Clinical Study Protocol

Protocol Title:	A Prospective, Open-Label, Single-Arm Clinical Trial to Assess the Anti-Hepatitis A Virus (HAV) Antibody Levels, Pharmacokinetics, and Safety of a Single Intramuscular Dose of a Polyvalent Human Immune Globulin in HAV-Seronegative Healthy Subjects
Investigational Product:	GamaSTAN® [Immune Globulin (Human)]
Sponsor's Name and Address:	Grifols Therapeutics LLC 79 TW Alexander Drive Research Triangle Park, NC 27709 USA
Sponsor's Telephone Number:	[redacted]
Study Number/Protocol Version Number/Date:	GC1703/Protocol Version 5.0/22 Jan 2018 includes GC1703/Protocol Version 4.0/30 Nov 2017, GC1703/Protocol Version 3.0/02 Nov 2017, GC1703/Protocol Version 2.0/20 Oct 2017, and GC1703/Protocol Version 1.0/21 Aug 2017
Additional Identifier:	GamaSTAN PK Study
Development Phase:	Phase IV

The undersigned confirm that they agree to conduct the study under the conditions described in this protocol:

Clinical Assessment Monitor: [redacted]

Signature: [redacted] Date: 23/Jan/18

Confidentiality Statement: *The following confidential information is the property of Grifols. As long as the information contained in this protocol has not been published, it may only be used after permission has been obtained from Grifols. It is not possible to make reproductions of all or sections of this protocol. Commercial use of the information is only possible with the permission of the proprietor and is subject to a license fee.*

Summary of Changes for Amendment 4

Protocol Version	Date of Approval
5.0 Amendment 4 + Integrated Protocol	22 Jan 2018
4.0 Amendment 3 + Integrated Protocol	30 Nov 2017
3.0 Amendment 2 + Integrated Protocol	02 Nov 2017
2.0 Amendment 1 + Integrated Protocol	20 Oct 2017
1.0 Original	21 Aug 2017

Amendment 4

The protocol for GC1703 (Version 4.0, dated 30 Nov 2017) has been amended and reissued as Protocol Amendment 4, Version 5.0, dated 22 Jan 2018.

Summary of Changes for Amendment 4

(Note: Administrative changes including minor administrative corrections and the changes in the protocol synopsis are not included in Protocol Summary of Changes.)

Sections	Change From: (Version 4.0, dated 30 Nov 2017)	Change To: (Version 5.0, dated 22 Jan 2018)	Rationale:
Global	Grifols Therapeutics Inc	Grifols Therapeutics LLC	Company name change
Synopsis, 5.1	Inclusion Criteria: 2. Subjects with a body mass index (BMI) of 18.5 to ~~25.0~~ kg/m^2	Inclusion Criteria: 2. Subjects with a body mass index (BMI) of 18.5 to <u>29.9</u> kg/m^2	To allow enrollment of subjects with a larger body mass index
4.3.1	Subject Numbering Subjects in the study will receive a consecutive subject number. Subject numbers are generated beginning with the study center number (3 digits, assigned by the sponsor) followed consecutively with a unique number for each subject (4 digits, including leading zeros). For example, if the Investigator's center number is 301, subject number will be 3010001, 3010002, 3010003, etc., in consecutive order. Subject numbers, once assigned, will not be reused.	Subject Numbering Subjects in the study will receive a consecutive subject number. Subject numbers are generated beginning with the study center number (3 digits, assigned by the sponsor) followed consecutively with a unique number for each subject (4 digits, including leading zeros). For example, if the Investigator's center number is 301, subject number will be 3010001, 3010002, 3010003, etc., in consecutive order. Subject numbers, once assigned, will not be reused. <u>If a subject is re-screened, a different subject number will be assigned to this subject.</u>	To provide the procedure for assigning subject numbers to rescreened subjects.
5.3.1	Screen Failures Screening evaluations will be used to determine the eligibility of each subject for enrollment. Subjects who fail to meet eligibility criteria during screening evaluations will be considered screen failures and will not participate in the study.	Screen Failures Screening evaluations will be used to determine the eligibility of each subject for enrollment. Subjects who fail to meet eligibility criteria during screening evaluations will be considered screen failures and will not participate in the study. <u>However, subjects who fail the first screening evaluation are allowed to be re-screened once.</u>	To allow rescreening of subjects who previously fail the first screening evaluation.

Investigator Signature Page

The undersigned confirms that he/she agrees to conduct the study under the conditions described in this protocol and comply with the International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) Good Clinical Practice (GCP) and all applicable regulatory requirements:

[redacted] USA

INVESTIGATOR NAME (Please Print) LOCATION

[redacted] 24 JAN 2018

INVESTIGATOR SIGNATURE DATE

Title of the Investigator: Principal Investigator

Address of the Site: [redacted]

Telephone Number: [redacted]

PROTOCOL SYNOPSIS

Title of Study:

A Prospective, Open-Label, Single-Arm Clinical Trial to Assess the Anti-Hepatitis A Virus (HAV) Antibody Levels, Pharmacokinetics, and Safety of a Single Intramuscular Dose of a Polyvalent Human Immune Globulin in HAV-Seronegative Healthy Subjects

Short Title:

GamaSTAN PK Study

Study Number:

GC1703

Phase:

Phase IV

Study Objectives:

Primary Efficacy Objective:

To evaluate whether a single 0.2 mL/kg intramuscular (IM) dose of the study treatment will provide protective levels of antibodies to HAV (anti-HAV) in HAV-seronegative healthy subjects for up to 60 days.

Pharmacokinetic (PK) Objectives:

To evaluate the PK parameters of anti-HAV antibodies following a single 0.2 mL/kg IM dose of the study treatment in HAV-seronegative healthy subjects.

The PK parameters of interest are:

- Area under the plasma and/or serum concentration time curve extrapolated to infinity ($AUC_{0-\infty}$)
- Cumulative area under the plasma and/or serum concentration time curve calculated from 0 to time of last observed quantifiable plasma and/or serum concentration (AUC_{0-T})
- Maximum observed plasma and/or serum concentration (C_{max})
- Time of maximum observed plasma and/or serum concentration (T_{max})
- Apparent elimination rate constant (λ_z)
- Terminal elimination half-life (T_{half})
- Apparent total plasma and/or serum clearance (Cl_{TOT}/F)
- Apparent volume of distribution (V_D/F)

Safety Objective:

To evaluate the safety and tolerability of a single 0.2 mL/kg IM dose of the study treatment in HAV-seronegative healthy subjects.

Overall Study Design and Description:

This is a single center, open-label, single-arm study. Approximately 28 HAV-seronegative healthy subjects will be enrolled in this study after obtaining their written informed consents. There will be a screening period of up to 28 days during which subjects will be screened for enrollment in the study.

The healthy subjects will receive a single IM dose of GamaSTAN (0.2 mL/kg), which is followed by a PK sampling period of 150 days (approximately 5 half-lives). The protective levels of anti-HAV antibodies will be assessed up to 60 days after the administration of GamaSTAN. A PK curve will be obtained during the PK sampling period.

Number of Subjects Planned:

Approximately 28 HAV-seronegative healthy subjects will be enrolled in the study which will provide about 20 evaluable subjects based on approximately 30% dropout rate.

Diagnosis and Main Criteria for Inclusion:

Only HAV-seronegative healthy adult subjects will be enrolled in this study.

Inclusion Criteria:

A subject must meet all the following inclusion criteria to be eligible for participation in this study.

1. Male subjects from 18 to 55 years of age, inclusive, or female subjects from 18 to 65 years of age, inclusive
2. Subjects with a body mass index (BMI) of 18.5 to 29.9 kg/m^2
3. Body weight greater than or equal to 50 kg at screening
4. Subjects willing and able to provide written informed consent
5. Subjects in good health in the judgment of the Investigator, as determined by medical history, physical examination, vital signs, ECG and laboratory assessments
6. A female study subject must meet one of the following criteria:
    a. If a female of childbearing potential – agrees to use one of the accepted contraceptive regimens from at least 30 days prior to study treatment administration and during the entire study duration. An acceptable method of contraception includes one of the following:
        - Abstinence from heterosexual intercourse (i.e. when abstinence is the preferred and usual lifestyle of the subject; periodic abstinence is not acceptable)
        - Non-estrogen containing hormonal contraceptives (birth control pills, injectable/implant/insertable hormonal birth control products, transdermal patch)
        - Intrauterine device without hormones
        - Condom with spermicide
        - Diaphragm or cervical cap with spermicide
        - Vasectomized partner (minimum 6 months since vasectomy prior to study treatment administration)

b. If a female of non-childbearing potential – should be surgically sterile (i.e. has undergone complete hysterectomy, bilateral oophorectomy, or tubal ligation) or in a menopausal state (at least 1 year without menses prior to study treatment administration)

7. A male study subject must agree to use one of the accepted contraceptive regimens during the entire study duration;
    - Abstinence from heterosexual intercourse
    - Female partner with condom with spermicide used by male study subject
    - Female partner of non-childbearing potential
    - Male sterilization (if proof of sterilization is not provided, the subject must agree to use one of the above accepted contraceptive methods)
8. A male study subject must agree not to impregnate a female or donate sperm during the entire study duration

Exclusion Criteria:

A subject meeting any of the following exclusion criteria is NOT eligible for participation in the study.

1. Subject vaccinated against HAV, as documented the in medical history at the screening visit
2. Subject positive for anti-HAV antibodies in blood sample at the screening visit
3. Subject who previously received any type of immune globulin (IG), including HAV IG within the past 12 months prior to study treatment administration
4. Subject with prolonged International Normalized Ratio (INR) or activated partial thromboplastin time (aPTT) at the screening visit
5. Subject with a platelet count below 100×10^9/L at the screening visit
6. Subject suffering from some acute or chronic medical, surgical or psychiatric significant condition or laboratory abnormality at the screening visit or prior to study treatment administration that, according to Investigator judgement, may increase the risk associated with study participation or study treatment administration, or may interfere with the successful completion or interpretation of the study results
7. Subject with a history of the following: angioedema, cardiac arrhythmia, angina pectoris, myocardial infarction, cerebrovascular accident, cardiac failure, thrombotic events, embolism, coagulopathy, diabetes mellitus, hyperlipidaemia, nephrotic syndrome, acute renal injury, chronic obstructive pulmonary disease, asthma, hepatic disease, reticuloendothelial system dysfunction, or nervous system disorder
8. Subject with known personal or family history of abnormal bleeding episodes
9. Subject not willing to receive study treatment via IM route of administration or unable to receive study treatment via IM route of administration
10. Subject with cardiovascular risk factors based on medical history: active tobacco smoking and/or ongoing diabetes mellitus at the screening visit
11. Subject with thrombosis risk factors: prolonged immobilization within 2 months prior to the screening visit, history of venous or arterial thrombosis, use of estrogens (30 days prior to the study drug administration), indwelling central vascular catheters and

hyperviscosity or hypercoagulable states
12. Subject with known history of hypersensitivity/allergic reaction to blood/plasma products
13. Subject with known selective Immunoglobulin A (IgA) deficiency (with or without antibodies to IgA)
14. Subject who received any plasma-derived product infusion within 12 months prior to study treatment administration
15. Subject who received a blood or plasma transfusion within 12 months prior to study treatment administration
16. Subject with systolic blood pressure >140 mmHg or diastolic blood pressure >90 mmHg at the screening visit and prior to treatment administration
17. Subject with anemia (hemoglobin <12 g/dL in women and 13 g/dL in men) at the screening visit
18. Subjects with proteinuria (>1+ on urine dipstick), blood urea nitrogen (BUN) or creatinine greater than the upper limit of normal at the screening visit
19. Subject with liver enzymes (aspartate aminotransferase [AST], alanine aminotransferase [ALT] and gamma-glutamyl transferase [GGT]) levels greater than the upper limit of normal at screening visit
20. Subject who received any dose of parenteral, oral, or inhaled corticosteroids, immunosuppressants, or immunomodulators within 6 weeks prior to the screening visit
21. Subject who received any live virus vaccine within five months prior to the screening visit
22. Subject not willing to postpone receiving any live virus vaccines until 6 months after receiving study treatment
23. Currently receiving any anti-viral treatment, regardless of the route of administration
24. Subject with virus safety laboratory results (serology and/or nucleic acid amplification technology [NAT]) indicative of a current infection with HAV, hepatitis B virus (HBV), hepatitis C virus (HCV), human immunodeficiency virus (HIV) or parvovirus B19 (B19V) at the screening visit
25. Participated in another clinical trial within 30 days prior to the screening visit or has received any investigational products within 3 months prior to the screening visit
26. Positive urine drug panel testing at the screening visit or prior to study treatment administration
27. Known or suspected abuse of alcohol, opiates, psychotropic agents or other drugs or chemical substances; or has done so in the 12 months prior to the screening visit
28. In the opinion of the Investigator, the subject may have compliance problems with the protocol and the procedures of the protocol
29. Subject who has already been included in a previous group for this clinical study

Investigational Product, Dose and Mode of Administration:

Investigational Product:

GamaSTAN [Immune Globulin (Human)]

Dose and Mode of Administration:

Single 0.2 mL/kg IM dose in the anterolateral aspects of the upper thigh or the deltoid muscle of the upper arm.

