC	System organ class
SRC	Safety Review Committee
$t_{½}$	Apparent terminal elimination half-life
TEAE	Treatment emergent adverse event
T_{max}	Time to maximum observed concentration
VOC	Vaso-occlusive crisis

Sponsor:	Imara, Inc.		
Protocol:	IMR-SCD-102		
Document Version No.:	1.0	**Document Date:**	3-NOV-2020

1. INTRODUCTION AND OBJECTIVES OF ANALYSIS

1.1. Introduction

The Sponsor is developing IMR-687 for the treatment of patients with sickle cell disease (SCD), which include homozygous sickle haemoglobin (HbSS) and sickle-β^0 thalassemia, collectively referred to as sickle cell anaemia (SCA).

With a neonatal incidence of 294,000 to 330,000 patients worldwide (Piel 2013), SCA is a rare inherited disorder of red blood cells (RBCs) that is both serious and life threatening. The most common manifestations of SCA include vaso-occlusive crisis (VOC), chronic and acute severe pain, acute chest syndrome, stroke, priapism, acute anaemia (particularly from aplastic crisis and splenic sequestration), increased susceptibility to infection, and progressive damage to major organs including the spleen, brain, kidney, heart, lung, skin, retina, vestibular cochlear systems, and bone. In developed countries where there is prenatal screening and widespread access to prophylactic and acute interventions, the median age of death remains in the 40s to 50s, though many patients succumb to the disease much earlier (Platt 1994; Lanzkron 2013; Paulukonis 2016).

SCA is caused by a specific point mutation in the gene encoding haemoglobin subunit beta that results in the substitution of a hydrophobic valine residue for glutamic acid in the 6th position from the N terminus of the β chain and leads to the production of abnormal haemoglobin ("sickle haemoglobin" or HbS), which polymerises when in the deoxygenated conformation. Under conditions of hypoxia, acidity, and/or dehydration, the intracellular concentration of deoxyHbS increases thereby favouring polymerization and causing RBCs to deform (i.e., sickle) or become rigid, leading to a complex cascade of haemolysis, inflammation, elevated cell adhesion, leucocytosis, oxidative stress, and endothelial dysfunction that culminates in the vascular obstruction and ischemia responsible for much of the observed morbidity.

Prior to July 2017, the only drug specifically approved for the treatment of SCA was hydroxyurea (HU), a small molecule inhibitor of ribonucleotide reductase that was originally developed for the treatment of myeloproliferative disorders. HU is also known as hydroxycarbamide and has been approved to treat SCA since 1998 in the United States and since 2001 in Europe. In SCA, HU reduces painful crises and the need for blood transfusions (Charache 1995), at least in part by increasing levels of foetal haemoglobin (HbF), which reduces RBC sickling and improves blood flow. These effects are mediated, at least in part, by increased nitric oxide (NO) and cyclic guanosine monophosphate (cGMP) production (Erard 1981; Platt 1984; Cokic 2003). Unfortunately, HU is often poorly tolerated and its widespread use is limited by concerns about its potential impact on fertility and reproduction, challenges achieving and maintaining an efficacious dose due to its hematologic toxicities, and requirements for monthly monitoring (Heeney 2008).

In July 2017, L-glutamine oral powder (Endari™) was approved in the US to reduce complications of SCD; however, the data available to date are limited primarily to the results from 230 patients (158 who received L-glutamine oral powder and 78 who received placebo) in the phase 3 pivotal trial. In this study, the median number of sickle cell crises through Week 48 (primary endpoint) was 3 in patients who received L-glutamine oral powder compared with 4 in patients who received placebo. Thus, additional novel, safe, and effective treatments to prevent the morbid complications of SCA in patients of all ages are still urgently needed.

1.2. Objectives of Statistical Analysis

This statistical analysis plan (SAP) is designed to outline the methods to be used in the final analysis of study data in order to answer the study objectives. Populations for analysis, data handling rules, statistical methods, and formats for data presentation are provided. The statistical analyses and summary tabulations described in this SAP will provide the basis for the results sections of the clinical study report (CSR) for this trial.

This SAP also will outline any differences in the currently planned analytical objectives relative to those planned in the study protocol.

1.3. Study Objectives

The primary objective of this study is:

- To assess the safety and tolerability of IMR-687 in adult patients with SCA, defined as HbSS or sickle-β^0 thalassemia, who are/are not receiving a stable dose of HU

The secondary objectives of this study are:

- To characterize the PK profile of IMR-687 in adult patients with SCA who are/are not receiving a stable dose of HU
- To characterize the PK profile of HU in adult patients with SCA before and after receiving IMR-687 to determine whether there is a clinically relevant PK interaction

The exploratory objectives of this study are:

- To assess the PD effects of IMR-687 in adult patients with SCA who are/are not receiving stable HU
- To assess the potential efficacy of IMR-687 in adult patients with SCA who are/are not receiving stable HU

2. STUDY DESIGN

2.1. Summary of Study Design

This is a randomized, double-blind, placebo-controlled study to evaluate the safety, tolerability, PK, and exploratory PD and clinical outcomes of the phosphodiesterase (PDE) type 9 (PDE9) inhibitor, IMR-687, administered once daily for 16 to 24 weeks in 2 populations of patients with SCA: those who are not receiving HU (Populations A and A1) and those who are currently receiving a stable dose of HU according to standard of care (Populations B and B1). The study design for Populations A, A1, B, and B1 are depicted graphically in Figure 1, Figure 2, Figure 3, and Figure 4, respectively Approximately 60 patients will be enrolled in Populations A and A1 combined and approximately 30 patients will be enrolled in Population B and B1 combined.

Throughout the study, all available clinical data is reviewed approximately every 2 weeks by the SRC, and dose escalation will occur on an individual-patient basis only if approved by the SRC based on review of each patient's individual clinical safety data. Note: because placebo and all dose levels of IMR-687 are the same in appearance, dose escalation will not affect study drug blinding.

Population A

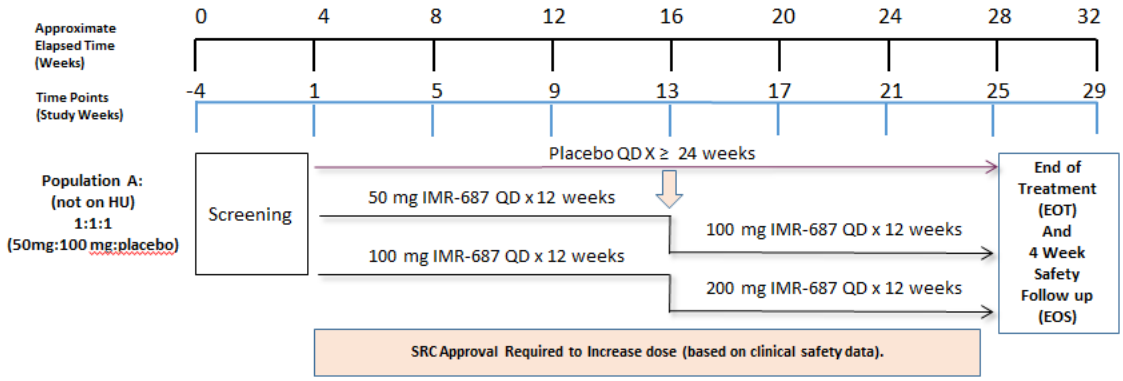
Population A is composed of those patients already enrolled at the time of Protocol Version 5 (19 December 2017) who are not receiving HU. These patients attend site visits at the time points specified in the Schedule of Assessments (SOA) provided in Table 1 (Population A).

Following a Screening period of up to 4 weeks, eligible patients in Population A receive either IMR-687 or placebo for a total of 24 weeks. On Day 1, patients are randomized 1:1:1 to receive oral IMR-687 50 mg, IMR-687 100 mg, or placebo daily for the first 12 weeks; for the second 12 weeks (Weeks 13-24), each patient's dose may be doubled (i.e., from 50 mg to 100 mg; from 100 mg to 200 mg; or placebo) based on SRC review.

The study design for Population A is depicted graphically in Figure 1.

Sponsor:	Imara, Inc.		
Protocol:	IMR-SCD-102		
Document Version No.:	1.0	**Document Date:**	3-NOV-2020

Figure 1: Study Design Schema for Population A



HU = hydroxyurea; QD = once daily; SRC = Safety Review Committee

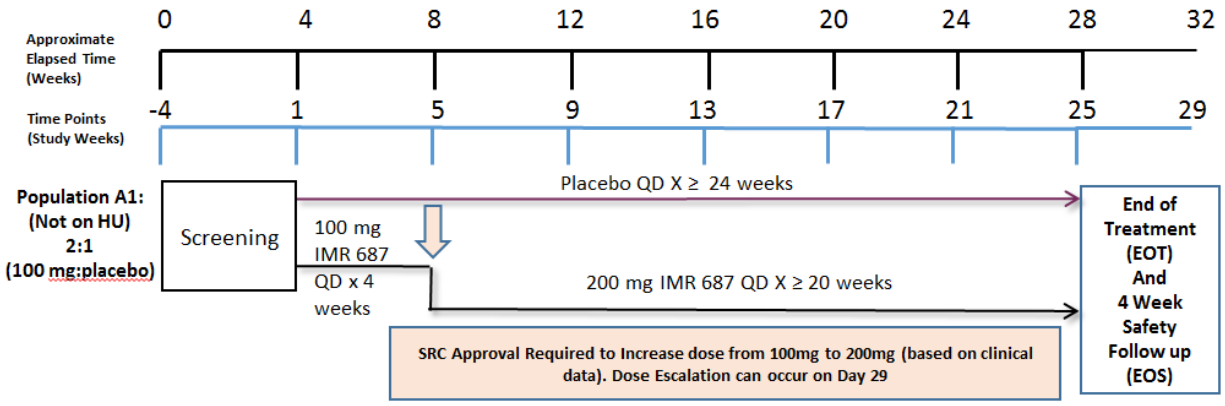
Population A1

Population A1 is composed of patients who are not receiving HU and were enrolled after Protocol Version US 5/UK 8 (7 January 2019). These patients attend site visits at the time points specified in the SOA provided in Table 2.

Following a Screening period of up to 4 weeks, eligible patients in Population A1 receive either IMR-687 or placebo for a total of 24 weeks. On Day 1, patients are randomized 2:1 to receive oral IMR-687 100 mg or placebo daily for 4 weeks; for the subsequent 20 weeks (Weeks 5-24), each patient's dose may be doubled (i.e., from 100 mg to 200 mg; or placebo) based on SRC review.

The study design for Population A1 is depicted graphically in Figure 2.

Figure 2: Study Design Schema for Population A1



HU = hydroxyurea; QD = once daily; SRC = Safety Review Committee

Population B

Population B consists of those patients already enrolled at the time of Protocol Version 6 (26 March 2018) who are receiving stable doses of HU. These patients attend site visits at time points specified in the SOA provided in Table 3.

