Statistical Analysis Plan

Protocol Number: GTI1307

A Prospective, Multicenter, Randomized, Double-Blind, Placebo-Controlled Study to Evaluate the Safety and Efficacy of Preoperative Antithrombin Supplementation in Patients Undergoing High-Risk Cardiac Surgery with Cardiopulmonary Bypass

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List of Abbreviations

ACT	activated clotting time
ADR	adverse drug reaction
AE	adverse event
AKI	acute kidney injury
ALT	alanine aminotransferase
ANCOVA	analysis of covariance
AR	adverse reaction
AST	aspartate transaminase
AT	antithrombin III
AT-III (Human)	antithrombin III (Human)
ATC	anatomical Therapeutic Chemical
BMI	body mass index
CABG	coronary artery bypass graft
CI	confidence interval
CK	creatine phosphokinase
CK-MB	creatine phosphokinase MB isoenzyme
CPB	cardiopulmonary bypass
CSR	clinical study report
CVP	central venous pressure
dL	deciliter
DVT	deep vein thrombosis
ECG	electrocardiogram
eCRF	electronic case report form
F1+2	prothrombin fragment 1+2
FFP	fresh frozen plasma
g	gram
Hb	hemoglobin
IABP	intra-aortic balloon pump
ICU	intensive care unit

INR	international normalized ratio
IP	investigational product
ITT	intention-to-treat
IU	international unit
kg	kilogram
L	liter
LBBB	left bundle branch block
LLOQ	lower limit of quantification
LV	left ventricular
MedDRA	medical dictionary for regulatory activities
mg	milligram
MI	myocardial infarction
mITT	modified intention-to-treat
mL	milliliter
mmol	millimole
NaCl	sodium chloride
NAT	nucleic acid testing
PCC	prothrombin complex concentrate
PLT	platelet
PP	per-protocol
PT	preferred term
RBC	red blood cell
SAE	serious adverse event
SAP	statistical analysis plan
SD	standard deviation
SOC	system organ class
TAT	thrombin-antithrombin complex
TEAE	treatment emergent adverse event
USP	United States pharmacopeia
WHO-Drug	world health organization drug classification

1 Purpose of the Analysis

The purpose of this statistical analysis plan (SAP) is to outline the planned analyses to support the completion of the clinical study report (CSR) for protocol GTI1307. Exploratory analyses not identified or defined in this SAP may be performed to support the clinical development program or manuscript. Any post-hoc or unplanned analyses performed and not identified in this SAP will be documented in the respective CSR.

2 Introduction

Cardiac surgery with cardiopulmonary bypass (CPB) causes a massive consumptive coagulopathy and thrombotic complications in a similar way that disseminated intravascular coagulation does. Therefore, suppressing excessive hemostatic activation is a plausible therapeutic strategy that may impact postoperative morbidity.

Several studies have consistently reported the association of low postoperative antithrombin (AT) levels with poor clinical outcomes and that preoperative AT supplementation is able to maintain AT levels within a normal range throughout the postoperative period. Conversely, past studies have also highlighted the potential anticoagulant action of purified AT. Likewise, a dose dependent trend towards higher bleeding rates and a trend towards an increased incidence of postoperative complications (namely bleeding related) in AT treated patients reaching the intensive care unit (ICU) with exceptionally high AT levels has been reported. The latter observations generate the hypothesis that an AT supplementation strategy aimed to avoid both excessively low (<58%) and excessively high (>100%) postoperative AT levels may represent an effective therapeutic goal to ameliorate the occurrence of bleeding and thrombotic events, respectively. However, these assumptions have been derived from post-hoc analysis and small studies performed with a limited number of patients.

Thus, this clinical study has been designed to evaluate the safety and efficacy of a preoperative treatment with AT supplementation (Antithrombin III [Human]) in subjects who undergo high-risk cardiac surgery with CPB targeting AT levels in such a way to avoid both excessively low and excessively high postoperative AT activity levels and to assess if management of AT levels in this range may decrease negative clinical outcomes during the ICU and hospital stay. The present clinical trial is intended to assess if directed preoperative AT supplementation will show favorable trends in decreasing the incidence of a composite of major morbidity when compared to placebo.