Doses over 5 mL are to be divided and injected into several muscle sites to reduce local pain and discomfort.

The gluteal region should not be used as an injection site due to the risk of injury to the sciatic nerve. The investigational product (IP) is not to be administered subcutaneously or intravenously.

Reference Therapy, Dose and Mode of Administration:

There is no reference therapy in this study.

Duration of Treatment:

After a screening period of up to 28 days, subjects will receive a single dose of the study treatment, which will be followed by a PK sampling period of 150 days. The total duration of participation for each subject in this study will be up to approximately 178 days.

Key Study Variables:

Primary Efficacy Variable:

- Proportion of subjects maintaining protective anti- HAV antibody levels (defined as anti-HAV antibody titer ≥10 mIU/mL in plasma and/or serum) up to 60 days after study treatment administration

PK Variables for anti-HAV antibodies in plasma and/or serum:

- $AUC_{0-\infty}$: Area under the plasma and/or serum concentration time curve extrapolated to infinity
- AUC_{0-T}: Cumulative area under the plasma and/or serum concentration time curve calculated from 0 to time of last observed quantifiable plasma and/or serum concentration
- C_{max}: Maximum observed plasma and/or serum concentration
- T_{max}: Time of maximum observed plasma and/or serum concentration
- λ_z: Apparent elimination rate constant
- T_{half}: Terminal elimination half-life
- Cl_{TOT}/F: Apparent total plasma and/or serum clearance
- V_D/F: Apparent volume of distribution

Safety Variables:

- Adverse events (AEs) including serious adverse events (SAEs), suspected adverse drug reactions (ADRs), and adverse reactions (ARs)
- Clinical laboratory parameters including chemistry, hematology, and urinalysis
- Physical examination
- Vital signs (heart rate (HR), blood pressure (BP), respiratory rate (RR), and body temperature)

Study Assessments and Procedures:

Screening Visit (Day -28 to Day -2):

Subjects will be screened for enrollment in this study. The following procedures will be

done:

- Obtain informed consent
- Review of inclusion and exclusion criteria for determination of subject's study eligibility
- Documentation of medical history
- Documentation of demographics
- Height and weight measurement and determination of BMI
- Full physical examination (excluding breast and genitourinary examination)
- ECG
- Vital signs measurement (HR, BP, RR, and body temperature)
- Clinical laboratory assessments (chemistry, hematology, urinalysis) (see Table 7-1 for details)
- Virus safety testing (see Table 7-1 for details)
- Coagulation tests (INR, aPTT)
- Immunoglobulin A (IgA) testing (see Table 7-1 for details)
- Urine alcohol and drug panel testing (see Table 7-1 for details)
- Urine pregnancy test for females
- Record prior (30 days prior to screening visit) and concomitant medications
- Record AEs

Treatment Period:

Day -1:

Subjects will be admitted to the clinical research unit on the day prior to study drug injection. The following procedures will be done upon admission:

- Review of subject eligibility (review of inclusion/exclusion criteria)
- Record concomitant medications
- Record AEs
- Vital signs measurement (HR, BP, RR, and body temperature)
- Symptom-directed physical examination
- ECG
- Clinical laboratory assessments (chemistry, hematology, urinalysis) (see Table 7-1 for details)
- Urine alcohol and drug panel testing (see Table 7-1 for details)
- Urine pregnancy test for females

Day 1:

The following procedures will be done (all pre-dose procedures are to be performed within 1 hour prior to dosing):

- Collection of virus safety retain samples prior to study drug injection
- Vital signs measurement (HR, BP, RR, and body temperature) prior to study drug injection, 60 minutes post study drug injection (a window of ±10 minutes is permitted) and 12 hours post study drug injection (a window of ±10 minutes is permitted)

- Record AEs and concomitant medications prior to and post study drug injection
- Study drug injection
- PK blood sampling prior to study drug injection, 60 minutes post study drug injection (a window of ±10 minutes is permitted) and 12 hours post study drug injection (a window of ±1 hour is permitted)
- Injection site evaluation 60 minutes (±10 minutes) post study drug injection (Note: any reactions will be recorded as AEs)

Day 2:

- Record concomitant medications
- Record AEs
- PK blood sample will be collected approximately 24 hours following study treatment administration (a window of ±1 hour is permitted)
- Injection site evaluation approximately 24 hours post study drug injection (prior to discharge) (Note: any reactions will be recorded as AEs)
- Symptom-directed physical examination approximately 24 hours post study drug injection (prior to discharge)
- Subjects will be discharged from the clinical research unit on Day 2, following clinical assessments and procedures, and will return for each of the remaining scheduled ambulatory visits (from Days 3 to 150).

Day 3 to Day 115:

A PK blood sample will be collected on each of the following days:

- Days 3, 4 and 5 (a window of ±4 hours is permitted)
- Days 7, 10, and 14 (a window of ±1 day is permitted)
- Days 21 and 28 (a window of ±2 days is permitted)
- Days 60, 79, and 115 (a window of ±4 days is permitted)

Procedures performed on Days 3, 4, 5, 7, 10, 14, 21, 28, 60, 79 and 115 (the window for these procedures is the same as that for the PK sample to be collected on the same study Day):

- Record concomitant medications
- Record AEs
- Vital signs measurement (HR, BP, RR, and body temperature) (Day 5, 28, and 60 visits only)
- Symptom-directed physical examination (Day 5, 28, and 60 visits only)
- Clinical laboratory assessments (chemistry, hematology, urinalysis) (Day 5, 28, and 60 visits only) (see Table 7-1 for details)

Final Visit (Day 150 ±7 days)/Early Discontinuation Visit:

- Record concomitant medications
- Record AEs

- A PK blood sample will be collected
- Full physical examination (excluding breast and genitourinary examination)
- Vital signs measurement (HR, BP, RR, and body temperature)
- Clinical laboratory assessments (chemistry, hematology, urinalysis) (see Table 7-1 for details)
- Collection of virus safety retain samples (see Table 7-1 for details)

Statistical Methods:

Efficacy analysis:

The percentage of subjects maintaining anti-HAV antibody levels ≥10 mIU/mL up to Day 60 following study treatment administration will be summarized. The anti-HAV antibody levels during the PK sampling period (up to Day 150 following study treatment administration) will be summarized. The efficacy analyses will be performed on the evaluable and safety populations.

PK Analysis:

PK analyses will be performed on the PK population. All PK parameters ($AUC_{0-\infty}$, AUC_{0-T}, C_{max}, T_{max}, λ_z, T_{half}, Cl_{TOT}/F and V_D/F) will be listed and summarized using arithmetic as well as geometric means and standard deviation (SD), percent coefficient of variation (CV), median, and minimum/maximum, as appropriate. PK parameters will be estimated using both anti-HAV antibody baseline uncorrected and baseline corrected levels.

Safety Analysis:

Safety analyses will be performed on the safety population. Safety data will be summarized with descriptive statistics and frequency tables and will include clinical laboratory values, vital signs and physical examination findings. The incidence of AEs, suspected ADRs, ARs, and AEs by severity and causality will be summarized. Deaths, subjects with SAEs and AEs leading to premature discontinuation from the study will be listed and presented in narrative form.

Determination of Sample Size:

Approximately 28 healthy subjects will be enrolled and treated in this study which will provide about 20 evaluable subjects based on approximately 30% dropout rate. The sample size is chosen based on clinical considerations but not on a formal sample size calculation.

TABLE OF CONTENTS

LIST OF IN-TEXT TABLES

LIST OF APPENDICES

GLOSSARY AND ABBREVIATIONS

ADR	Adverse drug reaction
AE	Adverse event
ALP	Alkaline phosphatase
ALT	Alanine aminotransferase
aPTT	Activated partial thromboplastin time
AR	Adverse reaction
AST	Aspartate aminotransferase
AUC	Area under the concentration time curve
AUC_{0-T}	Cumulative area under the plasma and/or serum concentration time curve calculated from 0 to time of last observed quantifiable plasma and/or serum concentration
$AUC_{0-\infty}$	Area under the plasma and/or serum concentration time curve extrapolated to infinity
B19V	Parvovirus B19
BMI	Body mass index
BUN	Blood urea nitrogen
CI	Confidence interval
CJD	Creutzfeldt-Jakob disease
Cl_{TOT}	Apparent total plasma and/or serum clearance
C_{max}	Maximum observed plasma and/or serum concentration
eCRF	Case report form/electronic case report form
CRO	Contract research organization
CV	Coefficient of variation
DBP	Diastolic blood pressure
DNA	Deoxyribonucleic acid
F	Bioavailability
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GGT	Gamma-glutamyl transferase
HAV	Hepatitis A virus
HBV	Hepatitis B virus

HCG	Human chorionic gonadotropin
HCV	Hepatitis C virus
HIV	Human immunodeficiency virus
HR	Heart rate
IB	Investigator's brochure
ICF	Informed consent form
ICH	The International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use
IG	Immune globulin
IgA	Immunoglobulin A
IgG	Immunoglobulin G
IgM	Immunoglobulin M
IM	Intramuscular
INR	International Normalized Ratio
IP	Investigational product
IRB/EC	Institutional Review Board/Ethics Committee
kg	Kilogram
LDH	Lactate dehydrogenase
MedDRA	Medical Dictionary for Regulatory Activities
NAT	Nucleic acid amplification technology
PK	Pharmacokinetic
PT	Prothrombin time
RR	Respiratory rate
RNA	Ribonucleic acid
SAE	Serious adverse event
SAP	Statistical analysis plan
SBP	Systolic blood pressure
SD	Standard deviation
S/D	Solvent/Detergent
TEAE	Treatment-emergent adverse event
T_{half}	Terminal elimination half-life
T_{max}	Time of maximum observed plasma and/or serum concentration

US	United States
V_D	Apparent volume of distribution
λ_z	Apparent elimination rate constant

1 GENERAL INFORMATION

Protocol title and other key study information are provided on the title page. Information regarding additional key personnel and organizations involved in the conduct of the study, including names and contact details of participating investigators, monitors, clinical laboratories, technical departments and/or institutions, as well as information on members of additional study committees, will be found in the study files of the sponsor and at the Investigator sites within the study reference manual/file.

Investigators and staff will receive training either via an Investigators meeting or other appropriate individual site training session(s).

2 BACKGROUND INFORMATION

In addition to the information provided below, please also refer to the Investigator's Brochure (IB) and any additional data supplied by the sponsor.

2.1 Name and Description of the Investigational Product(s)

See Section 4.4 Study Treatments for detail.