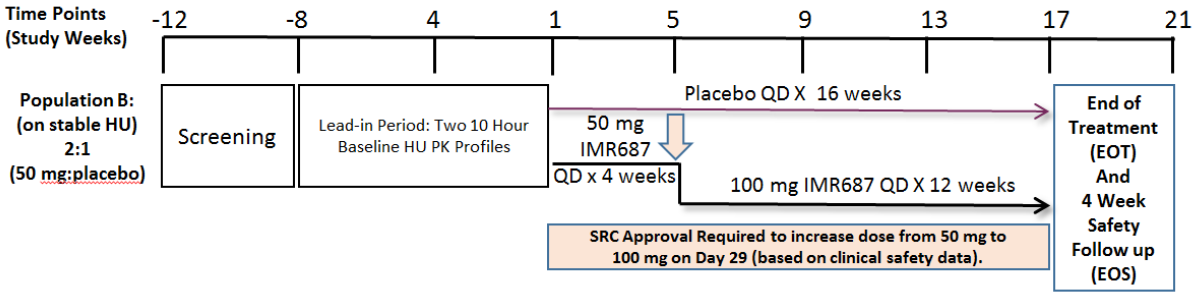
Following a Screening period of up to 4 weeks, eligible patients in Population B enter an approximately 8-week lead-in period and will have blood samples drawn to characterize the PK profile of the patient's prescribed dose of HU in the absence of IMR-687 (i.e., to characterize the patient's baseline HU PK profile). Two full baseline HU PK profile (with blood samples drawn over a 10-hour period) is determined.

IMR-687 dosing in Population B does not begin until at least 4 weeks of safety data from 6 patients in Population A have been reviewed by the SRC and the SRC has determined that it is safe and appropriate to begin dosing in Population B. Following SRC approval to initiate dosing in Population B and once the baseline HU PK blood draw are complete, patients are randomized 2:1 on Day 1 to receive oral IMR-687 50 mg or placebo for 16 weeks. For the first 4 weeks (Weeks 1-4), patients will receive study drug

according to their randomized treatment assignment; for the following 12 weeks (Weeks 5-16), each patient's dose may be doubled (i.e., from 50 mg to 100 mg; or placebo) based on SRC review.

The study design for Population B is depicted graphically in Figure 3.

Figure 3 Study Design Schema for Population B



HU = hydroxyurea; QD = once daily; SRC = Safety Review Committee

Population B1

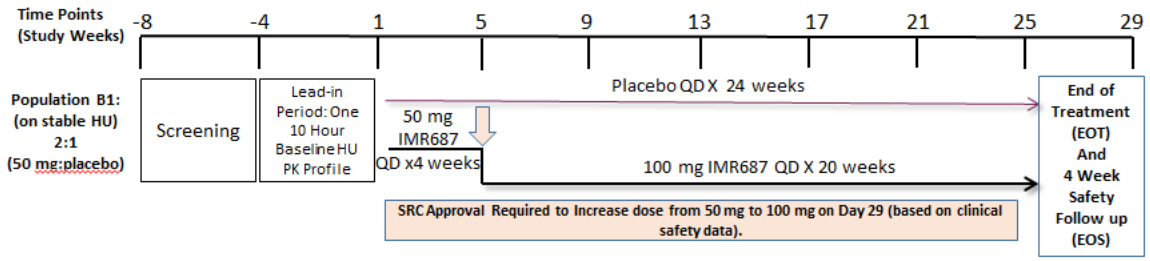
Population B1 consists of patients who are receiving stable doses of HU and were enrolled after Version 6 (26 March 2018). These patients attend site visits at the time points specified in the SOA provided in Table 4.

Following a Screening period of up to 4 weeks, eligible patients in Population B1 enter an approximate 4-week lead-in period and will have a single set of blood samples drawn to characterize the patient's baseline HU PK profile.

Once the baseline HU PK blood draw is complete, patients are randomized 2:1 on Day 1 to receive oral IMR-687 or placebo for 24 weeks. For the first 4 weeks (Weeks 1-4), patients receive study drug according to their randomized treatment assignment (i.e., 50 mg or placebo); for the following 20 weeks (Weeks 5-24), each patient's dose may be doubled (i.e., from 50 mg to 100 mg; or remain on placebo) based on SRC review.

The study design for Population B1 is depicted graphically in Figure 4.

Figure 4 Study Design Schema for Population B1



HU = hydroxyurea; QD = once daily; SRC = Safety Review Committee

2.2. Randomization Methodology

On Day 1, eligible patients in Populations A, A1, B, and B1 are assigned a unique number (randomization number) in sequential order. The randomization number codes the patient's initial treatment assignment to IMR-687 50 mg, IMR-687 100 mg, or placebo for Population A, IMR- 687 100 mg or placebo for Population A1, or IMR-687 50 mg or placebo for Populations B and B1, according to the randomization schedule generated prior to the study.

Randomization for Population A is 1:1:1 for each of the 3 groups; randomization for Population A1 is 2:1 for IMR-687 100 mg and placebo, and randomization for Populations B and B1 is 2:1 for IMR-687 50 mg and placebo.

Randomization numbers will not be re-used once assigned. In the event that a patient is replaced, the replacement patient will receive the same treatment as the replaced patient and will be assigned a randomization number incremented by PPD (e.g., PPD would replace PPD

2.3. Stopping Rules and Unblinding

In the exceptional circumstance where the investigator believes that knowledge of the study drug assignment is essential to provide appropriate medical management, the treatment assignment for that

patient is provided to the investigator. The Medical Monitor and Sponsor are available to the Investigator as needed for any considerations of unblinding a specific patient; however, the decision to unblind a specific patient relies solely on the clinical judgement of the Investigator and there is no requirement to discuss with the Medical Monitor and/or Sponsor prior to unblinding a specific patient by the Investigator. After breaking the blind, the site staff should record the reason(s) for breaking the blind and any AEs leading to the breaking of the blind in the source documents and the appropriate electronic case report form (eCRF) pages.

2.4. **Study Procedures**

The schedule of assessments, as outlined in the study protocol, is provided in Table 1-4.

Sponsor:	Imara, Inc.		
Protocol:	IMR-SCD-102		
Document Version No.:	1.0	**Document Date:**	3-NOV-2020

Table 1 Schedule of Assessments: Population A

	Screening	Double-Blind, Placebo-Controlled Treatment													End of Study (Safety FU)
Study Week(s)		**1**	**2**	**3**	**4**	**5**	**9**	**13**	**14**	**15**	**16**	**17**	**21**	**25[b] (EOT)**	**29 (EOS)**
Study Day(s)	**-28 to -1**	**1[a]**	**8 ± 2**	**15 ± 2**	**22 ± 2**	**29 ± 2**	**57 ± 2**	**85 ± 2**	**92 ± 2**	**99 ± 2**	**106 ± 2**	**113 ± 2**	**141 ± 2**	**169 ± 2**	**197± 5**
Informed Consent	X														
Demographic Information	X														
Medical/Disease History	X														
Inclusion/Exclusion Criteria	X														
Blood for Dx Confirmation & Pharmacogenomics[c]	X														
Randomization		X													
Telephonic Visits [d]				X	X					X	X				
Vital Signs[e]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight	X	X				X	X	X				X	X	X	
Height	X														
Physical Examination[f]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
12-Lead ECG[g]		X						X	X			X	X	X	
Safety Laboratory Assessments	X	X	X			X	X	X				X	X	X	X
Pregnancy Testing[h]	X	X				X	X	X				X	X	X	X
Exploratory PD Markers[i]		X				X	X	X				X	X	X	
IMR-687 Plasma PK[j]		X						X						X	
QOL Assessments (ASCQ-Me)		X						X						X	
App-based Pain Questionnaire		Daily													
Study Drug Administration		Once Daily Oral Administration IMR-687[k]													
AEs & Concomitant Medications		Continuous[l]													

Abbreviations: % F cells = percent of red blood cells containing foetal haemoglobin; AEs = adverse events; ASCQ-Me = Adult Sickle Cell Quality-of-Life Measurement Information System; BL = baseline; Dx = disease; FU = follow-up; ECG = electrocardiogram; EOT = end of treatment; PD = pharmacodynamics; PE = physical examination; PK = pharmacokinetic; QOL = quality of life; SFU = safety follow-up.

Note: Unless otherwise specified, all assessments should be completed prior to dosing at any given time point.

a The first day study drug is taken is considered "Day 1".
b The last day of dosing will be the end of Week 24 (Day 169 ± 2); patient should consume their last dose of study drug on site, after pre-dose assessments have been completed. During the study visit Day 169 ± 2, assessments should be collected from any patient who discontinues study drug or study prematurely. The week 25 visit will also be the EOT visit.
c Pharmacogenomic evaluation is optional and may be performed at any point during the study if patient provides informed consent.
d Site will contact the patient telephonically during Weeks 3, 4, 15, and 16. If any AEs of significant clinical concern are identified during the telephonic visit, the patient will be requested to come into the site to be assessed by targeted PE and vital sign measurements. Physical examinations and vital signs will not be assessed at these time points if patients do not have AEs of concern.
e On Days 1, 85, and the last day of dosing (End of Week 24 [Day 169 ± 2]), vital signs will be taken pre-dose and 2 hours (± 15 minutes) post-dose. At all other time points, vital signs will be taken pre-dose only. Vital sign measurements on Weeks 3, 4, 15, and 16 are obtained only if the patient is brought into the site after identification of AEs of significant clinical concern during the telephonic visits. Vital signs should be measured in the sitting or semi-supine position.
f At the Screening and Day 1 visits, the PE will include a general examination of the body including the abdomen, heart, chest, lungs, lymph nodes, dentition, back/neck, neurological system, skin, extremities, head, ears, eyes, nose, and throat. Subsequent PEs will be symptom directed; symptom-directed PEs on Weeks 3, 4, 15, and 16 are obtained only if the patient is brought into the site after identification of AEs of significant clinical concern during the telephonic visits.
g On Days 1, 85, and the last day of dosing (End of Week 24 [Day 169 ± 2]), ECGs will be obtained in triplicate at pre-dose and 2 hours (± 30 minutes) post-dose.
h Females of childbearing potential only. A serum pregnancy test will be performed at screening; all subsequent tests will be urine.
i Includes total haemoglobin (Hb), haemoglobin F (% HbF levels and % F cells), indices of red cell haemolysis (unconjugated bilirubin, reticulocyte counts, lactate dehydrogenase [LDH], haptoglobin, soluble E-selectin [sE-sel], soluble P-selectin [sP-sel], soluble intercellular adhesion molecule 1 [sICAM-1], and high-sensitivity C-reactive protein [hsCRP]). Blood samples should be obtained within 30 minutes prior to administration of study drug.
j On Days 1, 85, and the last day of dosing (End of Week 24 [Day 169 ± 2]), serial blood samples for IMR-687 plasma concentrations will be drawn pre-dose (within 30 minutes);at 0.5, 1, 1.5, 2, 4, 6, and 8 hours (± 5 minutes) after administration of study drug; and at 24 hours (± 1 hour) after administration of study drug.
k At each study visit, patients should be reminded to bring their study drug to their next site visit and to not take study drug on the day of the next visit until instructed to do so by the site during the visit. The last day of dosing will be the end of Week 24 (Day 169 ± 2); patient should consume the last dose on site after pre-dose assessments have been completed.
l Adverse events will be recorded throughout the study from Screening through the safety follow-up. In addition to the specified study time points in the Schedule of Assessments, patients will be followed weekly by the investigator for safety. Telephone-based check-ins may be performed on weeks where there are no scheduled site visits.