Antithrombin III (Human) with the trade name of THROMBATE III® is marketed in several countries for replacement therapy in subjects with hereditary deficiency of AT during pregnancy or surgery, or if at risk of thromboembolism. For simplicity, Antithrombin III (Human) is abbreviated as AT-III (Human) in this SAP.

3 Study Design and Objectives

3.1 Overall Study Design and Plan

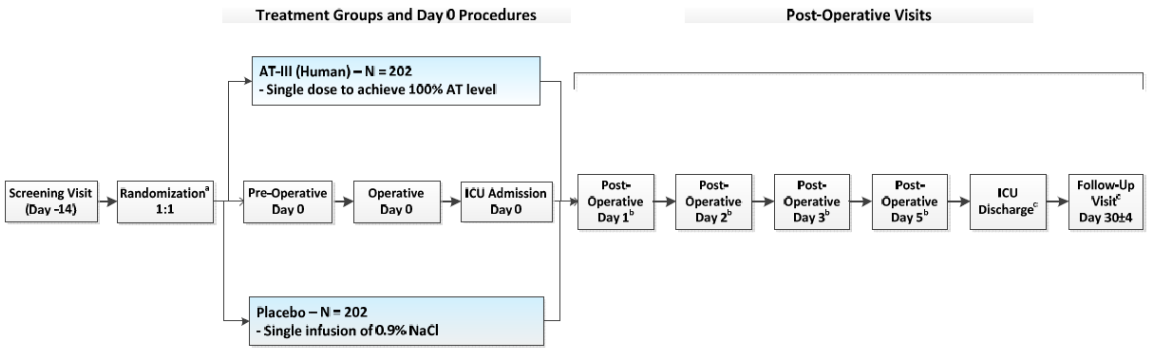
This study will be a prospective, multicenter, randomized, double-blind, placebo-controlled study. Participating subjects will be 404 adult subjects undergoing cardiac surgery with CPB who will be randomized into two treatment groups preoperatively in a 1:1 ratio: AT-III (Human) treatment group and Placebo control group. All subjects will be stratified by their previous history of myocardial infarction (MI). Subjects undergoing complex/combined procedures will not be prescreened or screened for AT levels using local lab values to assess eligibility. Only subjects undergoing isolated coronary artery bypass graft (CABG) or single valve repair/replacements and not receiving preoperative heparin may be prescreened locally for AT levels (must be AT <80%).

Subjects randomized to AT-III (Human) treatment group will receive preoperative supplementation with AT-III (Human) designed to achieve an absolute increase (Δ) of 20% above pretreatment AT levels. Subjects randomized to Placebo control group will receive 0.9% Sodium Chloride (NaCl) for Injection, USP (United States Pharmacopeia).

The maximum study duration for a given subject will be approximately 7 weeks (48 days). There will be 2 weeks for prescreening screening, and randomization into one of the two study groups (AT-III [Human] treatment or Placebo control). After the 2-week prescreening and screening period, the cardiac surgery procedure will take place with the preoperative administration of AT-III (Human) or Placebo supplementation. After surgery, the subject will be admitted to the ICU and be followed for approximately 1 month (30±4 days).

Both the primary efficacy endpoint (percentage of subjects with any component of the major morbidity composite) and the secondary endpoints will be evaluated for each subject after each cardiac surgery procedure, at the ICU admission, ICU discharge, and during a follow-up period of approximately 1 month (30±4 days).

The overall study schema is shown in diagram below.



[a] Randomization on Preoperative Visit (Day 0) may occur 24 hours before the day of surgery (Day 0).
[b] If this visit coincides in time with the ICU discharge, the only assessments to be performed are those detailed in the ICU discharge visit. Regardless of when the ICU discharge takes place, AT activity levels should be measured on Postoperative days 1, 2, 3, and 5.
[c] If the ICU stay is longer than 30 days, the assessments for both ICU Discharge visit and Follow-up visit (Day 30±4 days) will be performed at Day 30±4 days. There will be no additional ICU discharge visit and follow-up visit.