The following treatment will be used:

- GamaSTAN (Immune Globulin ([Human]): a 15 to 18% protein solution of human Immunoglobulin G (IgG) for intramuscular (IM) injection

2.2 Relevant Findings from Nonclinical and Clinical Trials

2.2.1 Pre-exposure Prophylaxis for Hepatitis A Infection

The World Health Organization estimates an annual total of 1.5 million clinical cases of hepatitis A infection worldwide, but seroprevalence data indicate that tens of millions of hepatitis A virus (HAV) infections occur each year [(1)]. Since the recommendation of HAV vaccination in 2006, vaccination rates and evidence of vaccine-induced immunity in young persons have increased in the past decade [(2, 3)]. Yet hepatitis A vaccination coverage levels remain lower than those for other routinely recommended vaccines [(4)]. Additionally, many older adults remain unvaccinated and susceptible to infection. International travel is the most commonly reported risk for HAV infection [(5)]. After adjusting for under-ascertainment and under-reporting [(6)], the estimated number of new HAV infections in the US in 2015 was 2,800 (95% confidence interval [CI] =1,900–3,100) [(7)].

Immune globulin (IG) provides protection against HAV infection through passive transfer of antibody. Efficacy is greatest when IG is administered early in the incubation period; when administered later, IG often only attenuates the clinical expression of hepatitis A infection [(8)].

It has been shown that prophylactic IM injection of IgG with anti-HAV activities provides protection against HAV infection. GamaSTAN S/D is one such licensed product that is commonly administered to provide pre-exposure prophylaxis for persons travelling to areas with endemic hepatitis A [9]. GamaSTAN S/D and GamaSTAN are both purified using a procedure based on the Cohn-Oncley process [10, 11], but GamaSTAN uses a caprylate/chromatography purification step.

2.2.2 Rationale for Clinical Investigation of GamaSTAN

GamaSTAN is manufactured from human plasma employing caprylate/chromatography steps in the purification scheme. This production process yields a final IgG product that is purer and more closely reflects the IgG subclass distribution found in plasma than earlier IgG products [12, 13]. GamaSTAN is being developed for the same indications as those of GamaSTAN S/D including pre-exposure prophylaxis for persons travelling to areas with endemic HAV.

2.3 Known and Potential Risks and Benefits to Human Subjects

Because GamaSTAN is made from human plasma and is therefore a biological product, it may carry a risk of transmitting infectious agents (e.g., viruses and, theoretically, the Creutzfeldt-Jakob disease (CJD) agent). No cases of transmission of viral diseases or CJD have ever been identified for other products manufactured with the same caprylate/chromatography purification process. The risk that GamaSTAN can transmit an infectious agent has been reduced by screening plasma donors for prior exposure, testing donated plasma, and by the inclusion of steps in the manufacturing process with the demonstrated capacity to inactivate and/or remove pathogenic agents. Despite these measures, it may carry a risk of transmitting infectious agents. [9]

There is clinical evidence of an association between the administration of all immunoglobulins and thromboembolic events such as myocardial infarction, stroke, pulmonary embolism and deep vein thrombosis. Therefore, caution should be exercised when prescribing and administering immunoglobulins. Thrombosis may occur even in the absence of known risk factors.

GamaSTAN should be given with caution to subjects with a history of prior systemic allergic reactions following the administration of human IG preparations [14]. GamaSTAN should not be given to subjects with isolated immunoglobulin A (IgA) deficiency. Such persons have the potential for developing antibodies to IgA and could have anaphylactic reactions to subsequent administration of blood products that contain IgA.Other antibodies in the GamaSTAN preparation may interfere with the response to live virus vaccines such as measles, mumps, polio, or rubella, and varicella. Therefore, use of such vaccines should be deferred until approximately 5 to 6 months after GamaSTAN administration. When administering GamaSTAN as prophylaxis for measles, wait 5 months before giving measles vaccine to a non-immunocompromised subject and wait 6 months for an immunocompromised subject [15].

Local pain and tenderness at the injection site, urticaria, and angioedema may occur. Anaphylactic reactions, although rare, have been reported following the injection of human IG preparations. Anaphylaxis is more likely to occur if GamaSTAN is given intravenously; therefore, GamaSTAN must be administered only intramuscularly.

As with all preparations administered by the IM route, bleeding complications may be encountered in subjects with thrombocytopenia or other bleeding disorders.

Grifols' licensed Immune Globulin (Human) product, GamaSTAN S/D, is commonly administered as prophylaxis to provide active immunization following exposure to HAV, to provide pre-exposure prophylaxis for persons travelling to areas with endemic hepatitis A. GamaSTAN S/D is also indicated to prevent or modify measles, passive immunization against varicella, and prophylaxis in pregnant females exposed to rubella. The immediate administration of an IG product is intended to provide immediate passive virus neutralizing antibody coverage [(2)].

Since the study treatment is not being given to subjects to treat any symptoms or illness, there will be no direct medical benefit from participation in this trial.

2.4 Description of and Justification for the Route of Administration, Dosage, Dosage Regimen, and Treatment Period(s)

2.4.1 Administration of Investigational Products

A single 0.2 mL/kg dose of the investigational product (IP) will be injected intramuscularly in the anterolateral aspects of the upper thigh or the deltoid muscle of the upper arm.

Doses over 5 mL are to be divided and injected into several muscle sites to reduce local pain and discomfort.

The gluteal region should not be used as an injection site due to the risk of injury to the sciatic nerve. The IP is not to be administered subcutaneously or intravenously.

2.4.2 Justification for Selection of Doses/Timing of Investigational Products

Recently the Food and Drug Administration (FDA) approved and instituted an anti-HAV titer specification (Not Less Than >20 IU/mL) and revised dosing for GamaSTAN S/D. Due to the recently instituted titer specifications and revised dosing, Grifols committed to the FDA to evaluate anti-HAV titers following IM administration at the recommended dose of 0.2 mL/kg in non-HAV (seronegative) healthy volunteers. An IM dose of 0.2 mL/kg was chosen, as it is the recommended dose for prophylaxis prior to exposure to hepatitis A for persons traveling to areas endemic with the virus.

Peak levels of IgG are obtained approximately 2 days after IM injection of immunoglobulins [(12)]. The half-life of IgG in the circulation of individuals with normal IgG levels is 23 days [(9)]. In this study, a 150-day follow-up period is chosen as it represents approximately 5 half-lives.

2.5 Compliance Statement

This study will be conducted under the conditions described in this protocol and in compliance with The International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH), Good Clinical Practice (GCP) and all applicable regulatory requirements.

2.6 Study Population

The study population will consist of HAV-seronegative healthy male and female subjects. Subjects must meet all the inclusion criteria and none of the exclusion criteria at the screening visit to be eligible for participation in this study. Continued eligibility will be assessed upon admission to the clinical site, prior to study treatment administration.

3 STUDY OBJECTIVES AND PURPOSE

3.1 Efficacy Objectives

The primary efficacy objective is to evaluate whether a single 0.2 mL/kg IM dose of the study treatment will provide protective levels of anti-HAV antibodies in HAV-seronegative healthy subjects for up to 60 days.

3.2 Pharmacokinetic Objectives

The pharmacokinetic (PK) objective is to evaluate the PK parameters of anti-HAV antibodies following a single 0.2 mL/kg IM dose of the study treatment in HAV-seronegative healthy subjects.

3.3 Safety Objectives

The safety objective is to evaluate the safety and tolerability of a single 0.2 mL/kg IM dose of the study treatment in HAV-seronegative healthy subjects.

4 STUDY DESIGN

4.1 Primary Endpoint and Secondary Endpoints

The primary efficacy endpoint is the proportion of subjects maintaining protective anti-HAV antibody levels (defined as anti-HAV antibody titer ≥10 mIU/mL in plasma and/or serum) up to 60 days after study treatment administration.

With respect to the PK endpoint, the parameters of interest for this study are: $AUC_{0-\infty}$, AUC_{0-T}, C_{max}, T_{max}, λ_z, T_{half}, Cl_{TOT}/F and V_D/F.

The safety endpoints will include a summary of the incidence of AEs, SAEs, suspected ADRs and ARs, as well as descriptive summary and statistics of the safety parameters (clinical laboratory values, vital signs and physical examination findings).

4.2 Study Design and Plan

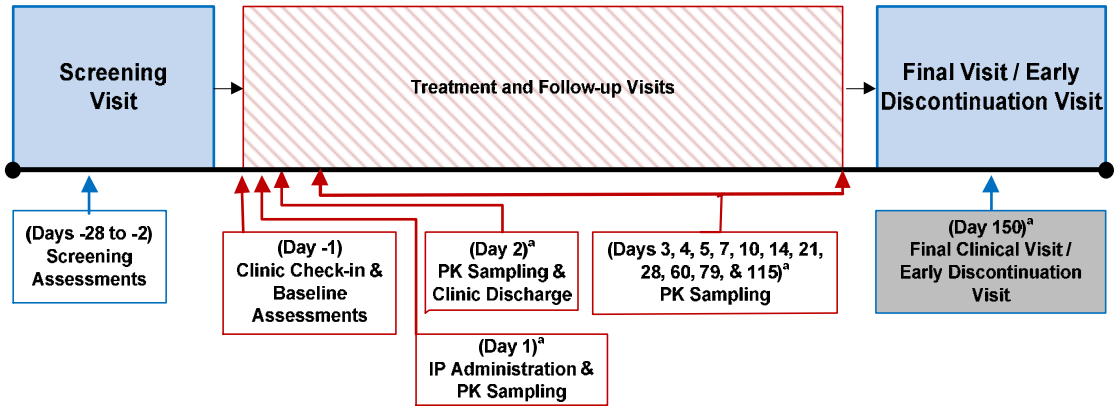
This is a single center, open-label, single-arm study design, in which approximately 28 subjects will receive the same study treatment (0.2 mL/kg dose via IM injection). There is no reference therapy in this study.

The study will be explained to each subject prior to the subject providing written informed consent. All subjects will be screened to ensure that all the inclusion criteria and none of the exclusion criteria are met.

A sufficient number of subjects will be qualified by screening assessments and procedures for reporting to the clinical site on Day -1. Subjects will be discharged from the clinic on Day 2, following the scheduled assessments and procedures, and will return to the clinical site for the remaining ambulatory PK samples and safety monitoring, and again for the final visit (Day 150).

The total duration of study participation for subjects who complete the study will be approximately 178 days.

The Schedule of Study Procedures and Events is provided in Appendix 1. The study consists of a Screening Visit, Treatment and Follow-up Visits, and Final Visit / Early Discontinuation Visit. The overall study schema is presented in Figure 4-1.



[a] *PK sampling visits will be conducted as close as possible to the exact time points. The PK sampling visits: Day 1: prior to study drug injection, 60 minutes (±10 minutes) post study drug injection and 12 hours post study drug injection (a window of ±1 hour is permitted); Day 2 (window of ±1 hour); Days 3, 4 and 5 (visits have a window of ±4 hours); Days 7, 10, and 14 (all 3 visits have a window of ±1 day); Days 21 and 28 (both visits have a window of ±2 days); Days 60, 79 and 115 (all 3 visits have a window of ±4 days); Day 150 (a window of ±7 days)*

Figure 4-1 Overall Study Diagram

4.3 Measures Taken to Minimize/Avoid Bias

4.3.1 Subject Numbering

Subjects in the study will receive a consecutive subject number. Subject numbers are generated beginning with the study center number (3 digits, assigned by the sponsor) followed consecutively with a unique number for each subject (4 digits, including leading zeros). For example, if the Investigator's center number is 301, subject number will be 3010001, 3010002, 3010003, etc., in consecutive order. Subject numbers, once assigned, will not be reused. If a subject is re-screened, a different subject number will be assigned to this subject.

4.3.2 Randomization

As this is an open-label, single-arm study design, no randomization code will be provided.

4.3.3 Blinding

Not applicable as this is an open-label study.

4.4 Study Treatments

4.4.1 Treatments to Be Administered

A single 0.2 mL/kg IM injection of GamaSTAN, administered in the anterolateral aspects of the upper thigh or the deltoid muscle of the upper arm.

Doses over 5 mL are to be divided and injected into several muscle sites to reduce local pain and discomfort.

The gluteal region should not be used as an injection site due to the risk of injury to the sciatic nerve. The IP is not to be administered subcutaneously or intravenously.

4.4.1.1 GamaSTAN

GamaSTAN (Immune Globulin ([Human]) is a 15 to 18% protein solution of human IgG for IM injection solution of human IgG for IM injection. GamaSTAN contains a minimum of 20 IU/mL of anti-HAV antibodies. GamaSTAN is a clear to opalescent, colorless to pale yellow sterile solution of polyvalent human IgG that contains no preservative.