Table 2 Schedule of Assessments: Population A1

	Screening	Double-Blind, Placebo-Controlled Treatment													End of Study (Safety FU)
Study Week(s)		1	2	3	4	5	9	13	14	15	16	17	21	25[b] EOT	29 (EOS)
Study Day(s)	-28 to -1	1[a]	8 ± 2	15 ± 2	22 ± 2	29 ± 2	57 ± 2	85 ± 2	92 ± 2	99 ± 2	106 ± 2	113 ± 2	141 ± 2	169 ± 2	197± 5
Informed Consent	X														
Demographic Information	X														
Medical/Disease History	X														
Inclusion/Exclusion Criteria	X														
Blood for Dx Confirmation & Pharmacogenomics[c]	X														
Randomization		X													
Telephonic Visits [d]				X						X	X				
Vital Signs[e]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight	X	X				X	X	X				X	X	X	
Height	X														
Physical Examination[f]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
12-Lead ECG[g]		X				X	X	X	X			X	X	X	
Safety Laboratory Assessments	X	X	X		X	X	X	X				X	X	X	X
Pregnancy Testing[h]	X	X			X	X	X	X				X	X	X	X
Exploratory PD Markers[i]		X				X	X	X				X	X	X	
IMR-687 Plasma PK[j]		X				X								X	
QOL Assessments (ASCQ-Me)		X						X						X	
App-based Pain Questionnaire		Daily													
Study Drug Administration		Once Daily Oral Administration IMR-687[k]													
AEs & Concomitant Medications		Continuous[l]													

Abbreviations: % F cells = percent of red blood cells containing foetal haemoglobin; AEs = adverse events; ASCQ-Me = Adult Sickle Cell Quality-of-Life Measurement Information System; BL = baseline; Dx = disease; FU = follow-up; ECG = electrocardiogram; EOT = end of treatment; PD = pharmacodynamics; PK = pharmacokinetic; QOL = quality of life; SFU = safety follow-up.

Note: Unless otherwise specified, all assessments should be completed prior to dosing at any given time point.

a The first day study drug is taken is considered "Day 1".
b The last day of dosing will be the end of Week 24 (Day 169 ± 2); patient should consume their last dose of study drug on site, after pre-dose assessments have been completed. During the study visit Day 169 ± 2, assessments should be collected from any patient who discontinues study drug or study prematurely.
c Pharmacogenomic evaluation is optional and may be performed at any point during the study if patient provides informed consent.
d Site will contact the patient telephonically during Weeks 3, , 15, and 16. If any AEs of significant clinical concern are identified during the telephonic visit, the patient will be requested to come into the site to be assessed by targeted PE and vital sign measurements. Physical examinations and vital signs will not be assessed at these time points if patients do not have AEs of concern.
e On Days 1, 29, and the last day of dosing (End of Week 24 [Day 169 ± 2]), vital signs will be taken pre-dose and 2 hours (± 15 minutes) post-dose. At all other time points, vital signs will be taken pre-dose only. Vital sign measurements on Weeks 3, 15, and 16 are obtained only if the patient is brought into the site after identification of AEs of significant clinical concern during the telephonic visits. Vital signs should be measured in the sitting or semi-supine position.
f At the Screening and Day 1 visits, the PE will include a general examination of the body including the abdomen, heart, chest, lungs, lymph nodes, dentition, back/neck, neurological system, skin, extremities, head, ears, eyes, nose, and throat. Subsequent PEs will be symptom directed; symptom-directed PEs on Weeks 3, 15, and 16 are obtained only if the patient is brought into the site after identification of AEs of significant clinical concern during the telephonic visits.
g On Days 1, 29, and the last day of dosing (End of Week 24 [Day 169 ± 2]), ECGs will be obtained in triplicate at pre-dose and 2 hours (± 30 minutes) post-dose.
h Females of childbearing potential only. A serum pregnancy test will be performed at screening; all subsequent tests will be urine.
i Includes total haemoglobin (Hb), haemoglobin F (% HbF levels and % F cells), indices of red cell haemolysis (unconjugated bilirubin, reticulocyte counts, lactate dehydrogenase [LDH], haptoglobin, soluble E-selectin [sE-sel], soluble P-selectin [sP-sel], soluble intercellular adhesion molecule 1 [sICAM-1], and high-sensitivity C-reactive protein [hsCRP]). Blood samples should be obtained within 30 minutes prior to administration of study drug.
j On Days 1, 29, and the last day of dosing (End of Week 24 [Day 169 ± 2]), serial blood samples for IMR-687 plasma concentrations will be drawn pre-dose (within 30 minutes); at 0.5, 1, 1.5, 2, 4, 6, and 8 hours (± 5 minutes) after administration of study drug; and at 24 hours (± 1 hour) after administration of study drug. If well-tolerated, dose escalation from 100 mg to 200 mg will occur after 4 weeks of treatment.
k At each study visit, patients should be reminded to bring their study drug to their next site visit and to not take study drug on the day of the next visit until instructed to do so by the site during the visit. The last day of dosing will be the end of Week 24 (Day 169 ± 2); patient should consume the last dose on site after pre-dose assessments have been completed.
l Adverse events will be recorded throughout the study from Screening through the safety follow-up. In addition to the specified study time points in the Schedule of Assessments, patients will be followed weekly by the investigator for safety. Telephone-based check-ins may be performed on weeks where there are no scheduled site visits.

Table 3 Schedule of Assessments: Population B

			Double-Blind, Placebo-Controlled Treatment											End of Study (Safety FU)
Study Week(s)	**Screening Period[a]**	**Lead-in Period[b]**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**13**	**17[d] (EOT)**	**21(EOS)**
Study Day(s)			**1[c]**	**8 ± 2**	**15 ± 2**	**22 ± 2**	**29 ± 2**	**36 ± 2**	**43 ± 2**	**50 ± 2**	**57 ± 2**	**85 ± 2**	**113 ± 2**	**141 ± 5**
Informed Consent	X													
Demographic Information	X													
Medical/Disease History	X													
Inclusion/Exclusion Criteria	X													
Blood for Dx Confirmation & Pharmacogenomics[e]	X													
Randomization			X											
Telephonic Visits[f]					X	X		X	X	X				
Vital Signs[g]	X		X	X	X	X	X	X	X	X	X	X	X	X
Weight	X		X				X				X	X	X	
Height	X													
Physical Examination[h]	X		X	X	X	X	X	X	X	X	X	X	X	X
12-Lead ECG[i]			X				X						X	
Safety Laboratory Assessments	X		X	X			X				X	X	X	X
Pregnancy Testing[j]	X		X				X				X	X	X	
Exploratory PD Markers[k]			X				X				X	X	X	
IMR-687 Plasma PK[l]			X				X						X	
HU PK[m]		X					X						X	
QOL Assessments (ASCQ-Me)			X				X						X	
App-based Pain Questionnaire			Daily											
Study Drug Administration			Once Daily Oral Administration IMR-687 and HU[n]											
AEs & Concomitant Medications			Continuous[o]											

Abbreviations: % F cells = percent of red blood cells containing foetal haemoglobin; AEs = adverse events; App = application; BL = baseline; Dx = disease; ECG = electrocardiogram; EOS = end of study; EOT = end of treatment; FU = follow-up; HU = hydroxyurea; PD = pharmacodynamic; PE = physical examination; PK = pharmacokinetic; QOL = quality of life; SRC = safety review committee.

Note: Unless otherwise specified, all assessments should be completed prior to dosing.

a Screening should occur within a 4-week period and can begin approximately 12 weeks prior to the anticipated first dose of study drug in Population B.
b Once all Screening assessments have been performed and the patient is eligible for the study, the patient will enter the lead-in period and will have serial blood samples drawn for 2 complete baseline HU PK profiles. For each profile, samples will be drawn pre-dose (within 30 minutes); at 0.5, 1, 1.5, 3, 6, 8, and 10 hours (± 5 minutes) after self administration of the prescribed dose of HU. The duration of the lead-in period is up to approximately 8 weeks, depending on when at least 4 weeks of safety data from at least 6 patients in Population A are available for SRC review.
c The first day study drug is taken is considered "Day 1"; unless otherwise specified. Administration of study drug (Day 1) in Population B will not begin until at least 4 weeks of safety data from at least 6 patients in Population A have been reviewed by the SRC and the SRC has determined that it is safe and appropriate to begin dosing in Population B.
d The last day of dosing will be the end of Week 16 (Day 113 ± 2), patient should consume the last dose on site after pre-dose assessments have been completed. During study visit Day 113 ± 2, assessments should be collected from any patient who discontinues study drug or study prematurely.
e Assessments are optional and may be performed at any point during the study if the patient consents.
f Site will contact the patient telephonically during study week 3, 4, 6, 7, and 8. If any AEs of significant clinical concern are identified during the telephonic visit, the patient will be requested to come into the site to be assessed by targeted PE and vital sign measurements. Physical examinations and vital signs will not be assessed at these time points if patients do not have AEs of concern.
g On Days 1, 29, and the last day of dosing [End of Week 16 (Day 113 ± 2)], vital signs will be taken pre-dose and 2 hours (± 15 minutes) post-dose. At all other time points, vital signs will be taken pre-dose only. Vital sign measurements on Weeks 3, 4, 6, 7, and 8 are obtained only if the patient is brought into the site after identification of AEs of significant clinical concern during the telephonic visits. Vital signs should be measured in the sitting or semi-supine position.
h At the Screening and Day 1 visits, the PE will include a general examination of the body including the abdomen, heart, chest, lungs, lymph nodes, dentition, back/neck, neurological system, skin, extremities, head, ears, eyes, nose, and throat. Subsequent PEs will be symptom-directed; symptom-directed PEs on Weeks 3, 4, 6, 7, and 8 are obtained only if the patient is brought into the site after identification of AEs of significant clinical concern during the telephonic visits.
i On Days 1, 29, and the last day of dosing (End of Week 16 [Day 113 ± 2]), ECGs will be obtained in triplicate at pre-dose and 2 hours (± 30 minutes) post-dose.
j Females of childbearing potential only; a serum pregnancy test will be performed at screening; all subsequent tests will be urine.
k Includes total haemoglobin (Hb), haemoglobin F (% HbF levels and % F cells), indices of red cell haemolysis (unconjugated bilirubin, reticulocyte counts, lactate dehydrogenase [LDH], haptoglobin, soluble E selectin [sE-sel], soluble P-selectin [sP-sel], soluble intercellular adhesion molecule 1 [sICAM-1], and high-sensitivity C-reactive protein [hsCRP]). Blood samples should be obtained within 30 minutes prior to administration of study drug.
l On Days 1, 29, and 113, serial blood samples for IMR-687 PK will be drawn pre-dose (within 30 minutes); at 0.5, 1, 1.5, 2, 4, 6, and 8 hours (± 5 minutes) after administration of study drug; and at 24 hours (± 1 hour) after administration of study drug.
m Baseline blood samples for HU PK will be drawn as described in footnote 'b'. On Days 29 and 113, serial blood samples for HU PK will be drawn pre-dose (within 30 minutes) and at 0.5, 1, 1.5, 3, 6, 8, and 10 hours (± 5 minutes) after self-administration of the prescribed dose of HU.
n At each study visit, patients should be reminded to bring their study drug and their HU to their next site visit and to not take either medication on the day of the next visit until instructed to do so by the site during the visit.
o Adverse events will be recorded throughout the study from Screening to end of Follow-up. In addition to the specified study time points in the Schedule of Assessments, patients will be followed weekly by the investigator for safety. Telephone-based check-ins may be performed on weeks where there are no scheduled site visits.