3.1.1 Treatment Administration

Every subject allocated in the AT-III (Human) treatment group will receive, immediately after anesthesia induction, a single dose of AT-III (Human) sufficient to achieve an absolute increase (Δ) of 20% above pretreatment AT levels according to the following formula, where IU means international unit:

$$AT-III\ dose(IU)\ required = \frac{(20)\times(subject\ weight\ in\ kg)}{1.4}$$

The only variable in the equation is the subject's body weight. For calculating AT-III (Human) dose, subject's weight obtained during the Screening visit will be used. Since the original protocol required that patients have an AT level < 80% for eligibility, the original equation differed (i.e., AT-III dose (IU) required = (100 - actual AT level in subject) x (subject weight in kg) / 1.4), as the aim was to raise AT level to 100%; given the near impossibility to recruit anyone with a subnormal AT level from the general pre-surgical population the above formula was instituted through protocol Amendment 3 (Version 4.0), 2 April 2015.

AT-III (Human) or Placebo (0.9% NaCl) will be prepared according to the volume calculated by unblinded pharmacist or designee.

AT-III (Human) will not be administered postoperatively unless the investigator decides otherwise for clinical reasons (if this is the case, the subject will be excluded from the per-protocol population for efficacy analysis and also from the mITT population as a sensitivity analysis of the primary efficacy variable).

3.2 Study Objectives

3.2.1 Primary Efficacy Objective

The primary objective of this clinical study is to compare the percentage of subjects with any component of a major morbidity composite between 2 groups of subjects randomly allocated to receive preoperative supplementation of AT-III (Human) or Placebo.

3.2.2 Secondary Efficacy Objectives

The secondary objectives of this clinical study are the following:

1. To compare postoperative AT levels at the ICU admission between the AT-III (Human) treatment group and Placebo control group
2. To compare the following postoperative outcomes between the AT-III (Human) treatment group and Placebo control group:

    - Postoperative chest-drain blood loss in the first 12 and 24 hours after surgery
    - Transfusion requirements
    - Need for surgical reexploration
    - Low cardiac output syndrome
    - Myocardial infarction (MI)
    - Stroke
    - Acute kidney injury (AKI)
    - Arterial or venous thromboembolic events
    - Infections
    - Prolonged mechanical ventilation (>24 hours)
    - All-cause postoperative mortality
    - ICU stay duration
    - Prolonged ICU stay (>6 days)
    - Length of hospital stay

3.2.3 Safety Objectives

Safety objectives include evaluation of AT-III (Human) for clinical safety including adverse events (AEs), risks for bleeding, clinical laboratory testing, physical exam, and vital signs.

4 Study Variables

4.2 Efficacy Variables

4.2.1 Primary Efficacy Variable

Percentage of subjects with any component of a **major morbidity composite** defined as a composite of any one or more of the following events (up to Day 30±4 days):

a. Postoperative **mortality** (defined as all deaths occurring within 30 days of the operation or occurring during the primary hospitalization).
b. **Stroke** (defined as a clinical diagnosis of focal or global neurological deficit of abrupt onset caused by a disturbance in cerebral blood supply).
c. **Acute kidney injury (AKI)** (defined as an increase of serum creatinine levels to >2.0 mg/dL and twice the baseline level or a new requirement for dialysis postoperatively).
d. **Surgical reexploration** (defined as having to return to the operating room because of bleeding, tamponade, graft occlusion or any other cardiac reason).
e. **Arterial or venous thromboembolic event** (consisting of perioperative MI, mesenteric infarction, peripheral thromboembolism, acute coronary graft thrombosis, intracardiac thrombosis, deep vein thrombosis [DVT], and pulmonary embolism).
f. **Prolonged mechanical ventilation** (defined as mechanical ventilation >24 hours).
g. **Infection** (defined as presence of deep sternal-wound infection and/or bloodstream infections).

4.2.2 Secondary Efficacy Variables

1. AT activity (levels) determinations:

To maintain the study blind, No unblinded AT concentration results will be provided to study sites or blinded Grifols personnel until after closure of the study database.

a. AT activity levels will be measured for both AT-III (Human) treatment and Placebo control groups at the following time points:
    - Prescreening visit (local lab)*
    - Screening visit (local lab)*§
    - Screening visit
    - Preoperatively
    - Immediately before study drug administration
    - Immediately after study drug administration
    - Postoperatively at ICU admission
    - Postoperatively Day 1
    - Postoperatively Day 2
    - Postoperatively Day 3
    - Postoperatively Day 5
    - ICU discharge
    - 1-month follow-up visit

* Only applicable for subjects undergoing isolated CABG or single valve repair/replacements (will be needed for eligibility only if subject not receiving preoperative heparin [see entry criteria]).