GamaSTAN is provided in vials for single use. The batch number(s) of the study formulation will be indicated in the study files.

4.4.2 Labeling of Investigational Product

IP will be labeled according to the requirements of local law and legislation. Label text will be approved according to agreed Grifols procedures, and a copy of the labels will be made available to the study site.

4.4.3 Packaging of Investigational Product

The sponsor will be responsible for ensuring that the IP is manufactured in accordance with applicable Good Manufacturing Practice regulations and requirements.

4.4.4 Storage of Investigational Product

GamaSTAN must be stored in a secure location accessible only to study personnel authorized by the Investigator, such as the pharmacy staff responsible for the preparation and dispensing of IP.

GamaSTAN must be stored at 2 to 8°C (36 to 46°F). Do not freeze.

Investigators or designees are responsible for maintaining storage temperature records and for immediately reporting deviations in temperature to the study monitor. Continuous 24 hour, 7 days a week temperature monitoring is preferred. IP storage monitoring should be recorded and maintained by pharmacy staff. Copies of the storage temperature monitoring records will be made available for review by the Site Monitor, if applicable, during the course of the study and will be provided to the Sponsor at completion of the study. Details of receipt, storage, dispensing, and destruction will be recorded and maintained in the pharmacy log book by pharmacy staff.

Unused IP will either be destroyed according to local or institutional disposal regulations and regulatory requirement or shipped return to the Sponsor at the end of the study. The Sponsor will provide written instructions regarding final disposition of the unused IP. Copies of the IP storage and destruction records will be provided to the Sponsor at completion of the study.

4.5 Expected Duration of Subject Participation in the Study

After a screening period of up to 28 days, subjects will receive a single dose of the study treatment, which will be followed by a PK sampling period of 150 days. The total duration of participation for each subject in this study will be up to approximately 178 days.

4.6 Discontinuation Criteria for Individual Subjects and Study

4.6.1 Discontinuation Criteria for Individual Subjects

See Section 5.3 Subject Withdrawal Criteria.

4.6.2 Premature Termination of Study

The sponsor, Institutional Review Board/Ethics Committee (IRB/EC), and/or regulatory authorities have the right to close this study or the study center, and an Investigator /sponsor has the right to close the study center, at any time, although this should occur only after consultation between involved parties. The IRB/EC must be informed. Should the study/center be closed prematurely, all study materials (except documentation that has to remain stored at site) must be returned to the sponsor. An Investigator will retain all other documents until notification given by the sponsor for destruction.

The study center can be closed for the following reasons:

- Lack of enrollment
- Non-compliance with the requirements of the study protocol
- Non-compliance with ICH, GCP

4.7 Accountability Procedures for Investigational Product(s)

IP is to be used only for the study in accordance with the directions given in this protocol. An Investigator, or designee such as the study pharmacist, is responsible for the distribution of the IP in accordance with directions given in the protocol.

An Investigator is responsible for maintaining accurate records of the IP for his/her site. IP inventory/dispensing documentation verifying the receipt, dispensing, damaged state or return must be maintained and kept current by an Investigator, or designee. The inventory must be made available for inspection by the monitor. IP supplies must be accounted for by the monitor and inventory/dispensing logs must be verified by the monitor prior to IP return or destruction. Written documentation of any used and unused inventory is required. At the end of the study, a copy of the inventory/dispensing log(s) will be retrieved by the monitor and returned to Grifols.

4.8 Maintenance of Treatment Randomization Codes

As this is an open-label, single-arm study design, no randomization code will be provided.

4.9 Data to Be Recorded Directly on the eCRFs

Source documents will be used to record all study-related data. Designated staff will enter the data required by the protocol into the Electronic Case Report Forms (eCRF).

5 SELECTION AND WITHDRAWAL OF SUBJECTS

This study will include HAV-seronegative healthy adult subjects.

5.1 Inclusion Criteria

A subject must meet all the following inclusion criteria to be eligible for participation in this study:

1. Male subjects from 18 to 55 years of age, inclusive, or female subjects from 18 to 65 years of age, inclusive
2. Subjects with a body mass index (BMI) of 18.5 to 29.9 kg/m^2
3. Body weight greater than or equal to 50 kg at screening
4. Subjects willing and able to provide written informed consent

5. Subjects in good health in the judgment of the Investigator, as determined by medical history, physical examination, vital signs, ECG and laboratory assessments
6. A female study subject must meet one of the following criteria:
    a. If a female of childbearing potential – agrees to use one of the accepted contraceptive regimens from at least 30 days prior to study treatment administration and during the entire study duration. An acceptable method of contraception includes one of the following:
        - Abstinence from heterosexual intercourse (i.e. when abstinence is the preferred and usual lifestyle of the subject; periodic abstinence is not acceptable)
        - Non-estrogen containing hormonal contraceptives (birth control pills, injectable/implant/insertable hormonal birth control products, transdermal patch)
        - Intrauterine device without hormones
        - Condom with spermicide
        - Diaphragm or cervical cap with spermicide
        - Vasectomized partner (minimum 6 months since vasectomy prior to study treatment administration)
    b. If a female of non-childbearing potential – should be surgically sterile (i.e. has undergone complete hysterectomy, bilateral oophorectomy, or tubal ligation) or in a menopausal state (at least 1 year without menses prior to study treatment administration)
7. A male study subject must agree to use one of the accepted contraceptive regimens during the entire study duration;
        - Abstinence from heterosexual intercourse
        - Female partner with condom with spermicide used by male study subject
        - Female partner of non-childbearing potential
        - Male sterilization (if proof of sterilization is not provided, the subject must agree to use one of the above accepted contraceptive methods)
8. A male study subject must agree not to impregnate a female or donate sperm during the entire study duration

5.2 Exclusion Criteria

A subject meeting any of the following exclusion criteria is NOT eligible for participation in the study:

1. Subject vaccinated against HAV, as documented the in medical history at the screening visit
2. Subject with positive anti-HAV antibodies in blood sample at the screening visit
3. Subject who previously received any type of IG, including HAV IG within the past 12 months prior to study treatment administration

4. Subject with prolonged International Normalized Ratio (INR) or activated partial thromboplastin time (aPTT) at the screening visit
5. Subject with a platelet count below 100×10^9/L at the screening visit
6. Subject suffering from some acute or chronic medical, surgical or psychiatric significant condition or laboratory abnormality at the screening visit or prior to study treatment administration that, according to Investigator judgement, may increase the risk associated with study participation or study treatment administration, or may interfere with the successful completion or interpretation of the study results
7. Subject with a history of the following: angioedema, cardiac arrhythmia, angina pectoris, myocardial infarction, cerebrovascular accident, cardiac failure, thrombotic events, embolism, coagulopathy, diabetes mellitus, hyperlipidaemia, nephrotic syndrome, acute renal injury, chronic obstructive pulmonary disease, asthma, hepatic disease, reticuloendothelial system dysfunction, or nervous system disorder
8. Subject with known personal or family history of abnormal bleeding episodes
9. Subject not willing to receive study treatment via IM route of administration or unable to receive study treatment via IM route of administration
10. Subject with cardiovascular risk factors based on medical history: active tobacco smoking and/or ongoing diabetes mellitus at the screening visit
11. Subject with thrombosis risk factors: prolonged immobilization within 2 months prior to the screening visit, history of venous or arterial thrombosis, use of estrogens (30 days prior to the study drug administration), indwelling central vascular catheters and hyperviscosity or hypercoagulable states
12. Subject with known history of hypersensitivity/allergic reaction to blood/plasma products
13. Subject with known selective IgA deficiency (with or without antibodies to IgA)
14. Subject who received any plasma-derived product infusion within 12 months prior to study treatment administration
15. Subject who received a blood or plasma transfusion within 12 months prior to study treatment administration
16. Subject with systolic blood pressure >140 mmHg or diastolic blood pressure >90 mmHg at the screening visit and prior to treatment administration
17. Subject with anemia (hemoglobin <12 g/dL in women and 13 g/dL in men) at the screening visit
18. Subjects with proteinuria (>1+ on urine dipstick), blood urea nitrogen (BUN) or creatinine greater than the upper limit of normal at the screening visit
19. Subject with liver enzymes (aspartate aminotransferase [AST], alanine aminotransferase [ALT] and gamma-glutamyl transferase [GGT]) levels greater than the upper limit of normal at screening visit
20. Subject who received any dose of parenteral, oral, or inhaled corticosteroids, immunosuppressants, or immunomodulators within 6 weeks prior to the screening visit

21. Subject who received any live virus vaccine within five months prior to the screening visit
22. Subject not willing to postpone receiving any live virus vaccines until 6 months after receiving IP
23. Currently receiving any anti-viral treatment, regardless of the route of administration
24. Subject with virus safety laboratory results (serology and/or nucleic acid amplification technology [NAT]) indicative of a current infection with hepatitis A virus (HAV), hepatitis B virus (HBV), hepatitis C virus (HCV), human immunodeficiency virus (HIV) or parvovirus B19 (B19V) at the screening visit
25. Participated in another clinical trial within 30 days prior to the screening visit or has received any IPs within 3 months prior to the screening visit
26. Positive urine drug panel testing at the screening visit or prior to study treatment administration
27. Known or suspected abuse of alcohol, opiates, psychotropic agents or other drugs or chemical substances; or has done so in the 12 months prior to the screening visit
28. In the opinion of the Investigator, the subject may have compliance problems with the protocol and the procedures of the protocol
29. Subject who has already been included in a previous group for this clinical study

5.3 Subject Withdrawal Criteria

5.3.1 Screen Failures

Screening evaluations will be used to determine the eligibility of each subject for enrollment. Subjects who fail to meet eligibility criteria during screening evaluations will be considered screen failures and will not participate in the study. However, subjects who fail the first screening evaluation are allowed to be re-screened once.

5.3.2 Removal of Subjects

Subjects may voluntarily withdraw from the study, or be removed from the study at the discretion of an Investigator or Sponsor at any time. An Investigator may withdraw a subject at any time if it is determined that continuing the study would result in a significant safety risk to the subject.

If such withdrawal occurs, or if the subject fails to return for visits, an Investigator should determine the primary reason for a subject's premature withdrawal from the study and record the reason in the respective subject's study documents.

Subjects may withdraw or be withdrawn from the study for the following reasons:

1. At their own request or at the request of their legally acceptable representative

2. If, in an Investigator's opinion, continuation in the study would be detrimental to the subject's well-being
3. At the specific request of the sponsor

Also subjects may be withdrawn for the following reasons:

- Noncompliance with the protocol requirements
- Difficulties with blood collection
- Pregnancy
- AE
- Unanticipated event that could result in an inadequately characterized PK profile (such as missed PK blood samples, for example)
- Other

In all cases, the reason for withdrawal must be recorded in the eCRF and in the subject's records.

5.3.3 Subject Replacement

Subjects who are withdrawn from the study will not be replaced.

5.3.4 Follow-up of Subjects Withdrawn from Study

Subjects who receive any amount of IP and discontinue early from the study will be requested to return for the early discontinuation visit procedures within approximately 7 days following discontinuation.

6 TREATMENT OF SUBJECTS

See Section 4.4 for the treatment to be administered, including the name of the IP, the dose, the dosing schedule and the route/mode of administration.

6.1 Administration and Timing of Investigational Products for Each Subject

Administration of the IP will be staggered between subjects for the purpose of accurate sampling time.

A single 0.2 mL/kg dose of the IP will be injected intramuscularly in the anterolateral aspects of the upper thigh or the deltoid muscle of the upper arm.

Doses over 5 mL are to be divided and injected into several muscle sites to reduce local pain and discomfort.

The gluteal region should not be used as an injection site due to the risk of injury to the sciatic nerve. The IP is not to be administered subcutaneously or intravenously.

The subjects will be discharged from the clinic on Day 2, following the scheduled assessments and clinical procedures. Subjects will be asked to return to the clinic for the remaining PK blood samples and clinical assessments, and for the final visit.