Table 4 Schedule of Assessments: Population B1

			Double-Blind, Placebo-Controlled Treatment													End of Study (Safety FU)
Study Week(s)	**Screening Period[a]**	**Lead-in Period[b]**	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**13**	**17**	**21**	**25[d] (EOT)**	**29 (EOS)**
Study Day(s)			**1[c]**	**8 ± 2**	**15 ± 2**	**22 ± 2**	**29 ± 2**	**36 ± 2**	**43 ± 2**	**50 ± 2**	**57 ± 2**	**85 ± 2**	**113 ± 2**	**141 ± 2**	**169 ± 2**	**197± 5**
Informed Consent	X															
Demographic Information	X															
Medical/Disease History	X															
Inclusion/Exclusion Criteria	X															
Blood for Dx Confirmation & Pharmacogenomics[e]	X															
Randomization			X													
Telephonic Visits[f]					X	X		X	X	X						
Vital Signs[g]	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X
Weight	X		X				X				X	X	X	X	X	
Height	X															
Physical Examination[h]	X		X	X	X	X	X	X	X	X	X	X	X	X	X	X
12-Lead ECG[i]			X				X								X	
Safety Laboratory Assessments	X		X	X			X				X	X	X	X	X	X
Pregnancy Testing[j]	X		X				X				X	X	X	X	X	
Exploratory PD Markers[k]			X				X				X	X	X	X	X	
IMR-687 Plasma PK[l]			X				X								X	
HU PK[m]		X					X								X	
QOL Assessments (ASCQ-Me)			X				X						X		X	
App-based Pain Questionnaire			Daily													
Study Drug Administration			Once Daily Oral Administration IMR-687 and HU[n]													
AEs & Concomitant Medications			Continuous[o]													

Abbreviations: % F cells = percent of red blood cells containing foetal haemoglobin; AEs = adverse events; App = application; BL = baseline; Dx = disease; ECG = electrocardiogram; EOS = end of study; EOT = end of treatment; FU = follow-up; HU = hydroxyurea; PD = pharmacodynamic; PE = physical examination; PK = pharmacokinetic; QOL = quality of life; SRC = safety review committee.

Note: Unless otherwise specified, all assessments should be completed prior to dosing.

a Screening should occur within a 4-week period and can begin approximately 8 weeks prior to the anticipated first dose of study drug in Population B.
b Once all Screening assessments have been performed and the patient is eligible for the study, the patient will enter the lead-in period and will have serial blood samples drawn for 1 complete baseline HU PK profile. For the baseline HU PK profile, samples will be drawn pre-dose (within 30 minutes); at 0.5, 1, 1.5, 3, 6, 8, and 10 hours (± 5 minutes) after self-administration of the prescribed dose of HU. The duration of the lead-in period is approximately 4 weeks.
c The first day study drug is taken is considered "Day 1"; unless otherwise specified.
d The last day of dosing will be the end of Week 24 (Day 169 ± 2); patient should consume the last dose on site after pre-dose assessments have been completed. During study visit Day 169 ± 2, assessments should be collected from any patient who discontinues study drug or study prematurely.
e Assessments are optional and may be performed at any point during the study if the patient consents.
f Site will contact the patient telephonically during study week 3, 4, 6, 7, and 8. If any AEs of significant clinical concern are identified during the telephonic visit, the patient will be requested to come into the site to be assessed by targeted PE and vital sign measurements. Physical examinations and vital signs will not be assessed at these time points if patients do not have AEs of concern.
g On Days 1, 29, and the last day of dosing (End of Week 24 [Day 169 ± 2]), vital signs will be taken pre-dose and 2 hours (±15 minutes) post-dose. At all other time points, vital signs will be taken pre-dose only. Vital sign measurements on Weeks 3, 4, 6, 7, and 8 are obtained only if the patient is brought into the site after identification of AEs of significant clinical concern during the telephonic visits. Vital signs should be measured in the sitting or semi-supine position.
h At the Screening and Day 1 visits, the PE will include a general examination of the body including the abdomen, heart, chest, lungs, lymph nodes, dentition, back/neck, neurological system, skin, extremities, head, ears, eyes, nose, and throat. Subsequent PEs will be symptom-directed; symptom-directed PEs on Weeks 3, 4, 6, 7, and 8 are obtained only if the patient is brought into the site after identification of AEs of significant clinical concern during the telephonic visits.
i On Days 1, 29, and the last day of dosing (End of Week 24 [Day 169 ± 2]), ECGs will be obtained in triplicate at pre-dose and 2 hours (±30 minutes) post-dose.
j Females of childbearing potential only; a serum pregnancy test will be performed at screening; all subsequent tests will be urine.
k Includes total haemoglobin (Hb), haemoglobin F (% HbF levels and % F cells), indices of red cell haemolysis (unconjugated bilirubin, reticulocyte counts, lactate dehydrogenase [LDH], haptoglobin, soluble E selectin [sE-sel], soluble P-selectin [sP-sel], soluble intercellular adhesion molecule 1 [sICAM-1], and high-sensitivity C-reactive protein [hsCRP]). Blood samples should be obtained within 30 minutes prior to administration of study drug.
l On Days 1, 29, and 169, serial blood samples for IMR-687 PK will be drawn pre-dose (within 30 minutes); at 0.5, 1, 1.5, 2, 4, 6, and 8 hours (± 5 minutes) after administration of study drug; and at 24 hours (± 1 hour) after administration of study drug.
m Baseline blood samples for HU PK will be drawn as described in footnote 'b'. On Days 29 and 169, serial blood samples for HU PK will be drawn pre-dose (within 30 minutes) and at 0.5, 1, 1.5, 3, 6, 8, and 10 hours (± 5 minutes) after self-administration of the prescribed dose of HU.
n At each study visit, patients should be reminded to bring their study drug and their HU to their next site visit and to not take either medication on the day of the next visit until instructed to do so by the site during the visit.
o Adverse events will be recorded throughout the study from Screening to end of Follow-up. In addition to the specified study time points in the Schedule of Assessments, patients will be followed weekly by the investigator for safety. Telephone-based check-ins may be performed on weeks where there are no scheduled site visits.

2.5. Efficacy, Pharmacokinetic, Pharmacodynamic and Safety Variables

2.5.1. Efficacy Variables

The following SCA-related clinical outcome measures will be evaluated to assess the potential efficacy of IMR-687:

- Pain-related measures as captured using an app-based daily questionnaire, including:
  - Frequency and severity of pain
  - Frequency and severity of fatigue
  - Impact of pain/fatigue on work/school and on activities of daily living (ADL)
  - Need for professional medical attention due to sickle cell pain
  - Use of medication for sickle cell pain
- VOCs, including number of events of acute painful crisis and frequency of acute chest symptoms, including fever, cough, sputum production, shortness of breath, tachypnea, hypoxia, chest pain.
- Use of pain medications including opioids
- ASCQ-Me Outcomes, including:
  - Emotional impact
  - Pain episode frequency and severity
  - Pain impact
  - Sleep impact
  - Social functioning impact
  - Stiffness impact
  - SCD medical history checklist

2.5.2. Pharmacokinetic Analysis

IMR-687

The following single dose pharmacokinetic endpoints will be estimated for IMR-687 on Study Day 1:

- C_{max}, T_{max}, AUC_{last}, and $AUC_{0\text{-}tau}$, where tau = 24 hours. Half-life ($t_{½}$) and $AUC_{0\text{-}inf}$ will be generated as data permit.

The following steady-state pharmacokinetic endpoints will be generated for IMR-687 on Study Day 85 (for population A) and Day 29 (for Populations A1, B and B1):

- C_{max}, T_{max}, AUC_{last}, $AUC_{0\text{-}tau}$, where tau = 24 hours, and Accumulation ratio (R_{ac}). Half-life ($t_{½}$) will be generated as data permit.

The following steady-state pharmacokinetic endpoints will be generated for IMR-687 at the end of therapy:

- C_{max}, T_{max}, AUC_{last}, $AUC_{0\text{-}tau}$, where tau = 24 hours, and Accumulation ratio (R_{ac}). Half-life ($t_{½}$) will be generated as data permit.

HU

The following pharmacokinetic endpoints will be estimated for HU prior to receiving the first dose of test drug (i.e. IMR-687 or placebo), on study day 29, and at the end of therapy:

- C_{max}, T_{max}, AUC_{last}, $AUC_{0\text{-}12}$, and $AUC_{0\text{-}tau}$, where tau = 24 hours.
- The geometric mean ratios of C_{max}, AUC_{last}, $AUC_{0\text{-}12}$, and $AUC_{0\text{-}tau}$ for HU in the presence (End of Therapy) or absence of IMR-687 (baseline).
- The geometric mean ratio of C_{max}, AUC_{last}, $AUC_{0\text{-}12}$, and $AUC_{0\text{-}tau}$ for HU in the presence of maintenance therapy (End of Therapy) or at the end of the lead-in dose period (Study Day 29).

2.5.3. Pharmacodynamic Variables

The following endpoints will be evaluated to assess the PD activity of IMR-687 (additional exploratory PD biomarkers may also be assayed):

- Total haemoglobin (Hb) level

- HbF value (%)
- % F cells
- Indices of red cell haemolysis (indirect bilirubin, reticulocyte count [% and absolute], lactate dehydrogenase [LDH], and haptoglobin levels)
- sE-Selectin, sP-Selectin, sICAM-1 and sVCAM-1
- High-sensitivity C-reactive protein (hs-CRP)
- Mean corpuscular volume (MCV)
- Myeloperoxidase (MPO)
- N-terminal prohormone of brain natriuretic peptide (NT-proBNP)

2.5.4. **Safety Variables**

The safety and tolerability of IMR-687 will be assessed by the following endpoints:

- Incidence and severity of AEs and SAEs
- Observed values and changes from baseline in 12-lead ECG parameters, clinical laboratory tests (chemistry, haematology, coagulation, urinalysis), and vital signs
- Physical examination findings
- Use of concomitant medications and therapies

3. PATIENT POPULATIONS

3.1. Analysis Populations

The following patient populations will be evaluated and used for presentation and analysis of the data:

- Safety Analysis Set: is defined as all patients who have received any amount of study drug and from whom informed consent has been obtained and will be used to summarize all safety and tolerability data. The Safety Analysis Set will be used for listings of all data, including safety, tolerability, and PK/PD concentrations and parameters. Patients will be included in a treatment group based on the actual treatment received.
- PK Concentration Set: The PK Concentration Set is a subset of the safety analysis set and includes all patients who are enrolled in the study, have received at least one dose of IMR-687, and have any measurable IMR-687 concentration-time data, without protocol deviations or events expected to affect PK. Pharmacokinetic IMR-687 (and HU for patients in Populations B and B1) concentration profiles will be summarized based on the PK concentration set.
- Pharmacokinetic Analysis Set (PKA): The PKA consists of the subset of the Safety analysis set for which sufficient concentration and dosing/sampling date/time data are available. Inclusion of participants with missing data or protocol deviations in the PKA will be considered by the pharmacokineticist on a case-by-case basis. The PKA will be used for all tables and graphical summaries of the PK parameter data.
- PD Evaluable Set: is a subset of the safety analysis set and includes all patients who have provided samples for PD analysis sufficient to obtain at least one valid PD observation, without protocol deviations or events that would be expected to affect the PD analysis. PD observation and parameter data will be summarized based on the PD Evaluable Set. Patients will be included in a treatment group based on the actual treatment received.

3.2. **Protocol Deviations**

At the discretion of the sponsor, major protocol deviations as determined by a review of the data prior to unblinding of the study results and the conduct of statistical analyses may result in the removal of a patient's data from the PK Evaluable Set and PD Evaluable Set. Use of any prohibited medication and/or therapy must also be recorded on the eCRF and must be documented as a protocol deviation.