6.2 Prior and Concomitant Therapy

Concomitant medications must be recorded in the eCRF, including the trade and generic names of the medication, the dose, the route of administration, and the duration of the medication (frequency).

6.2.1 Prohibited Medications Prior to Study Participation

As per the exclusion criteria (see section 5.2), subjects are prohibited from taking any of the following prior to study participation:

1. Any type of IG, including HAV IG
2. Estrogens (for at least 30 days prior to the study drug administration and during the entire study duration)
3. Any plasma-derived product infusion
4. Any blood or plasma transfusion
5. Any live virus vaccine
6. Any anti-viral treatment, regardless of the route of administration
7. Any dose of parenteral, oral, or inhaled corticosteroids, immunosuppressants, or immunomodulators

6.2.2 Prohibited Concomitant Medications during the Study

In addition to the medications listed in section 6.2.1, subjects are prohibited from taking any IPs that are not for this study.

6.2.3 Restricted Concomitant Medications during the Study

No concomitant medications will be restricted during the study with the exception of any medication described in section 6.2.1.

6.3 Treatment Compliance

Reasons for any deviation from the administration of less than 100% of the IP dose, as prepared by the pharmacist or designee, must be recorded in the eCRF and in the subject's medical records.

To ensure treatment compliance, study treatment administrations will be observed and verified by a site staff member.

7 ASSESSMENT OF EFFICACY AND PHARMACOKINETICS

7.1 Efficacy Variables

7.1.1 Primary Efficacy Variable

The primary efficacy variable in this study is the proportion of subjects maintaining protective anti- HAV antibody levels (defined as anti-HAV antibody titer ≥10 mIU/mL in plasma and/or serum) up to 60 days after study treatment administration.

7.2 Pharmacokinetic Variables

The PK Variables for anti-HAV antibody in plasma and/or serum are:

- $AUC_{0-\infty}$: AUC from time zero to time infinity
- AUC_{0-T}: AUC from time zero to time of last measured concentration
- C_{max}: maximum concentration
- T_{max}: time to C_{max}
- λ_z: terminal elimination rate
- T_{half}: estimated elimination half-life
- Cl_{TOT}/F: apparent total plasma and/or serum clearance
- V_D/F: apparent volume of distribution

PK parameters will be estimated using both anti-HAV antibody baseline uncorrected and baseline corrected levels.

7.3 Methods and Timing for Assessing, Recording, and Analyzing Efficacy Parameters

7.3.1 Observations and Measurements

The following is a description of the procedures/assessments to take place at each study visit. See the Schedule of Study Procedures and Events in Appendix 1.

Screening Visit (Day -28 to Day -2):

Subjects will be screened for enrollment in this study. The following procedures will be done:

- Obtain informed consent
- Review of inclusion and exclusion criteria for determination of subject's study eligibility
- Documentation of medical history
- Documentation of demographics
- Height and weight measurement and determination of BMI
- Full physical examination (excluding breast and genitourinary examination)
- ECG

- Vital signs measurement (heart rate [HR], blood pressure [BP], respiratory rate [RR], and body temperature)
- Clinical laboratory assessments (chemistry, hematology, urinalysis) (see Table 7-1 for details)
- Virus safety testing (see Table 7-1 for details). Note: the viral safety test results for HAV serology (HAV antibody differential (Immunoglobulin M [IgM]/IgG)) will be used to determine subject eligibility
- Coagulation tests (INR, aPTT)
- Immunoglobulin A (IgA) testing (see Table 7-1 for details)
- Urine alcohol and drug panel testing (see Table 7-1 for details)
- Urine pregnancy test for females
- Record prior (30 days prior to screening visit) and concomitant medications
- Record AEs

Treatment Period:

Day -1:

Subjects will be admitted to the clinical research unit on the day prior to study drug injection. The following procedures will be done upon admission:

- Review of subject eligibility (review of inclusion/exclusion criteria)
- Record concomitant medications
- Record AEs
- Vital signs measurement (HR, BP, RR, and body temperature)
- Symptom-directed physical examination
- ECG
- Clinical laboratory assessments (chemistry, hematology, urinalysis) (see Table 7-1 for details)
- Urine alcohol and drug panel testing (see Table 7-1 for details)
- Urine pregnancy test for females

Day 1:

The following procedures will be done (all pre-dose procedures are to be performed within 1 hour prior to dosing):

- Collection of virus safety retain samples prior to study drug injection
- Vital signs measurement (HR, BP, RR, and body temperature) prior to study drug injection, 60 minutes post study drug injection (a window of ±10 minutes is permitted) and 12 hours post study drug injection (a window of ±10 minutes is permitted)
- Record AEs and concomitant medications prior to and post study drug injection
- Study drug injection

- PK blood sampling prior to study drug injection, 60 minutes post study drug injection (a window of ±10 minutes is permitted) and 12 hours post study drug injection (a window of ±1 hour is permitted)
- Injection site evaluation 60 minutes (±10 minutes) post study drug injection (Note: any reactions will be recorded as AEs)

Day 2:

- Record concomitant medications
- Record AEs
- PK blood sample will be collected approximately 24 hours following study treatment administration (a window of ±1 hour is permitted)
- Injection site evaluation approximately 24 hours post study drug injection (prior to discharge) (Note: any reactions will be recorded as AEs)
- Symptom-directed physical examination approximately 24 hours post study drug injection (prior to discharge)
- Subjects will be discharged from the clinical research unit on Day 2, following clinical assessments and procedures, and will return for each of the remaining scheduled ambulatory visits (from Days 3 to 150).

Day 3 to Day 115:

A PK blood sample will be collected on each of the following days:

- Days 3, 4 and 5 (a window of ±4 hours is permitted)
- Days 7, 10, and 14 (a window of ±1 day is permitted)
- Days 21 and 28 (a window of ±2 days is permitted)
- Days 60, 79, and 115 (a window of ±4 days is permitted)

Procedures performed on Days 3, 4, 5, 7, 10, 14, 21, 28, 60, 79 and 115 (the window for these procedures is the same as that for the PK sample to be collected on the same study Day):

- Record concomitant medications
- Record AEs
- Vital signs measurement (HR, BP, RR, and body temperature) (Day 5, 28, and 60 visits only)
- Symptom-directed physical examination (Day 5, 28, and 60 visits only)
- Clinical laboratory assessments (chemistry, hematology, urinalysis) (Day 5, 28, and 60 visits only) (see Table 7-1 for details)

Final Visit (Day 150 ±7 days)/Early Discontinuation Visit:

- Record concomitant medications
- Record AEs

- A PK blood sample will be collected
- Full physical examination (excluding breast and genitourinary examination)
- Vital signs measurement (HR, BP, RR, and body temperature)
- Clinical laboratory assessments (chemistry, hematology, urinalysis) (see Table 7-1 for details)
- Collection of virus safety retain samples (see Table 7-1 for details)

7.3.2 Description of Laboratory Tests and Procedures

Detailed descriptions of laboratory test procedures are located in the study Laboratory Manual. Table 7-1 provides a summary of the laboratory tests to be conducted for this study.

Table 7-1 Name, Description, and Location of Laboratory Tests and Procedures

Test Panel	Description	Location
Hematology	Hemoglobin, hematocrit, platelets, red blood cell count, white blood cell count (absolute and % with differential)	Central
Chemistry	Sodium, potassium, creatinine, calcium, BUN, LDH, AST, ALT, GGT, ALP, glucose, total bilirubin, direct and indirect bilirubin	Central
Coagulation [1]	INR and aPTT	Central
Immunoglobulin A (IgA)[1]	Quantitative	Central
Urine pregnancy test [2]	Qualitative urine β-HCG for female subjects	Local
Urine test for alcohol and drugs of abuse	Alcohol, marijuana, opiates, cocaine, amphetamines, methamphetamines, and benzodiazepines	Central
Urinalysis	Microscopic evaluation is done only with cause. pH, protein, glucose and blood	Central
Virus safety (NAT) testing	Screening: HAV ribonucleic acid (RNA), HBV DNA, HCV RNA, HIV-1 RNA, and B19V DNA testing	Central
Virus safety (serology) testing	Screening: hepatitis A antibody differential (IgM/IgG), hepatitis B core antibody differential (IgM/IgG), hepatitis C antibody, HIV-1/-2 + Group O antibody, and B19V antibody differential (IgM/IgG) testing	Central
Virus safety (NAT and serology) retain samples [3]	Day 1 prior to study drug injection and final visit	

1. Screening visit only.
2. Screening and on Day -1, prior to study treatment administration.
3. Virus safety (NAT and serology) retain samples collected during the study will only be tested if subject exhibits clinical signs and symptoms consistent with HAV, HBV, HCV, HIV or B19V infection while participating in the study. Virus safety retain samples will be retained until all analyses in support of the study are complete.

If samples collected for laboratory analyses are non-analyzable due to various factors (i.e., lost, quantity not sufficient/useful, laboratory error), they will need to be recollected by contacting the subject and arranging for re-sampling.

Blood and/or urine samples may be repeated at the discretion of the Principal Investigator and/or Sponsor.

7.3.3 Drug Concentration Measurements

Total anti-HAV antibody levels will be measured in the subjects throughout the study.

Table 7-2 PK Blood Sampling Schedule

Sample Number	Study Day	Time (Window)
01	1	Prior to treatment administration
02	1	60 minutes following treatment administration (±10 minutes)
03	1	12 hours following treatment administration (±1 hour)
04	2	24 hours following treatment administration (±1 hour)
05	3	48 hours following treatment administration (±4 hours)
06	4	72 hours following treatment administration (±4 hours)
07	5	96 hours following treatment administration (±4 hours)
08	7	(±1 day)
09	10	(±1 day)
10	14	(±1 day)
11	21	(±2 days)
12	28	(±2 days)
13	60	(±4 days)
14	79	(±4 days)
15	115	(±4 days)
16	150	(±7 days)

8 ASSESSMENT OF SAFETY

8.1 Safety Parameters

Safety of the IP will be evaluated in this study. Safety endpoints will include:

- AEs including SAEs, suspected ADRs, and ARs
- Clinical laboratory parameters including chemistry, hematology, and urinalysis
- Physical examination
- Vital signs (HR, BP, RR, and body temperature)

8.2 Methods and Timing for Assessing, Recording, and Analyzing Safety Parameters

8.2.1 Adverse Events

Adverse events occurring at any time between signing of the subject's informed consent form (ICF) and the last day of the subject's participation in the clinical trial will be reported and recorded on the appropriate subject's eCRF entry.

It is the Investigator's responsibility to ensure that all AEs are appropriately recorded.

Adverse events will be elicited by spontaneous reporting by the study individual or by a non-leading inquiry or direct observation by the study staff.

8.2.2 Clinical Laboratory Evaluations

All clinical laboratory data will be listed for each subject (refer to Table 7-1).

An Investigator will be required to classify laboratory results out of the normal range reported by the laboratory as clinically significant or not according to his/her criteria. Clinical laboratory evaluations may be repeated at an Investigator's discretion.

Laboratory results out of the normal range judged by an Investigator as clinically significant will be considered AEs.

8.2.3 Virus Safety Testing

Virus safety (NAT and serology) retain samples collected on Day 1 prior to IP administration and at the final visit or early discontinuation visit will be tested only if the subject exhibits clinical signs and symptoms consistent with hepatitis A, hepatitis B, hepatitis C, HIV, or B19V infection while participating in the study. Virus safety retain samples will be retained until all analyses in support of the study are complete. Additional samples for viral NAT and viral serology testing may be collected and tested during the study only if the subject exhibits clinical signs and symptoms consistent with hepatitis A, hepatitis B, hepatitis C, HIV, or B19V infection while participating in the study.

8.2.4 Vital Signs

Vital signs will be measured at the following visits: screening visit, study Days -1, 1, 5, 28 and 60, and at final visit on Day 150 (or early discontinuation visit).

The following vital signs will be assessed:

- Temperature
- Blood Pressure (systolic blood pressure [SBP]) and diastolic blood pressure [DBP]),
- HR
- RR

Vital signs will be routinely monitored by the study staff as detailed in Appendix 1. An Investigator will be required to classify vital signs abnormalities as clinically significant or not according to his/her criteria. Results will be recorded in source documents and on the subject eCRF. Vital signs abnormalities judged by an Investigator as clinically significant will be considered AEs.

8.2.5 Physical Examinations

A medically certified individual will conduct a physical examination, as necessary, at the following visits:

Screening visit, study Days -1, 2, 5, 28 and 60, and at final visit on Day 150 (or early discontinuation visit).

Physical examinations will be routinely monitored by the study staff as detailed in Appendix 1. A complete physical examination will be performed by a medically qualified and licensed individual. The physical examination will include a review of the following: head and neck, heart, lungs, abdomen and general appearance. Any abnormalities judged by an Investigator as clinically significant will be considered AEs.

8.3 Procedures for Eliciting Reports of and for Recording and Reporting Adverse Event and Intercurrent Illnesses

8.3.1 Warnings/Precautions

For complete information on GamaSTAN, refer to the GamaSTAN IB and to protocol section 2.3.

8.3.2 Adverse Event Monitoring

Subjects must be carefully monitored for AEs. This monitoring includes clinical and laboratory tests and physical signs. AEs should be assessed in terms of their seriousness, severity, and causal relationship to the IP.

AEs will be elicited by spontaneous reporting by the study individual or by a non-leading inquiry or direct observation by the study staff.

8.3.3 Adverse Event Definitions

8.3.3.1 Adverse Events

An AE is defined as any untoward medical occurrence in a subject or clinical investigation subject administered a medicinal product or study treatment and which does not necessarily have a causal relationship with this administration. An AE can therefore be any unfavorable and unintended sign (including any abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product. In this study, all local site reactions are to be considered as AEs.

8.3.3.2 Suspected Adverse Drug Reactions/Adverse Reactions

All noxious and unintended responses to a medicinal product or study treatment related to any dose should be considered suspected ADRs. The phrase "responses to a medicinal product" means that a causal relationship between a medicinal product or study treatment and an AE is at least a reasonable possibility.

In the framework of this study, a suspected ADR for which there is a reason to conclude that the drug caused the event will be labeled as an adverse reaction (AR); thus, ARs are a subset of suspected ADRs.

An AE will be considered a Suspected ADR when either the sponsor or an Investigator considers that the drug might have caused the adverse event.

8.3.4 Assessment of Causality of Adverse Event

An Investigator is required to provide a causality assessment for each AE reported to the Sponsor. The Sponsor will consider an Investigator's causality assessment and also provide its own assessment.

Causal relationship to the study drug will be established according to medical judgment on whether there is a **reasonable possibility of a causal relationship between the AE and the IP administration**:

An Investigator must determine and classify the AE causality according to the following categories:

Unrelated/Not related: there is not a reasonable possibility of causal relationship between the AE and the study drug.

Possibly related: there is evidence to suggest a causal relationship between the study drug and the AE.

Definitively related: there is a reason to conclude that the study drug caused the AE.

Criteria to assess the causal relationship should take into account of the following conditions: 1) a plausible temporal sequence from the study drug administration to the AE onset; 2) whether the event follows a known response pattern to the suspected treatment; 3) whether the AE could be reasonably explained by the subject's clinical state, comorbidities, or concomitant medications, as well as 4) the occurrence of improvement on stopping/reducing the treatment (positive dechallenge) and/or reappearance of the event on repeated exposure (positive rechallenge).

For expedited safety reporting purposes, AEs assessed as either "definitively related" or "possibly related" will be considered POTENTIALLY RELATED or just RELATED.

For any subject, all AEs that occur at any time from the beginning of IP administration until the final visit of the clinical trial will be considered treatment-emergent AEs (TEAEs).

8.3.5 Severity of Adverse Event or Suspected Adverse Drug Reaction

AEs and suspected ADRs will be classified depending on their severity according to the following definitions:

Mild: an AE which is well tolerated by the subject, causing minimum degree of malaise and without affecting normal activities.

Moderate: an AE that interferes with the subject’s normal activities.

Severe: an AE that prevents the subject from performing their normal activities.

AE and suspected ADR severity gradation must be distinguished from AE and suspected ADR seriousness gradation, which is defined according to event consequence. For example, headache can be mild, moderate or severe but not necessarily serious in all these cases.

An Investigator will be responsible for assessing the AE and suspected ADR intensity during the clinical trial, taking into account current criteria included in this section.

8.3.6 Expectedness of Adverse Event or Suspected Adverse Drug Reaction

An AE or suspected ADR is considered "unexpected" if the nature, seriousness, severity or outcome of the reaction(s) is not consistent with the reference information. The expectedness shall be determined by the Sponsor according to the reference document (i.e., IB) for any serious suspected ADRs (potentially related SAEs) for expedited safety reporting purposes.

8.3.7 Seriousness of Adverse Event or Suspected Adverse Drug Reaction

An AE or suspected ADR is considered "serious" if, in the view of either an Investigator or Sponsor, it results in any of the following outcomes:

- Death

- Life-threatening AE (life-threatening in the definition of "serious" refers to an event in which the subject was at risk of death at the time of the event; it does not refer to an event which hypothetically might have caused death if it were more severe)
- In-patient hospitalization or prolongation of existing hospitalization
- A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly/birth defect
- An important medical event (important medical event in the definition of "serious" refers to those events which may not be immediately life-threatening, or result in death, or hospitalization, but from medical and scientific judgment may jeopardize the subject or/and may require medical or surgical intervention to prevent one of the other outcomes listed above)

*Hospitalization is to be considered only hospital stay for equal or more than 24 hours. The following hospitalizations should not be reported as SAEs:

- hospitalization or prolongation of hospitalization needed for procedures required by the clinical trial protocol
- hospitalization or prolongation of hospitalization as part of a routine procedure followed by the center
- hospitalization for a survey visit, annual physicals, or social reasons
- elective or pre-planned hospitalizations for a pre-existing condition that had not worsened from Baseline (e.g. elective or scheduled surgery arranged prior to start of the study)
- admissions not associated with an AE (e.g. social hospitalization for purposes of respite care)

This definition permits either the Sponsor or an Investigator to decide whether an event is "serious". If either the Sponsor or an Investigator believes that the event is serious, the event must be considered "serious" and evaluated by the Sponsor for expedited reporting.

8.3.8 Adverse Event Documentation

All AEs occurring after the subject has **signed the ICF through the final visit (i.e., end of study)** must be fully recorded in the subject's eCRF as well as in the medical record. If no AE has occurred during the study period, this should also be indicated in the eCRF. Each SAE must be fully recorded on the SAE Report Form as well as in the medical records and indicated as serious in the eCRF.

It is the responsibility of an Investigator to ensure that AEs including SAEs are appropriately recorded.

At each visit, AEs will be elicited by asking the individual a non-leading question such as "Do you feel different in any way since the last visit?" Moreover, AEs will also be collected through directly observed events or spontaneously volunteered by the subject. Clearly related

signs, symptoms and abnormal diagnostic procedures should preferably be grouped together and recorded as a single diagnosis or syndrome wherever possible.

The following variables must be recorded in the AE eCRF:

- The verbatim term (a diagnosis is preferred)
- Date/time of onset
- Date/time of resolution
- Severity (mild, moderate, severe)
- Causality (unrelated, possibly related, definitely related)*
- Seriousness (yes, no)
- Action taken (with regard to IP)
- Other action (to treat the event)
- Outcome and sequel (follow-up on AE)

*Causality assessment will be made only when the AE occurs after the subject has initiated at least one infusion of the IP. An AE occurring before subject's exposure to IP will be always labeled as "unrelated".

In addition to an Investigator's own description of the AEs, each AE will be encoded according to the Medical Dictionary for Regulatory Activities (MedDRA®).

For example, a laboratory test abnormality considered clinically significant, e.g., causing the subject to withdraw from the study, requiring treatment or causing apparent clinical manifestations, or judged clinically significant in the context of the subject's medical history by an Investigator, should be reported as an AE. Each event must be described in detail along with start and stop dates, severity, relationship to IP, action taken and outcome. Each event must be adequately supported by documentation as it appears in the subject's medical or case file.

8.3.9 Reporting of Serious Adverse Events

8.3.9.1 Reporting of Serious Adverse Events

Any SAE (see section 8.3.7) that occurs after **signing the study ICF through the final visit (i.e., end of study)** must be expeditiously reported whether or not considered attributable to the study drug.

SAEs will be reported using the designated SAE Report Form. When an Investigator becomes aware of an SAE, she/he must submit a completed, signed and dated SAE Report Form (in English) within 24 hours to the Sponsor by email/fax. The date of this SAE discovery by the site staff should be documented in the source documents (i.e., medical records).

Each SAE must be followed up until resolution or stabilization. After the initial report, all relevant information for SAE follow-up, and for the outcome, must also be supplied to the Sponsor in a timely manner (within 3 days from its identification or within 24 hours for relevant new information) by means of the SAE Report Form. In addition, the Sponsor or contract research organization (CRO) may request additional information and/or reports.

All SAE Report Forms must be reported by e-mail to:

Grifols Global Pharmacovigilance

[redacted]

When required, and according to local law and regulations, SAEs must be reported to the IRB/EC and regulatory authorities.

8.3.9.2 Reporting Pregnancy

While pregnancy itself is not a true "AE," pregnancy occurring in a clinical study must be followed, to collect information regarding the experiences of gestation and pregnancy with IP exposure. An Investigator must report any pregnancy that occurs in a female study subject / partner of a male study subject subsequent to first treatment exposure to the Final Study Visit (Day 150)/Early Discontinuation Visit. Any female subject who becomes pregnant during the study will be discontinued from IP treatment and will be followed for pregnancy outcome.

A pregnancy not verified prior to study treatment administration but occurring during the course of the study will be not considered an AE, unless a relation to the study drug is suspected. In any case, a *Pregnancy Report Form* must be completed and sent as soon as possible to the Sponsor. A copy of the form should be filed at the study site for follow-up until the end of the pregnancy. Any pregnancy must be followed by an Investigator until delivery or to the end of pregnancy. Any anomalies, complications, abnormal outcomes, or birth defect observed in the child must be reported as an SAE within 24 hours of Investigator or study personnel's first knowledge.

Please use the email address or fax numbers (back up only) in section 8.3.9.1 for reporting pregnancy.

8.4 Type and Duration of the Follow-Up of Subjects after Adverse Events

In so far as is possible, all individuals will be followed up until the AE or suspected ADR has been resolved. If an AE/suspected ADR/SAE is present when the subject has completed the study, the course of the event must be followed until the final outcome is known, or the event has been stabilized and no further change is expected and an Investigator decides that no further follow-up is necessary.

9 STATISTICS

9.1 Statistical Methods

Unless otherwise specified, descriptive statistics will include the number of non-missing observations (n), mean, standard deviation (SD), median, minimum and maximum values for the continuous/quantitative data or absolute and relative frequency counts and percentages for categorical/qualitative data.

Data handling and evaluation procedures will be described in the Statistical Analysis Plan (SAP).

9.1.1 Demographic and Baseline Characteristics

Demographic and baseline characteristics will be listed and summarized. For quantitative variables, mean, SD, median, and minimum/maximum will be provided. For qualitative variables, the frequency and percentage will be provided.

9.1.2 Efficacy Analysis

The percentage of subjects maintaining anti-HAV antibody levels ≥10 mIU/mL up to Day 60 following study treatment administration will be listed and summarized. The anti-HAV antibody levels during the PK sampling period (up to Day 150 following study treatment administration) will be listed and summarized. N, mean (SD), 90% CI for mean, % coefficient of variation (CV), median, minimum, maximum, geometric mean will be provided. The individual plasma and/or serum concentration/time profiles will be presented using the actual sampling times whereas the mean plasma and/or serum concentration/time profiles will be presented using the nominal sampling times.

The efficacy analyses will be performed on the evaluable and safety populations (if different).

9.1.3 Safety Analysis

Safety analyses will be performed on the safety population. Adverse events, clinical laboratory values, vital signs and physical examination findings will be analyzed. Data will be described using descriptive analyses.