The sponsor will be responsible for producing the final protocol deviation file, in collaboration with the data monitoring group. This file will include a description of the protocol deviation, and clearly identify whether or not this protocol deviation warrants exclusion from the PK Analysis Set and PD Evaluable Set. This file will be finalized prior to hard database lock.

4. STATISTICAL METHODS

4.1. Sample Size Justification

A total of approximately 90 evaluable patients are expected to enroll in this study.

- In Populations A and A1, approximately 60 adult patients with a confirmed diagnosis of SCA who have not received HU for at least 90 days prior to Screening and who are not planning to take HU within the next 6 months will be enrolled.
- In Populations B and B1, approximately 30 adult patients with a confirmed diagnosis of SCA who have received HU for at least 6 months, have been on a stable dose for at least 60 days prior to Screening, and are not planning to change the dose or discontinue HU within the next 6 months will be enrolled.

The sample sizes for Populations A, A1, B, and B1 were not based on formal statistical considerations. However, the number of patients involved in this placebo-controlled study is considered to be sufficient to achieve the principal objectives of this exploratory study.

4.2. General Statistical Methods and Data Handling

4.2.1. General Methods

All output will be incorporated into Portable Document Format (PDF) files, sorted and labeled according to the International Council for Harmonisation (ICH) recommendations, and formatted to the appropriate page size(s).

Tabulations will be produced for appropriate demographic, baseline, efficacy, pharmacokinetic, pharmacodynamic, and safety parameters. Unless otherwise specified, separate tables will be generated for each population.

Continuous data will be summarized using descriptive statistics (number of patients, mean, standard deviation [SD], median, Q1, Q3, minimum, and maximum) and, where appropriate, coefficient of variation (%CV). Graphs of actual values and changes over time will also be created. Graphs of percent changes over time will also be created for selected data. Categorical data will be summarized including number and percentage within each category (with a category for missing data).

Formal statistical hypothesis testing will not be performed. No p-values or confidence intervals will be presented. Data will be summarized separately by study population (Populations A, A1, B, and B1) and by treatment group. Listings will be sorted by study population, treatment group, and patient ID.

4.2.2. **Computing Environment**

All statistical analyses will be performed using SAS statistical software Version 9.4 or later, unless otherwise noted. Medical history and adverse events will be coded using Medical Dictionary for Regulatory Activities (MedDRA) version 21.0. Concomitant medications will be coded using World Health Organization (WHO) Drug version Global B3 (March 2019).

4.2.3. **Methods of Pooling Data**

For select summaries of adverse events, patients will be presented by background medication usage (No-HU and HU). In these tables, data from Populations A and A1 will be pooled for the No-HU summary, and data from Populations B and B1 will be pooled for the HU summary.

4.2.4. **Adjustments for Covariates**

No formal statistical analysis that adjusts for possible covariate effects is planned.

4.2.5. **Definition of Baseline**

Unless otherwise specified, baseline is defined as the last non-missing measurement prior to the first dose of study drug.

4.2.6. Multiple Comparisons/Multiplicity

Multiplicity is not of concern for this study with a descriptive interpretation.

4.2.7. Subpopulations

No analyses of subgroups of patients are planned.

4.2.8. Withdrawals, Dropouts, Loss to Follow-up

If a patient withdraws from the study, he/she must be provided with a point of contact to obtain further information about the study, if desired. Data and samples collected to the point of withdrawal may only be used after withdrawal if the patient consented to this. Data collected from discontinued patients will be included in the CSR. Available data for all patients will be reported in data listings.

4.2.9. Missing, Unused, and Spurious Data

In general, there will be no substitutions made to accommodate missing data points, except for imputation of partial dates.

4.2.10. Partial and Missing Dates for Adverse Events

4.2.10.1. Partial and Missing Start Dates

If an AE start date is completely missing, and either the available (complete, partial) AE end date/time information indicates that the AE ended before the date/time of first dosing of study treatment on Day 1, or the patient did not receive study treatment, impute the AE start date as the date of informed consent.

If an AE start date is completely missing, the patient received study treatment and either the AE end date information is completely missing or the available AE end date/time information indicates that the AE did not end before the date/time of first dosing of study treatment on Day 1, impute the AE start date to Day 1. The adverse event will be assumed to be treatment-emergent following the first dose of study treatment.

If a partial AE start date is consistent with the actual start date being on Day 1, and either the AE end date information is completely missing, or the complete or imputed AE end date/time information indicates that the AE did not end before Day 1, the imputed AE start date will be Day 1 in the following scenarios:

- The AE start time is missing (the adverse event will be flagged as treatment-emergent following the first dose of study treatment)
- The AE start time is after the date/time of first dosing of study treatment on Day 1 (the adverse event will be flagged as treatment-emergent following the first dose of study treatment)
- The AE start time is before the time of first dosing of study treatment on Day 1, and imputing the AE start date as Day 2 instead of Day 1 would be inconsistent with the partially recorded AE start date (eg, the calendar month for Day 2 is not the same as for Day 1), or with the complete or imputed AE end date/time (in this case, the adverse event will not be flagged as treatment-emergent following the first dose of study treatment).

If none of these conditions are met, the AE start date should be imputed as Day 2 (so that the adverse event will be flagged as treatment-emergent following the first dose of study treatment).

Otherwise, if the start date for an AE is partially recorded, the AE start date will be imputed as follows:

- The first day of the month if day is missing
- January if month is missing
- If only year is recorded, 1 January

If dates imputed to the 1st of the month and/or to the month of January are before the date of informed consent, the AE start date will be re-imputed as the date of the informed consent.

4.2.10.2. Partial and Missing End Dates

If the end date for an AE is completely missing, the AE will be considered ongoing. If the end date for an AE is partially recorded, the AE end date will be imputed as follows:

- The last day of the month if day is missing
- December if month is missing
- If only year is recorded, 31 December

If an AE end date imputed in this way is after the date of the patient's last visit, then the AE end date will be re-imputed as the date of the patient's last visit.

4.2.11. Partial and Missing Dates for Concomitant Medications

4.2.11.1. Partial and Missing Start Dates

If a medication start date is completely missing, the medication start date will be imputed as the date of informed consent. If a partial medication start date is consistent with occurring on Day 1, the patient received study treatment and the available medication end date information indicates that the medication did not end before Day 1, the imputed medication start date will be Day 1. Otherwise, if the medication start date is partially recorded, the medication start date will be imputed as follows:

- The first day of the month if day is missing
- January if month is missing
- If only year is recorded, 1 January

If medication start date imputed in this way is before the date of the patient's informed consent, the medication start date will be re-imputed as the date of the informed consent.

4.2.11.2. Partial and Missing End Dates

If the end date for a medication is completely missing, the end date will be imputed as the date of the patient's last visit. If the end date for a medication is partially recorded, the medication end date will be imputed as follows:

- The last day of the month if day is missing
- December if month is missing
- If only year is recorded, 31 December

If a medication end date imputed in this way is after the date of the patient's last visit, the medication end date will be re-imputed as the date of the patient's last visit.

4.2.12. Visit Windows

The study visit windows are defined in Tables 1 through 4. The accepted window of time for blood draws for PK and PD parameters is shown in Table 5. The actual date and time of each sample will be recorded. Data from all blood draws will be included in data listings. Actual dates and times will be used for pharmacokinetic analyses rather than nominal days and times.

Table 5 Accepted Time Window for PK and PD Blood Draws

Procedure	Time Point	Interval for Analysis
PK and PD	Pre-dose	Within 30 minutes prior to dose
PK	Post-dose time points up through 10 hours post-dose	±5 minutes for each time point
PK	24 hours post-dose	±1 hour for each time point

4.2.13. **Unscheduled Visits**

Data from unscheduled visits will be assigned to a planned visit based on visit windowing. Data summaries will include data from scheduled visits, if available. In the case that data is missing for a planned visit but there is data available from an unscheduled visit that occurred during the visit window, the data from the unscheduled visit will be included in data summaries. Data from both scheduled and unscheduled visits will be included in data listings.

4.3. Interim Analyses

Two interim analyses were planned to review pooled data for specific biomarkers. Statistical considerations for each of the interim analyses are included in two separate interim analysis plans (IAPs).

4.4. Patient Disposition

An overall tabulation of patient disposition will be presented, including the number of patients screened and the number of patients enrolled in Populations A, A1, B, and B1. Tabulations of the number of patients enrolled and treated, the number in each patient population for analysis, the number that completed the study, the number that withdrew prior to completing the study, and reasons for withdrawal will be presented for Populations A, A1, B, and B1. A by-patient listing of study completion information, including the reason for premature study withdrawal, if applicable, will be presented.

4.5. Demographic Characteristics

Demographic information will be summarized for the Safety Analysis Set using descriptive statistics, including age, age category, gender, race and ethnicity. Demographic data will be provided in a data listing.

4.6. Baseline and Sickle Cell Anemia Disease History

Baseline information will be summarized for the Safety Analysis Set using descriptive statistics, including height, weight, BMI and BMI category based on the following categories:

- Underweight (BMI < 18.5 kg/m^2)
- Normal (18.5 kg/m^2 ≤ BMI < 25 kg/m^2)
- Overweight (25 kg/m^2 ≤ BMI < 30 kg/m^2)
- Obese (BMI ≥ 30 kg/m^2)

Sickle cell anemia disease history information will be summarized for the Safety Analysis Set using descriptive statistics, including genotype, use of hydroxyurea (HU), number of hospitalizations for VOCs, and baseline ASCQ-Me SCD Medical History Total Score.

Baseline and sickle cell anemia disease history data will be provided in a data listing.

4.7. General Medical History

General medical history information coded using MedDRA version 21.0 will be summarized for the Safety Analysis Set using descriptive statistics. General medical history data will be provided in a data listing.

4.8. Study Drug Exposure and Compliance

Summaries of exposure to study drug will be presented for the Safety Analysis Set. A summary of the duration of treatment at each dose level will be presented using descriptive statistics, with duration calculated in days as:

Duration = date of last dose-date of first dose+1

Compliance will be derived as a percentage as follows:

Compliance = 100 × number of administered doses ÷ number of expected doses

Compliance will be summarized as a continuous variable, using descriptive statistics

For patients receiving HU, exposure and compliance will be tabulated separately for IMR-687 and HU. Listings of study drug exposure and compliance will be provided.

4.9. Protocol Deviations

The number and percentage of patients with at least one major protocol deviation will be presented for the Safety Analysis Set. Additionally, incidence by category of deviation will be presented. In these tabulations, patients could be counted in more than one category if they have a deviation attributed to multiple categories. All protocol deviations will be presented in a data listing.

4.10. Pharmacokinetic Evaluations

4.10.1. PK Concentration Analysis

Individual plasma IMR 687 and HU concentration-time data will be displayed graphically, summarized and listed. Plasma concentrations will be summarized to 3 significant figures.

Plasma concentration will be summarized by Population, Treatment dose cohort, day/ timepoint including all sample assessments. The following descriptive statistics will be presented for plasma concentrations obtained at each nominal time point (described in Table #): n (number of non-missing observations), arithmetic mean, SD, median, minimum, maximum, geometric mean, geometric CV (GCV), where GCV (%) =SQRT(exp(s^2)-1)*100 and s is the standard deviation of the log-transformed values.

For the tabulation and plotting of the PK concentrations, PK concentrations below the limit of quantification (BLQ) will be replaced with 0.01 prior to the generation of the corresponding descriptive

statistics. When the mean, geometric mean, minimum, median or maximum are BLQ (< 1.00 ng/mL), these stats will be reported as "BLQ" while the SD, %CV and geometric %CV will be reported as not determined (ND). When the geometric mean concentration is BLQ, this will be plotted as zero on the linear plots.