9.1.3.1 Adverse Events

Safety analysis will be primarily focused on a descriptive analysis of suspected ADRs. Safety assessment will be based on the prevalence of suspected ADRs that occurred during the clinical trial.

Suspected ADRs are the AEs with causality assessment of possibly or definitely related. Adverse reactions are the AEs with causality assessment of definitely related.

Adverse events will be coded and classified using MedDRA® terms (system organ class and preferred terms).

Adverse events will be classified as TEAEs or non-TEAEs depending on the comparison of AE onset date/time with the start date/time of study treatment with the IP. A TEAE will be defined as an AE which occurs between the beginning of the study drug injection and the final visit of the clinical trial. A non-TEAE will be defined as an AE which occurs prior to the start of study treatment. Non-TEAEs and TEAEs will be summarized separately.

All AEs will be summarized by presenting subject incidences and percentages, and they will also be listed by body systems with subject number.

In addition, TEAEs, including suspected ADRs, will be summarized by system organ class, preferred term, causal-relationship, intensity (severity) and seriousness (serious vs non-serious) using descriptive statistics. For the summary by severity or causality at each level of summarization (number of subjects, System Organ Class, or Preferred Term), a subject will only be counted once per system organ class or preferred term using the most severe AE or the AE with the strongest causal relationship to the IP.

Subjects with an SAE or who withdraw from the study because of an AE will also be individually listed and summarized.

AEs for which an Investigator causality assessment is missing or is not determined will be individually listed.

9.1.3.2 Clinical Laboratory Values

All clinical laboratory data collected in this study will be listed for each subject.

An Investigator will be required to classify out of the normal range laboratory results reported by the laboratory as clinically significant or not according to his/her criteria.

Out of the normal range laboratory results judged by an Investigator as clinically significant in the context of the subject's medical history will be considered AEs.

9.1.3.3 Vital Signs

Vital signs [HR, BP (systolic and diastolic), RR, and body temperature] will be listed for each clinical trial subject.

Clinically significant vital signs abnormalities will be considered AEs. Clinical relevance will be based on an Investigator's criteria. For each subject, every vital sign will be considered.

9.1.3.4 Physical Assessment

Physical examination findings (normal and abnormal) will be listed for each clinical trial subject. Any clinically significant abnormality developed by individual during the clinical trial and not already present at screening and/or baseline will be reported as AE.

9.1.4 PK Analysis

Anti-HAV antibody plasma and/or serum concentrations produced by the administration of the study treatment will be determined in order to establish the PK profile of the IP. The PK parameters that will be derived from the plasma and/or serum concentrations *versus* time profiles are listed in Table 9-1. All reported sampling time deviations will be taken into consideration for evaluation of PK parameters. It is recognized that there may be circumstances where PK sampling may fall outside of the time windows. The actual times for collection of PK samples will be captured in the source data and eCRF for analysis.

In the case where concentrations of anti-HAV antibodies cannot be determined due to bioanalytical or clinical reasons, these values will be set to missing for the statistical and PK analyses.

The main PK parameters of interest for this study will be $AUC_{0-\infty}$, AUC_{0-T}, C_{max}, T_{max}, λ_z, T_{half}, Cl_{TOT}/F and V_D/F.

Table 9-1 Pharmacokinetic Parameters

PK Parameter	Definition
C_{max}	Maximum observed plasma and/or serum concentration
T_{max}	Time of maximum observed plasma and/or serum concentration; if it occurs at more than one time point, T_{max} is defined as the first time point with this value
T_{LQC}	Time of last observed quantifiable plasma and/or serum concentration
AUC_{0-T}	Cumulative area under the plasma and/or serum concentration time curve calculated from 0 to T_{LQC} using the linear trapezoidal method, where T_{LQC} represents time of last observed quantifiable plasma and/or serum concentration
$AUC_{0-\infty}$	Area under the plasma and/or serum concentration time curve extrapolated to infinity, calculated as $AUC_T + C_{LQC}/\lambda_Z$, where C_{LQC} is the measured concentration at time T_{LQC}
T_{LIN}	Time point where the log-linear elimination phase begins
λ_Z	Apparent elimination rate constant, estimated by linear regression of the terminal linear portion of the log concentration *versus* time curve
T_{half}	Terminal elimination half-life, calculated as $\ln(2)/\lambda_Z$
V_D/F	Apparent Volume of Distribution, calculated as follows: $V_D / F = \left(\frac{Dose}{K_{el} * AUC_{\infty}} \right)$
Cl_{TOT}/F	Apparent Total plasma and/or serum Clearance, calculated as follows: $Cl_{TOT} / F = \left(\frac{Dose}{AUC_{\infty}} \right)$

PK parameters will be estimated using both anti-HAV antibody baseline uncorrected and baseline corrected levels. The main absorption and disposition parameters will be estimated using a non-compartmental approach with a log-linear terminal phase assumption. The trapezoidal rule will be used to estimate the area under the curve (linear trapezoidal linear interpolation) and the terminal phase will be estimated by maximizing the coefficient of determination estimated from the log-linear regression model. However, $AUC_{0-\infty}$, λ_Z, T_{half}, Cl_{TOT}/F and V_D/F parameters will be estimated for individual concentration-time profiles only when the terminal log-linear phase can be reliably characterized using the following criteria:

- Phoenix® WinNonlin® Best fit range selection
- R^2 of at least 80%

PK parameters will be listed and summarized using descriptive statistics for the PK population. N, mean (SD), 90% CI for mean, % CV, median, minimum, maximum, geometric mean (except T_{max}), and 90% CI for geometric mean (except T_{max}) will be provided.

The individual plasma and/or serum concentration/time profiles will be presented using the actual sampling times whereas the mean plasma and/or serum concentration/time profiles will be presented using the nominal sampling times.

PK analyses will be generated using validated PK software.

9.2 Determination of Sample Size

Approximately 28 healthy subjects will be enrolled and treated in this study which will provide about 20 evaluable subjects based on approximately 30% dropout rate. The sample size is chosen based on clinical considerations but not on a formal sample size calculation.

9.3 Level of Significance to Be Used

Not applicable since only descriptive statistics will be provided and there will be no statistical hypothesis testing.

9.4 Criteria for Termination of the Study

The sponsor or its representative may terminate the study at any time for scientific or corporate reasons.

If the trial is prematurely terminated or suspended for any reason, the Principal Investigator should promptly inform the trial subjects, should assure appropriate therapy and follow-up for the subjects and should inform the regulatory authority (ies) when required.

9.5 Procedure for Accounting for Missing, Unused, and Spurious Data (if applicable)

Handling of missing, unused and spurious data will be described in the SAP. All available safety, efficacy and PK data will be included in data listings.

9.6 Reporting Deviation(s) from the Statistical Analysis Plan

The detailed statistical analysis methodologies will be documented in the SAP. If there are any deviations from the originally planned analyses in the SAP, they will be fully described and justified in the protocol amendment(s) and/or final Clinical Study Report.

9.7 Subject Population(s) for Analysis

9.7.1 Safety Population:

The safety population consists of all subjects who received any amount of study drug.

9.7.2 Evaluable Population:

The evaluable population consists of all subjects who received the entire dose of IP and had no major protocol deviations that would impact the efficacy analysis up to Day 60. Any deviations from the protocol will be recorded in the protocol deviation list and evaluated before database lock.

9.7.3 PK population:

The PK population consists of all subjects who received the entire dose of the IP and who provided sufficient plasma and/or serum concentration data to facilitate calculation of PK parameters. Subjects who do not complete the PK sampling schedule may be included in the PK analysis only for the PK parameters that are judged not to be affected by the missing sample(s). This decision is to be documented and approved by the sponsor before the start of the sample analysis by the bioanalytical facility.

10 DIRECT ACCESS TO SOURCE DATA/DOCUMENTS

The data will be recorded and kept current in eCRFs by the study site personnel directly responsible for the information and reviewed for completeness by the monitor. Grifols personnel or designee can review the records.

In accordance with ICH GCP guidelines, the monitor must have direct access to the Investigator’s source documentation in order to verify the data recorded in the eCRFs for consistency and to verify adherence to the protocol, and the completeness, consistency, and accuracy of data entered. "Source documentation" includes individual subject files, separate from the eCRFs, which should be maintained and include visit dates, laboratory results, concomitant treatment, vital signs, ECGs, medical history, examinations, AEs, IP dispensing logs, and other notes as appropriate. An Investigator agrees to cooperate with the monitor to ensure that any problems noted during the course of these monitoring visits are resolved.

11 QUALITY CONTROL AND QUALITY ASSURANCE

Monitoring and auditing procedures defined/agreed by the sponsor will be followed, in order to comply with ICH GCP guidelines. Each center will be visited at regular intervals by a monitor to ensure compliance with the study protocol, ICH GCP and legal aspects. The on-site verification of the eCRF for completeness and clarity will include cross checking with source documents, and clarification of administrative matters. Query verification of data will be described in the Data Management Plan.

Representatives of regulatory authorities or of Grifols may conduct audits or inspections or audits of the Investigator study site. If an Investigator is notified of an audit or inspection by a regulatory authority, the Investigator agrees to notify the Grifols Representative immediately. An Investigator agrees to provide to representatives of a Regulatory Agency or Grifols access to records, facilities, and personnel for the effective conduct of an audit or inspection.

12 ETHICS

12.1 Institutional Review Board/Ethics Committee

Documented approval from appropriate IRBs/ECs will be obtained for all participating centers/countries prior to study start, according to ICH GCP guidelines, local laws, regulations and organizations. When necessary, an extension, amendment or renewal of the IRBs/ECs approval must be obtained and also forwarded to the sponsor. The IRBs/ECs must supply to the sponsor, upon request, a list of the IRBs/ECs members involved in the vote and a statement to confirm that the IRBs/ECs is organized and operates according to ICH GCP guidelines and applicable laws and regulations.

12.2 Ethical Conduct of the Study

The procedures set out in this protocol, pertaining to the conduct, evaluation, and documentation of this study, are designed to ensure that the sponsor and Investigator abide by ICH GCP guidelines. The study will also be carried out in keeping with applicable local law(s) and regulation(s). This may include an audit by the sponsor representatives and/or an inspection by Regulatory Authority representatives at any time. An Investigator must agree to the audit or inspection of study-related records by the sponsor representatives and/or Regulatory Authority representatives, and must allow direct access to source documents to the sponsor and/or Regulatory Authority representatives.

Modifications to the study protocol will not be implemented by either the sponsor or an Investigator without agreement by both parties. However, an Investigator may implement a deviation from, or a change to, the protocol to eliminate an immediate hazard(s) to the study subjects without prior IRB/EC/Sponsor approval/favorable opinion. As soon as possible, the implemented deviation or change, the reasons for it and if appropriate the proposed protocol amendment should be submitted to the IRB/EC/Sponsor. Any deviations from the protocol must be fully explained and documented by an Investigator.

No medical waivers for protocol inclusion/exclusion criteria will be allowed by the sponsor, and that in case the need for a change to the protocol is identified, it will be submitted as a protocol amendment to the competent regulatory authority and/or ethics committee as applicable per regulations.

12.3 Regulatory Authority Approvals/Authorizations

Regulatory Authority approvals/authorizations/notifications, where required, must be in place and fully documented prior to study start. Study information including contact information for Investigator sites responsible for conducting the study will be posted on a publicly accessible clinical registry(ies) as required by local law.

12.4 Subject Information and Consent

Subject information and ICF will be provided to Investigator sites. Prior to the beginning of the study, an Investigator must have the IRB/EC written approval/favorable opinion of the written ICF and any other written information to be provided to subjects. The written approval of the IRB/EC together with the approved subject information/ICF must be filed in the study files and a copy of the documents must also be provided to Sponsor by the Investigator site.

Written ICF must be obtained before any study specific procedure takes place. Participation in the study and date of ICF given by the subject should be documented appropriately in the subject's files. A signed copy of the subject ICF will be provided to the subject or subject's authorized representative.

12.5 Confidentiality

All records identifying the subject will be kept confidential and, to the extent permitted by the applicable laws and/or regulations, will not be made publicly available.

Subject names will not be supplied to the sponsor. Only the subject code number and subject initials will be recorded in the eCRF, and if the subject's name appears on any other document (e.g., pathologist report), it must be retracted before a copy of the document is supplied to the sponsor. Study findings stored on a computer will be stored in accordance with local data protection laws. Subjects will be informed in writing that representatives of the sponsor, IRB/EC, or Regulatory Authorities may inspect their medical records and personal health information to verify the information collected, and that all personal information made available for an audit or inspection will be handled in strictest confidence and in accordance with local data protection laws.

If the results of the study are published, the subject's identity will remain confidential.

An Investigator will maintain a list to enable subjects' records to be identified.

13 DATA HANDLING AND RECORD KEEPING

13.1 Data Handling

The study data will be recorded and kept current in the eCRF by the site study personnel directly responsible for the information. Entries made in the eCRF must be verifiable against source documents, or have been directly entered into the eCRF, in which case the entry in the eCRF will be considered the source data. The data in the eCRF will be monitored at the site by Grifols representatives at regular intervals and reviewed for completeness and compared with the source documents. Examples of acceptable source documents include individual subject medical records, prospective information gathered on source documentation worksheets, lab reports and other diagnostics pertinent to this study which are separate from the eCRFs. The listing of types of source documents which will be defined in the source data agreement will be filed in trial master file.

All adverse events and SAEs must be recorded. All SAEs must be recorded on the SAE form. The SAE form must be kept in site records with a copy provided to the designated person as detailed in the study file.

13.2 Record Retention

At study completion, all study data will be transferred to Grifols according to ICH GCP guidelines, local laws, regulations, and Grifols requirements. The study file and all source data should be retained until notification is given by the sponsor for destruction.

An Investigator is required by ICH GCP guidelines to retain the study files. If an Investigator moves, withdraws from an investigation or retires, the responsibility for maintaining the records may be transferred to another person (e.g., other investigator). Grifols must be notified in writing of the person responsible for record retention and the notification will be retained in the sponsor study file and the Investigator site file.

14 FINANCING AND INSURANCE

In the event of subject injury as a direct result of either administration of IP or any non-standard of care study procedure, Sponsor will pay for the costs of treatment, provided the subject has followed the instructions given by a study Investigator and the illness or injury is not due to the natural progression of any conditions existing before the subject participated in the study. Financial compensation for such things as lost wages, disability, or discomfort due to any research-related injury is not available.

Sponsor shall maintain comprehensive general liability insurance or self-insurance in amounts adequate to cover any damage, demand, claim, loss or liability caused or incurred by Sponsor, or as otherwise required by applicable laws and/or regulations.

15 PUBLICATION POLICY

Institution and the Investigator agree that the first publication shall be made in conjunction with the presentation of a joint publication of the study results. If such a publication is not submitted within twelve (12) months after conclusion of the study at the investigational site or after Grifols confirms there will be no joint publication, then institution and/or Investigator shall have the right, at their discretion, to publish, either in writing or orally, the results of the study performed under the protocol, subject to the conditions outlined below:

- The results of the study will be reported in the publicly accessible registry(ies).
- Institution and/or Investigator shall furnish Grifols with a copy of any proposed publication at least thirty (30) days in advance of the date of submission for publication.
- Within said thirty (30) day period, Grifols shall:
  - Review such proposed publication for Confidential Information (other than Study results) and for subject information subject to the Health Insurance Portability and Accountability Act of 1996 ("HIPAA") and other applicable privacy laws;
  - Review such proposed publication for the unauthorized use of the name, symbols and/or trademarks of Grifols;
  - By written notice to the Investigator, identify with specificity the text or graphics in such proposed publication that Grifols contends contains Confidential Information, protected subject information, or the unauthorized use of Grifols' name, symbols and/or trademarks so that the proposed publication may be edited appropriately to remove such text or graphics before publication; and
  - By written request, Grifols may delay proposed publications up to sixty (60) days to allow Grifols to protect its interests in Grifols Inventions described in such publications.
- Institution and/or Investigator shall give Grifols the option of receiving an acknowledgment for its sponsorship of the study in all such publications or presentation.

16 REFERENCES

1. Wasley A, Fiore A, Bell BP. Hepatitis A in the era of vaccination. Epidemiol Rev. 2006;28:101-111.

2. Centers for Disease Control and Prevention. Prevention of hepatitis A through active or passive immunization: recommendations of the Advisory Committee on Immunization Practices. MMWR. 2006;55(RR-07):1-23.

3. Klevens RM, Kruszon-Moran D, Wasley AM, Gallagher K, McQuillan GM, Kuhnert WL, et al. Seroprevalence of hepatitis A antibodies in the United States: results from the National Health and Nutrition Examination Survey. Public Health Rep. 2011;126(4):522-532.

4. Centers for Disease Control and Prevention. Vaccination coverage among children aged 19-35 months – United States, 2015. MMWR. 2016 65(39):1065-1071.

5. Klevens RM, Miller JT, Iqbal KI, Thomas A, Rizzo EM, Hanson H, et al. The evolving epidemiology of hepatitis A in the United States. Arch Intern Med 2010; 170(20):1811-18.

6. Klevens RM, Liu SJ, Roberts H, Jiles RB, Holmberg SD. Estimating acute viral hepatitis infections from nationally reported cases. Am J Public Health. 2014;104(3):482-7.

7. Viral Hepatitis Surveillance, United States, 2015.https://www.cdc.gov/hepatitis/statistics/2015surveillance/pdfs/2015HepSurveillanceRpt.pdf. Accessed on 27 July 2017.

8. Fiore AE, Wasley A, Bell BP. Prevention of hepatitis A through active or passive immunization: recommendation of the Advisory Committee on Immunization Practices (ACIP). MMWR 1999; 48(RR12):1-37.

9. Product Information: GamaSTAN(R) S/D intramuscular injection solution, immune globulin (human) intramuscular injection solution. Grifols Therapeutics Inc., Research Triangle Park, NC. 2013

10. Cohn EJ, Strong LE, Hughes WL, Mulford DJ, Ashworth JN, Melin M, et al. Preparation and properties of serum and plasma proteins. IV. A system for the separation into fractions of the protein and lipoprotein components of biological tissues and fluids. J Am Chem Soc 1946 Mar;68(3):459-75.

11. Cohn EJ, Gurd FRN, Surgenor DM, Barnes BA, Brown RK, Derouaux G, et al. A system for the separation of the components of human blood: quantitative procedures for the separation of the protein components of human plasma. J Am Chem Soc. 1950 Jan; 72(1):465-74.

12. Barnette D, Roth NJ, Hotta J, Cai K, Gall M, Hartwell R, Kent JD, Willis T. Pathogen safety profile of a 10% IgG preparation manufactured using a depth filtration-modified process. Biologicals. 2012 Jul;40(4):247-253.

13. Lebing W, Remington KM, Schreiner C, Paul HI. Properties of a new intravenous immunoglobulin (IGIV-C, 10%) produced by virus inactivation with caprylate and column chromatography. Vox Sang. 2003 Apr;84(3):193-201.

14. Fudenberg HH. Sensitization to immunoglobulins and hazards of gamma globulin therapy. In: Merler E, editor. Immunoglobulins: biologic aspects and clinical uses. Washington DC: Nat Acad Sci; 1970. p. 211-220.

15. Kroger AT, Vázquez DJ. General recommendations on immunization. Best Practices Guidance of the Advisory Committee on Immunization Practices (ACIP). https://www.cdc.gov/vaccines/hcp/acip-recs/general-recs/downloads/general-recs.pdf Accessed on 27 July 2017

16. Immune Globulin (Human) – GamaSTAN Investigator's Brochure, Grifols Therapeutics Inc.

17 APPENDICES

Appendix 1 Schedule of Study Procedures and Events

Visits Procedures and Evaluation	Screening Visit Days -28 to -2	Day -1	Day 1[a]	Day 2	PK Sampling Visits[b] Days 3, 4, 5, 7, 10, 14, 21, 28, 60, 79, 115	Final Visit (Day 150)[b]/ Early Discontinuation Visit
Informed consent	X					
Admission to clinical research unit		X				
Inclusion/exclusion criteria	X					
Continued eligibility verification		X				
Medical history & demographics	X					
Height and weight	X					
ECG	X	X				
Full physical exam [c]	X					X
Symptom-directed physical exam		X		X	X (Days 5, 28, and 60 visits only)	
Vital signs [d]	X	X	X		X (Days 5, 28, and 60 visits only)	X
Clinical lab assessments [e]	X	X			X (Days 5, 28, and 60 visits only)	X
Coagulation tests (INR, aPTT)	X					
Immunoglobulin A (IgA)	X					
Pregnancy test [f]	X	X				
Urine alcohol and drug panel testing [g]	X	X				
Virus safety testing [h]	X					
Virus safety retain samples [i]			X			X
IP injection			X			
Injection site evaluation			X	X		

Visits Procedures and Evaluation	Screening Visit Days -28 to -2	Day -1	Day 1[a]	Day 2	PK Sampling Visits[b] Days 3, 4, 5, 7, 10, 14, 21, 28, 60, 79, 115	Final Visit (Day 150)[b]/ Early Discontinuation Visit
PK sampling (anti-HAV antibody level)			X	X	X	X
Concomitant medications	X	X	X	X	X	X
Adverse events	X	X	X	X	X	X
Discharge from clinical research unit				X		

[a] Collection of virus safety retain samples on Day 1 will be performed prior to study treatment administration. Vital signs will be measured prior to study drug injection, approximately 60 minutes (±10 minutes) and 12 hours (±10 minutes) after study treatment administration. Injection site evaluation will be performed approximately 60 minutes after study treatment administration. PK blood sampling on Day 1: prior to study drug injection, 60 minutes (±10 minutes) post study drug injection and 12 hours post study drug injection (a window of ±1 hour is permitted)

[b] PK sampling visits will be conducted as close as possible to the exact time points. The PK sampling visits: Day 2 (window of ±1 hour); Days 3, 4 and 5 (visits have a window of ±4 hours); Days 7, 10, and 14 (all 3 visits have a window of ±1 day); Days 21 and 28 (both visits have a window of ±2 days); Days 60, 79 and 115 (all 3 visits have a window of ±4 days); Day 150 (a window of ±7 days)

[c] Full physical examination (excluding breast and genitourinary examination)

[d] Vital signs include systolic blood pressure, diastolic blood pressure, body temperature, heart rate, respiratory rate

[e] Clinical laboratory assessments will include hematology, chemistry, and urinalysis. Hematology panel: hemoglobin, hematocrit, platelets, red blood cell count, and white blood cell count (absolute and % with differential). Chemistry panel: Sodium, potassium, creatinine, BUN, calcium, LDH, AST, ALT, GGT, ALP, glucose, total bilirubin, direct and indirect bilirubin. Urinalysis panel: Microscopic evaluation is done only with cause, pH, protein, glucose, and blood.

[f] Dipstick urine pregnancy test at screening visit and on Day -1 administration.

[g] Urine alcohol and drug panel testing (drug panel includes marijuana, opiates, cocaine, amphetamines, methamphetamines and benzodiazepines)

[h] Virus safety testing: HAV RNA, HBV DNA, HCV RNA, HIV-1 RNA and B19V DNA by NAT methods as well as HAV antibody differential (IgM/IgG), hepatitis B core antibody differential (IgM/IgG), hepatitis C antibody, HIV-1/-2 + Group O antibody and B19V antibody differential (IgM/IgG) testing by serological methods.

[i] Virus safety retain samples: serum and/or plasma samples for HAV RNA, HBV DNA, HCV RNA, HIV-1 RNA and B19V DNA testing by NAT methods as well as HAV antibody differential (IgM/IgG), hepatitis B core antibody differential (IgM/IgG), hepatitis C antibody, HIV-1/-2 + Group O antibody and B19V antibody differential (IgM/IgG) testing by serological methods will be collected on Day 1 prior to IP injection and at final visit (Day 150)/early discontinuation visit. These samples will be tested only if the subject exhibits clinical signs and symptoms consistent with HAV, HBV, HCV, HIV or B19V infection while participating in the study. Virus safety retain samples will be retained until all analyses in support of the study are complete.