Arithmetic mean plasma concentration will be displayed graphically in linear and semi-log scale. Nominal sampling times will be used in the table summaries and summary figures of plasma concentrations.

Individual patient plasma, concentration versus nominal time plots will be produced in linear and semi-log scale. Per treatment group, dose cohort, day, a plot will be produced for each patient with available individual patient plasma concentration.

4.10.2. **PK Parameter Analysis**

PK parameters of IMR-687 and HU will be analyzed using the assigned doses and actual sampling times. All parameters will be derived using model-independent methods (non-compartmental analysis – NCA) as implemented in Phoenix® WinNonlin® version 8 or higher (Certara USA Inc., Princeton, New Jersey).

Table 6. Planned IMR 687 PK Parameters

Parameter	Definition	Method of Determination	Study Day 1	Mid-Study[a]	End of Therapy
C_{max} (ng/mL)	Maximum observed plasma concentration	Observed Value	X	X	X

T_{max} (hr)	Time to maximum observed plasma concentration	Observed Value	X	X	X
Terminal elimination half-life ($t_{1/2}$) (hr)	Apparent first-order terminal elimination half-life	$\ln(2)/\lambda_z$, where λ_z is the apparent first-order terminal elimination rate constant.	X	X	X
$AUC_{0\text{-last}}$ (hr*ng/mL)	Area under the concentration versus time curve from 0 to the time of the last quantifiable (above LLOQ) sample	The area under the concentration-time curve time 0 to the time of the last quantifiable (above LLOQ) sample will be calculated using the linear up/log down variant of the trapezoidal rule	X	X	X
$AUC_{0\text{-inf}}$ (hr*ng/mL)	Area under the concentration versus time curve from 0 to infinity	Calculated as $AUC_{0\text{-last}} + C_{est,last}/\lambda_z$, where $C_{est,last}$ is the estimated last measurable concentration will be calculated using the	X		

		linear up/log down variant of the trapezoidal rule			
$AUC_{0\text{-}tau}$ (hr*ng/mL)	Area under the concentration versus time curve from 0 to the end of the dosing interval, tau.	The area under the concentration-time curve time 0 to the end of the dosing interval will be calculated using the linear up/log down variant of the trapezoidal rule	X	X	X
R_{ac}	Accumulation Ratio	Mid-Study $AUC_{0\text{-}tau}$/End of Therapy $AUC_{0\text{-}tau}$,			X

[a]Mid-Study: Study Day 85 for population A; Study Day 29 for populations A1, B and B1.

Table 7. Planned HU PK Parameters

Parameter	Definition	Method of Determination
C_{max} (ng/mL)	Maximum observed plasma concentration	Observed Value

T_{max} (hr)	Time to maximum observed plasma concentration	Observed Value
$AUC_{0\text{-last}}$ (hr*ng/mL)	Area under the concentration versus time curve from 0 to the time of the last quantifiable (above LLOQ) sample	The area under the concentration-time curve time 0 to the time of the last quantifiable (above LLOQ) sample will be calculated using the linear up/log down variant of the trapezoidal rule
$AUC_{0\text{-}12}$	Area under the concentration versus time curve from 0 to 12 hours post-dose.	The area under the concentration-time curve time 0 to 12 hours post-dose will be calculated using the linear up/log down variant of the trapezoidal rule

The AUC parameters will be calculated using the linear trapezoidal method for ascending concentrations and the log trapezoidal method for descending concentrations ('linear up, log down') calculation method option in WinNonlin.

Onset of the apparent terminal log-linear phase used in the calculation of λ_z will be determined using WinNonlin Auto Selection and will be reviewed by the pharmacokineticist for potential outliers. No values for $AUC_{0\text{-inf}}$ or Apparent Terminal $T_{1/2}$ will be reported for cases that do not exhibit a terminal log-linear phase in the concentration versus time profile or for profiles where the R^2 value of the regression line through terminal elimination points is less than 0.80.

Parameters "Rsq_adjusted" (adjusted R^2), $AUC_{0\text{-last}}$, and "AUC%extrap_predicted" (predicted percent extrapolated $AUC_{0\text{-inf}}$) will be included as part of WinNonlin parameter output for supportive information on determination of the individual Apparent Terminal $T_{1/2}$ and $AUC_{0\text{-}\infty}$ values, but will not be referenced in the CSR except in cases where the extrapolated $AUC_{0\text{-inf}}$ is ≥ 25% of predicted, when it will be starred with added footnote in the text with justification for inclusion/exclusion and percent extrapolated

All PK parameters will be listed by patient and summarised by treatment, dose, day. The following descriptive statistics will be presented: n (number of non-missing observations), arithmetic mean, SD, SE, median, minimum, maximum, CV, geometric mean, geometric CV. Geometric CV (GCV) will be calculated as GCV%=SQRT(e^{s^2}-1)*100 and *s* is the standard deviation of the log-transformed values. PK parameters will be summarised to 3 significant figures and T_{max} to 2 decimal places.

4.10.3. **Drug-Drug Interaction Assessment**

The effect of co-administration of IMR-687 with HU will be assessed for drug-drug interactions (DDI) using data from populations B and B1. Changes in systemic exposure ($AUC_{0\text{-}12}$, AUC_{last}) and peak exposure (C_{max}) will be assessed by comparing each subject's baseline HU PK profile (i.e. in the absence of IMR 687) to their Day 29 and End of Therapy Profiles. The following tests will be performed:

Table 8. Planned Comparisons of HU Exposure in the Presence and Absence of IMR 687

Comparison	Test	Reference
1	Day 29 HU $AUC_{0\text{-tau}}$	Baseline HU $AUC_{0\text{-tau}}$
2	End of Study HU $AUC_{0\text{-tau}}$	Baseline HU $AUC_{0\text{-tau}}$
3	End of Study HU $AUC_{0\text{-tau}}$	Day 29 HU $AUC_{0\text{-tau}}$

*Similar comparisons will be performed for $AUC_{0\text{-}12}$ and C_{max}

For some subjects, PK profiling of baseline HU exposure will be performed twice. For these subjects, two sets of baseline HU PK parameters will be generated and presented in the PK parameter listings. Because these subjects will be pooled with subjects that only contribute a single baseline HU PK profile,

the replicate PK parameters (i.e. C_{max} and AUC) from both baseline profiles will be averaged so that each subject is only represented once during statistical analysis.

Additionally, because the dose of HU was not uniformly collected during the entirety of study duration, HU doses will be assumed to be consistent with the original dose assignment across the DDI assessments. For these purposes, the baseline dose of HU will be considered the HU dose at the time of test profiling. If it is known that HU doses were up- or down-titrated, or that the dosing scheme was changed between the test and reference profiles, the subject will be considered inevaluable and excluded from the primary statistical analysis.

A secondary analysis of dose-normalized values may be performed to assess the impact of these subjects on interpretation of the results.

The effect of multiple-dose administration of IMR-687 on the systemic exposure to HU will be evaluated using generalized linear model (GLM) procedures in SAS®, comparing IMR-687 + HU at the end of study to reference and with Subject as a random factor on log-transformed AUC_{0-t} at the alpha level of 0.05. The Test effect will be tested against the residual mean square error. All sums of squares (Types I, II, III and IV) will be reported. Probability (p) values will be derived from Type III sums of squares. Intra- and Inter-subject coefficients of variation will be estimated.

Based on pairwise comparison of the log-transformed AUC_{0-t} data for HU, the ratios (Test/Ref) of the geometric least-squares means, calculated according to the formula “exp(X-Y) * 100”, as well as the corresponding 90% confidence intervals for geometric means, will be determined.

Additionally, because HU is a titratable drug and dosing information was not uniformly collected, the results of the planned statistical comparisons will be considered exploratory in nature and no hypothesis testing will be performed.

Boxplots representing the geometric mean ratios for Cmax or $AUC_{0\text{-}tau}$ for HU in the presence and absence of IMR-687 in population B and B1 will be generated to assist evaluation of the potential for meaningful DDI

Additionally, the current study design does not support direct comparisons of IMR-687 exposure in the presence and absence of HU. However, visualizations of Day 1 PK profiles for 100 mg in Population A and A1 vs. steady state PK profiles (100 mg only) in Population B and B1 may be prepared to help assess any potential effects of HU administration on the PK of IMR-687.

4.11. Pharmacodynamic Evaluations

The following endpoints will be evaluated to assess the PD activity of IMR-687:

- Total haemoglobin (Hb) level
- HbF value (%)
- % F cells
- Indices of red cell haemolysis (indirect bilirubin, reticulocyte count [% and absolute], lactate dehydrogenase [LDH], and haptoglobin levels)
- sE-Selectin, sP-Selectin, sICAM-1 and sVCAM-1
- High-sensitivity C-reactive protein (hs-CRP)
- Mean corpuscular volume (MCV)
- Myeloperoxidase (MPO)
- N-terminal prohormone of brain natriuretic peptide (NT-proBNP)

In analyses of PD parameters, baseline is defined as the average of non-missing values from the screening and baseline visit. The end of end-of-study endpoint is the average of non-missing values from Week 21 and Week 25. PD endpoints will be summarized for the PD Evaluable Set using descriptive statistics for the observed value and change from baseline at each scheduled study visit. Figures displaying mean value and mean change from baseline for PD endpoints over time will also be generated, as well as bar plots of mean baseline and end-of study values. A by-patient listing of PD

endpoints will be generated. A sensitivity analyses of PD parameters will be performed with baseline defined as the value from baseline visit.

4.12. Clinical Outcome Measures

4.12.1. ASCQ-Me Outcomes

ASCQ-Me Outcomes will be analyzed, including:

- Emotional impact
- Pain impact
- Sleep impact
- Social functioning impact
- Stiffness impact
- Pain frequency and severity
- SCD medical history checklist

The emotional impact, pain impact, sleep impact, social functioning impact, and stiffness impact assessments will be analyzed similarly. For each patient, a total score will be calculated for each ASCQ-Me outcome type by summing the raw scores for each question and converting the total to a t-score based on the tables in Appendix A. Continuous t-scores will be converted to categorical severity classes based on the tables in Appendix B. Continuous summaries of t-scores and categorical summaries of severity classes will be generated at each scheduled visit based on the FAS using descriptive statistics. Bar plots of mean values at baseline and end-of study will be generated.

The pain frequency and severity will be analyzed by deriving two separate composite scores. The frequency composite score represents the sum of the values for the first questions in the questionnaire and the severity score composite represents the sum of the values for the other three items. The

composite scores are standardized by calculating z-scores. Each patient's z-score for each component score is derived by subtracting the mean of the score distribution from the individual score and dividing by the standard deviation of the score distribution. The score is then transformed by multiplying by 10 and adding 50 (so that the range of scores is greater than 1).

The distribution means and standard deviations are as follows:

Score Type	Mean	Standard Deviation
Frequency	7.525	2.573
Severity	15.018	4.275

Continuous z-scores will be converted to severity classes based on the tables in Appendix B. Continuous summaries of z-scores and categorical summaries of severity classes will be generated at each scheduled visit based on the FAS using descriptive statistics.

A numerical score for the SCD medical history checklist is derived by summing the number of positive responses. Continuous summaries of composite scores for SCD medical history checklist will be generated at each scheduled visit based on the FAS using descriptive statistics.

A by-patient listing of ASCQ-Me outcome measures will be generated.

4.12.2. **Additional SCA-Related Clinical Outcomes**

The following SCA-related clinical outcome measures will be evaluated to assess the potential efficacy of IMR-687:

Pain-related measures, as captured using an app-based daily questionnaire, including:

- Frequency and severity of pain
- Frequency and severity of fatigue
- Impact of pain/fatigue on work/school and on activities of daily living (ADL)

- Need for professional medical attention due to sickle cell pain
- Use of medication for sickle cell pain

Continuous and categorical summaries of SCA-related outcomes will be generated at each scheduled visit based on the FAS using descriptive statistics. A by-patient listing of SCA-related outcome measures will be generated.

VOCs will be summarized using descriptive statistics, including number of events of acute painful crisis and frequency of acute chest symptoms, including fever, cough, sputum production, shortness of breath, tachypnea, hypoxia, chest pain. Results of x-rays will also be summarized, as well as the primary setting for VOC treatment. A by-patient listing of VOC data will also be generated, including the use of pain medications, including opioids.

4.13. **Safety Analyses**

Safety analyses will be conducted using the Safety Analysis Set.

4.13.1. **Adverse Events**

Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA) and displayed in tables and listings using System/Organ/Class (SOC) and Preferred Term.

An SAE is any AE that results in one or more of the following outcomes:

- Death
- Requires or prolongs hospitalization
- Is life-threatening
- Persistent or significant disability/incapacity
- Congenital anomaly or birth defect

- Other medically important event

Any AE, including any SAE, that occurs after the patient has signed informed consent but prior to assignment of a patient identification number will be recorded on the medical history page of the eCRF. AEs that occur following assignment of a patient identification number (randomization), including those that may occur prior to administration of study drug, will be recorded on the AE page(s) of the eCRF. AEs that occur following administration of study drug will be considered treatment emergent adverse events (TEAEs). Missing or partial start and end dates will be imputed as described in Section 4.2.10 before determining whether an AE is treatment emergent. Adverse events (related and unrelated), including SAEs, will be recorded from the signing of informed consent through the end-of-study safety follow-up visit.

AE severity (intensity) will be graded using the Common Terminology Criteria for Adverse Events (CTCAE) version 4.0, as follows:

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

The investigator will assess the potential relatedness of each AE to the investigational product. An investigator causality assessment (Unrelated, Unlikely, Possible, Probable, or Definite) must be provided for all AEs (both serious and non-serious), as follows:

- Not Related/Unrelated
- Unlikely Related
- Possibly Related:
- Probably Related:
- Definitely Related:

Outcome describes the status of the AE. Once the outcome is clear or at the end of the study, the investigator assigns one of the following outcomes for each AE: fatal, not recovered/not resolved, recovering/resolving, recovered/resolved, recovered/resolved with sequelae, or unknown.

Adverse events are summarized by patient incidence rates, therefore, in any tabulation, a patient contributes only once to the count for a given adverse event (SOC or preferred term). The total number of events and the number and percentage of patients with any treatment-emergent adverse event, with any treatment-emergent adverse events assessed by the Investigator as related to treatment (definite, probable, or possible relationship), with any treatment-emergent adverse event leading to study discontinuation, and with any serious adverse event will be summarized by treatment group and overall. In these tabulations, each patient will contribute only once (i.e., the most related occurrence or the most intense occurrence) to each of the incidence rates in the descriptive analysis, regardless of the number of episodes. No formal hypothesis-testing analysis of adverse events incidence rates will be performed for summaries of adverse events. All adverse events occurring on study will be listed in patient data listings. By-patient listings also will be provided for the following: patient deaths; serious adverse events; and adverse events leading to withdrawal.

4.13.2. **Laboratory Data**

Samples for haematology, clinical chemistry, coagulation, and urinalysis will be collected as outlined in the Schedules of Assessments in Table 1 (Population A), Table 2 (Population A1), Table 3 (Population B),

and Table 4 (Population B1). A list of parameters included in each of these categories is presented in Table 6. Laboratory tests may be repeated at any visit if there was an abnormal finding at the most recent previous evaluation or if additional information is clinically necessary to appropriately evaluate the patient's current condition, follow up, and/or manage an AE.

Table 6: Safety Laboratory Parameters

Chemistry:	Sodium, potassium, calcium, chloride, bicarbonate, magnesium, phosphate, glucose, BUN, creatinine, CPK, total protein, ALP, LDH, albumin, bilirubin (total; direct; indirect), ALT, AST, and GGT
Haematology:	Absolute and differential WBC count, erythrocyte count, reticulocyte count, Hb (including HbF), haematocrit, platelet count, and RBC indices (mean corpuscular volume, mean corpuscular haemoglobin, and mean corpuscular haemoglobin concentration)
Coagulation:	Prothrombin time (PT) and activated partial thromboplastin time (aPTT)
Urinalysis:	Specific gravity, pH, protein, glucose, bilirubin, urobilinogen, ketones, and blood

ALP = alkaline phosphatase; ALT = alanine aminotransferase; aPTT = activated partial thromboplastin time;
AST = aspartate aminotransferase; BUN = blood urea nitrogen; CPK = creatinine phosphokinase;
GGT = gamma-glutamyl transferase; Hb = haemoglobin; HbF = fetal haemoglobin; LDH = lactate dehydrogenase;
PT = prothrombin time; RBC = red blood cell; WBC = white blood cell

Clinical laboratory values will be expressed using SI units. The actual value and change from baseline at scheduled study visits will be summarized for each clinical laboratory parameter, including haematology and clinical chemistry using descriptive statistics. In the event of repeat values, the last non-missing value per study day/time will be used. Separate data listings for chemistry, haematology, coagulation and urinalysis will be generated.

4.13.3. **Vital Signs**

Vital signs should be measured in the sitting or semi-supine position, on the days and time points indicated in the Schedules of Assessments in Table 1 (Population A), Table 2 (Population A1) Table 3 (Population B), and Table 4 (Population B1). Vital signs will include systolic and diastolic blood pressure

(mmHg), heart rate (beats/minute), respiratory rate (breaths/minute), temperature (°C), and weight (kg).

The actual value and change from baseline at scheduled study visits will be summarized for vital signs using descriptive statistics. All vital signs data will be provided in data listings.

4.13.4. **Physical Examinations**

A complete physical examination will be performed at the Screening and Day 1 visits as outlined in the Schedules of Assessments in Table 1 (Population A), Table 2 (Population A1), Table 3 (Population B), and Table 4 (Population B1). All physical examination findings will be presented in a data listing.

4.13.5. **Electrocardiogram**

A standard 12-lead ECG will be performed in triplicate predose and postdose on the days and time points indicated in the Schedules of Assessments in Table 1 (Population A), Table 2 (Population A1), Table 3 (Population B), and Table 4 (Population B1). The average of triplicate values by day and time point will be included in the summary table. ECG results will be summarized descriptively for quantitative measures such as ventricular heart rate, PR interval, RR interval, QRS interval, QT interval, and QTcF interval. Categorical summaries of the number and percent of patients with normal, not clinically significant abnormal, and clinically significant abnormal results will also be presented at baseline and each study visit. All ECG data for each patient (including triplicate measurements), as well as the average by day and time point, will be provided in data listings.

4.13.6. **Concomitant Medications**

Any medication or vaccine (including over-the-counter or prescription medicines, vitamins, and/or herbal supplements) that the patient receives from the time of enrollment through the end of the study must be recorded along with reason for use, dates of administration, including start date/time and stop date/time, and dose and frequency of administration.

If the end date for use of a medication is before the date of the first dose of study treatment, it will be considered a prior medication. If the start date for use of a medication is after the date of the first dose of study treatment, or if the start date is prior to first dose date but the end date is after the first dose of study treatment or the medication use is ongoing during study treatment, it will be considered a concomitant medication.

The use of prior and concomitant medications will be included in by-patient data listing. For Populations B and B1, separate listings for HU and non-HU medications will be generated.

5. CHANGES TO PLANNED ANALYSES

The following PD parameters are not currently in listed in the protocol but will be analyzed as described in Section 4.12:

- Myeloperoxidase (MPO)
- N-terminal prohormone of brain natriuretic peptide (NT-proBNP)
- VCAM

6. REFERENCES

American Institutes for Research. ASCQ-Me Scoring Manual. http://www.healthmeasures.net/images/ASQMe/ASCQ-Me_Scoring_Manual.pdf

Charache S, Terrin ML, Moore RD, Dover GJ, Barton FB, et al. Effect of HU on the frequency of painful crises in sickle cell anaemia. Investigators of the Multicenter Study of Hydroxyurea in Sickle Cell Anemia. N Engl J Med. 1995;332(20):1317-22.

Cokic VP, Smith RD, Beleslin-Cokic BB, Njoroge JM, Miller JL, Gladwin MT, et al. Hydroxyurea induces foetal haemoglobin by the nitric oxide-dependent activation of soluble guanylyl cyclase. J Clin Invest. 2003;111(2):231-9.

Erard F, Dean A, Schechter AN. Inhibitors of cell division reversibly modify haemoglobin concentration in human erythroleukemia K562 cells. Blood. 1981 Dec;58(6):1236-9.

Johnson JJ. Use of pharmacokinetic data below lower limit of quantitation values. Pharm Res. 2012 Sep; 29(9): 2628–2631.

Keller S, Yang M, Treadwell MJ and Hassell KL. Sensitivity of alternative measures of functioning and wellbeing for adults with sickle cell disease: comparison of PROMIS® to ASCQ-Me. Health and Quality of Life Outcomes. 2017 15(117): 1-14.

Lanzkron S, Carroll CP, Haywood C Jr. Mortality rates and age at death from sickle cell disease: U.S., 1979-2005. Public Health Rep. 2013 Mar-Apr;128(2):110-6. PMID: 23450875.

Paulukonis ST, Eckman JR, Snyder AB, Hagar W, Feuchtbaum LB, Zhou M, Grant AM, Hulihan MM. Defining sickle cell disease mortality using a population-based surveillance system, 2004through 2008. Public Health Rep. 2016 Mar-Apr;131(2):367-375. PMID: 26957672.

Platt OS, Orkin SH, Dover G, Beardsley GP, Miller B, Nathan DG. Hydroxyurea enhances foetal haemoglobin production in sickle cell anaemia. J Clin Invest. 1984 Aug;74(2):652-6.

Platt OS, Brambilla DJ, Rosse WF, Milner PF, Castro O, Steinberg MH, et al. Mortality in sickle cell disease. Life expectancy and risk factors for early death. N Engl J Med.1994;330(23):1639-44.

7. APPENDIX A: ASCQ-ME SCORING TABLES

Emotional Impact Short Form Conversion Table		
Raw Score	**T-Score**	**SE[a]**
5	26.8	4.5
6	30.8	3.5
7	33.3	3.1
8	35.3	2.9
9	37.0	2.8
10	38.5	2.7
11	39.9	2.6
12	41.2	2.6
13	42.5	2.6
14	43.7	2.6
15	44.9	2.6
16	46.2	2.7
17	47.4	2.7
18	48.7	2.8
19	50.1	2.8
20	51.5	3.0
21	53.3	3.3
22	55.2	3.6
23	57.3	3.8
24	60.5	4.4
25	65.6	5.8

[a]SE = Standard Error for T-Score

Social Functioning Impact Short Form Conversion Table		
Raw Score	**T-Score**	**SE[a]**
5	26.0	4.3
6	29.8	3.2
7	32.5	2.8
8	34.7	2.8
9	36.8	2.7
10	38.7	2.7
11	40.4	2.7
12	42.1	2.7
13	43.9	2.6
14	45.6	2.6
15	47.2	2.6
16	48.8	2.6
17	50.5	2.6
18	52.2	2.5
19	54.0	2.5
20	55.8	2.5
21	57.7	2.5
22	59.8	2.6
23	62.1	2.7
24	64.9	3.1
25	69.8	4.6

[a]SE = Standard Error for T-Score

Pain Short Form Conversion Table		
Raw Score	**T-Score**	**SE[a]**
5	24.8	3.9
6	28.8	2.5
7	31.0	2.2
8	33.0	2.2
9	34.9	2.2
10	36.7	2.2
11	38.3	2.2
12	39.9	2.1
13	41.5	2.1
14	43.0	2.1
15	44.4	2.1
16	45.7	2.1
17	47.1	2.1
18	48.5	2.0
19	49.9	2.0
20	51.2	2.0
21	52.5	2.0
22	54.0	2.1
23	55.8	2.3
24	58.0	2.8
25	63.8	5.2

[a]SE = Standard Error for T-Score

8. APPENDIX B: ASCQ-ME SEVERITY GROUP DEFINITIONS

ASCQ-Me														
SCDMHC Normative	Sample Mean[1]			LS Mean[3]			Tertile	Mean	Standard	Severity Cutoff[4]		Severity Distribution		
Short Form	Severity Group			Severity Group			Cutoff	Tertile	Cutoff	High	Low	Severity Group		
(Higher score is better)	High	Middle	Low	High	Middle	Low	T	C	T/C	Q1	Q3	High	Middle	Low
Emotional	47.04	49.75	51.84	47.14	49.54	51.94	0.4307	1.0908	0.3949	48.60	50.49	< 48.60	[48.60, 50.49]	> 50.49
Social Functioning	46.72	48.91	52.72	46.45	49.45	52.45	0.4307	1.0908	0.3949	48.27	50.63	< 48.27	[48.27, 50.63]	> 50.63
Pain	46.30	48.88	53.26	46.00	49.48	52.96	0.4307	1.0908	0.3949	48.11	50.85	< 48.11	[48.11, 50.85]	> 50.85
Stiffness	45.81	48.81	53.80	45.48	49.47	53.47	0.4307	1.0908	0.3949	47.90	51.05	< 47.90	[47.90, 51.05]	> 51.05
Sleep	48.10	48.79	52.24	47.64	49.71	51.78	0.4307	1.0908	0.3949	48.89	50.53	< 48.89	[48.89, 50.53]	> 50.53
SCDMHC Normative	Sample Mean[1]			LS Mean[3]			Tertile	Mean	Standard	Severity Cutoff[4]		Severity Distribution		
Pain Episode	Severity Group			Severity Group			Cutoff	Tertile	Cutoff	High	Low	Severity Group		
(Lower score is better)	Low	Middle	High	Low	Middle	High	T	C	T/C	Q1	Q3	Low	Middle	High
Frequency	48.31	50.33	51.99	48.37	50.21	52.05	0.4307	1.0908	0.3949	49.48	50.94	< 49.48	[49.48, 50.94]	> 50.94
Severity	48.38	50.09	52.10	48.33	50.19	52.05	0.4307	1.0908	0.3949	49.46	50.92	< 49.46	[49.46, 50.92]	> 50.92
PROMIS General Health	Sample Mean[2]			LS Mean[3]			Tertile	Mean	Standard	Severity Cutoff[4]		Severity Distribution		
Pain Episode	Very Good		Poor	Very Good		Poor	Cutoff	Tertile	Cutoff	Low	High	Very Good		Poor
(Lower score is better)	/Excellent	Good	/Fair	/Excellent	Good	/Fair	T	C	T/C	Q1	Q3	/Excellent	Good	/Fair
Frequency	44.6	49.1	52.8	44.7	48.8	52.9	0.4307	1.0908	0.3949	47.21	50.45	< 47.21	[47.21, 50.45]	> 50.45
Severity	45.2	48.6	52.9	45.1	48.9	52.8	0.4307	1.0908	0.3949	47.38	50.42	< 47.38	[47.38, 50.42]	> 50.42
PROMIS Overall Quality of Life	Sample Mean[2]			LS Mean[3]			Tertile	Mean	Standard	Severity Cutoff[4]		Severity Distribution		
Pain Episode	Very Good		Poor	Very Good		Poor	Cutoff	Tertile	Cutoff	Low	High	Very Good		Poor
(Lower score is better)	/Excellent	Good	/Fair	/Excellent	Good	/Fair	T	C	T/C	Q1	Q3	/Excellent	Good	/Fair
Frequency	47.7	50.3	52.2	47.8	50.1	52.3	0.4307	1.0908	0.3949	49.18	50.96	< 49.18	[49.18, 50.96]	> 50.96
Severity	48.0	49.6	52.4	47.8	50.0	52.2	0.4307	1.0908	0.3949	49.13	50.87	< 49.13	[49.13, 50.87]	> 50.87

[1] From Table 4 of Keller et al. Sensitivity of alternative measures of functioning and wellbeing for adults with sickle cell disease: comparison of PROMIS to ASCQ_Me. Health and Quality of Life Outcomes (2017) 15:117

[2] From Exhibits 4-3 and 4-4 of ASCQ-Me User's Manual (December 2017)

[3] Least squares means were calculated as follows: The least squares mean for the Middle (Good) group is the average of the three sample means. The adjusted means for the High (Poor/Fair) and Low (Very Good/Excellent) groups are derived so that: (1) the average of the 3 adjusted means is the same as the average of the 3 sample means, and (2) the adjusted mean of the middle group is equidistance from both adjusted means of the other two groups.

[4] Cutoff points (Q1 and Q3) for the SCDMHC were based on tertiles of the distribution of scores.

Note: SCD-MHC scores were the sum of the number conditions checked. Cutoffs for low, medium and high groups were SCD-MHC scores less than 2, equal to 2, and greater than 2, respectively.

9. APPENDIX C: DETERMINING CUT-OFF POINTS BETWEEN SEVERITY GROUPS FOR ASCQ-ME

Consider X a normal distribution with mean μ and standard deviation σ, *i.e.* $X \sim N(\mu,\sigma)$. Let t_1 and t_2 denote the lower and upper tertiles of the standard normal distribution, respectively. Then $t_1 = \Phi^{-1}(1/3)$, $t_2 = \Phi^{-1}(2/3)$ and $t_1 = -t_2 \approx 0.4307$. Let X_1, X_2 and X_3 be independent random variables from the first, second and third tertile groups of $N(\mu,\sigma)$, respectively. That is, X_1, X_2 and X_3, respectively, have density functions

$$f_1(x) = \frac{3}{\sqrt{2\pi}} e^{-x^2/2} \text{ for } -\infty < x < \mu + \Phi^{-1}(1/3)\sigma,$$

$$f_2(x) = \frac{3}{\sqrt{2\pi}} e^{-x^2/2} \text{ for } \mu + \Phi^{-1}(1/3)\sigma < x < \mu + \Phi^{-1}(2/3)\sigma$$

and $$f_3(x) = \frac{3}{\sqrt{2\pi}} e^{-x^2/2} \text{ for } \mu + \Phi^{-1}(2/3)\sigma < x < \infty.$$

Let $\overline{X}_1$, $\overline{X}_2$ and $\overline{X}_3$ be sample means of the first, second and third tertiles, respectively. Then

$$E(\overline{X}_1) = \mu + \sigma \int_{-\infty}^{t_1} \frac{3t}{\sqrt{2\pi}} e^{-t^2/2} dt = \mu - [\frac{3}{\sqrt{2\pi}} e^{-(\Phi^{-1}(1/3))^2/2}]\sigma,$$

$$E(\overline{X}_2) = \mu + \sigma \int_{t_1}^{t_2} \frac{3t}{\sqrt{2\pi}} e^{-t^2/2} dt = \mu$$

and $$E(\overline{X}_3) = \mu + \sigma \int_{t_2}^{-\infty} \frac{3t}{\sqrt{2\pi}} e^{-t^2/2} dt = \mu + [\frac{3}{\sqrt{2\pi}} e^{-(\Phi^{-1}(1/3))^2/2}]\sigma.$$

Define the sum of squared errors as

$$f(\mu,\sigma) = (\overline{X}_1 - \mu + c\sigma)^2 + (\overline{X}_2 - \mu_2)^2 + (\overline{X}_3 - \mu - c\sigma)^2,$$

where $c = \frac{3}{\sqrt{2\pi}} e^{-(\Phi^{-1}(1/3))^2/2} \approx 1.0908$.

The critical point for $f(\mu,\sigma)$ satisfies $\frac{\partial f}{\partial \mu}(\mu,\sigma) = 0$ and $\frac{\partial f}{\partial \sigma}(\mu,\sigma) = 0$. That is

$$\frac{\partial f}{\partial \mu}(\mu,\sigma) = 2(\mu - c\sigma - \overline{X}_1) + 2(\mu - \overline{X}_2) + 2(\mu + c\sigma - \overline{X}_3) = 6(\mu - \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3}) = 0$$

and $$\frac{\partial f}{\partial \sigma}(\mu,\sigma) = -c(\mu - c\sigma - \overline{X}_1) + c(\mu + c\sigma - \overline{X}_3) = 2c^2(\sigma - \frac{\overline{X}_3 - \overline{X}_1}{2c}) = 0.$$

Hence $\hat{\mu} = \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3}$ and $\hat{\sigma} = \frac{\overline{X}_3 - \overline{X}_1}{2c}$.

The determinant of the Hessian matrix of second-order derivatives of $f(\mu,\sigma)$ at the critical point $(\hat{\mu}, \hat{\sigma})$ is

$$D = \frac{\partial^2 f}{\partial \mu^2}(\hat{\mu},\hat{\sigma}) \frac{\partial^2 f}{\partial \sigma^2}(\hat{\mu},\hat{\sigma}) - [\frac{\partial^2 f}{\partial \mu \partial \sigma}(\hat{\mu},\hat{\sigma})]^2 = 6 \times 2c^2 - 0 = 12c^2 > 0.$$

Therefore, the minimum sum of squared errors is attained at $(\frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3}, \frac{\overline{X}_3 - \overline{X}_1}{2c})$. The least squares means of first, second and third tertiles, respectively, are

$$\hat{\mu} - c\hat{\sigma} = \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3} - \frac{\overline{X}_3 - \overline{X}_1}{2} = \overline{X}_1 + (\overline{X}_2 - \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3})/2,$$

$$\hat{\mu} = \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3}$$

and $$\hat{\mu} + c\hat{\sigma} = \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3} + \frac{\overline{X}_3 - \overline{X}_1}{2} = \overline{X}_3 + (\overline{X}_2 - \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3})/2.$$

Hence, the least squares estimates of lower and upper tertiles of $N(\mu,\sigma)$ are

$$\hat{\mu} + t_1\hat{\sigma} = \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3} + \frac{t_1}{c}(\frac{\overline{X}_3 - \overline{X}_1}{2}) \approx \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3} - 0.3949 \times (\frac{\overline{X}_3 - \overline{X}_1}{2})$$

and $$\hat{\mu} + t_2\hat{\sigma} = \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3} + \frac{t_2}{c}(\frac{\overline{X}_3 - \overline{X}_1}{2}) \approx \frac{\overline{X}_1+\overline{X}_2+\overline{X}_3}{3} + 0.3949 \times (\frac{\overline{X}_3 - \overline{X}_1}{2}).$$