§ This AT (local lab) determination will not be required if the Prescreening visit is conducted.

b. Percentage of subjects with AT levels of 58% or higher at the ICU admission will be calculated for both treatment groups and compared.

2. **Chest-drain blood loss** (mL) in the first 12 and 24 hours of chest closure is defined as the amount of blood collected in the cardiotomy reservoir from chest closure through the following 12 and 24 hours measured.
3. **Transfusion requirements** will be measured as the number of units of Fresh frozen plasma (FFP), red blood cells (RBCs), platelets (PLTs), cryoprecipitate, fibrinogen concentrate, prothrombin complex concentrates (PCCs), plasma, and whole blood from the day of ICU admission and throughout the follow-up period.

A specific perioperative transfusion algorithm will be applied:

Standardized Transfusion Algorithm

Red blood cells (RBC):

Administration of RBCs should be performed one unit at a time after which the hemoglobin (Hb) levels should be reanalyzed to check the suitability for the next unit.

- Hb <6.0 g/dL (≤3.72 mmol/L [millimoles/liter]): Transfusion is mandatory
- Hb = 6.0-8.0 g/dL (3.72-4.96 mmol/L): Transfusion is optional
- Hb = (8.0-10.0 g/dL). Transfusion is acceptable only if clinically justified*
- Hb >10.0 g/dL: Transfusion is not permitted.

* One or more of the following clinical justifications must be provided:

- Central venous saturation (Swan Ganz / Central Venous Pressure (CVP) <60% with an arterial oxygen saturation of at least 90%.
- Cardiac Index <2.0 unresponsive to inotropic or mechanical support (catecholamine, intra-aortic balloon pump [IABP], etc.).
- Signs / symptoms of organ ischemia, such as: change in ST-segment or left ventricular wall motion abnormalities (LV).

Coagulation products:

Only if subject is bleeding 4 mL/kg/hour or more over at least 30 minutes or 1.5 mL/kg/hour or more over 2 consecutive hours.

- FFP (10-15 mL/kg) / PCC: If subject is actively bleeding and presents an international normalized ratio (INR) ≥1.5.
- PLT: 1 apheresis unit or 4-6 pooled units if the PLT count is <100,000/µL.
- Cryoprecipitate / Fibrinogen concentrates: Administration of 8-10 units of cryoprecipitate or 2 grams of fibrinogen concentrate if the concentration of fibrinogen ≤1.5 g/L.

Shed blood collected during surgery can be retransfused according to institutional practice. Retransfusion of chest-drain collected blood is not permitted.

4. Percentage of subjects needing **surgical reexploration** defined as having to return to the operating room because of bleeding, tamponade, graft occlusion or any other cardiac reason will be considered from chest closure to the end of the follow-up period.
5. Percentage of subjects with **low cardiac syndrome** defined as the need for major inotropic support or intra-aortic balloon pump will be considered from the day of ICU admission and throughout the follow-up period.
6. Percentage of subjects with postoperative **MI** defined through enzymatic criteria plus new Q-waves at the electrocardiogram (ECG) will be considered from the day of ICU admission and throughout the follow-up period.

   A perioperative MI is diagnosed by the following criteria:

   - (0-24 hours postoperation):

     The creatine phosphokinase-MB isoenzyme CK-MB (or creatine phosphokinase [CK] if MB not available) must be greater than or equal to 5 times the upper limit of normal, with or without new Q waves present in two or more contiguous ECG leads. No symptoms required.

   - (>24 hours postoperation): Documented by at least one of the following criteria:

   a. Evolutionary ST-segment elevations
   b. Development of new Q-waves in two or more contiguous ECG leads
   c. New or presumably new left bundle branch block (LBBB) pattern on the ECG
   d. CK-MB (or CK if MB not available) ≥3 times the upper limit of normal
7. Percentage of subjects with postoperative **stroke** defined as a clinical diagnosis of focal or global neurological deficit of abrupt onset caused by a disturbance in cerebral blood supply will be considered from the day of ICU admission and throughout the follow-up period.
8. Percentage of subjects with **AKI** defined as an increase of serum creatinine levels to >2.0 mg/dL and twice the baseline levels or new requirement for dialysis postoperatively, will be considered from the day of ICU admission and throughout the follow-up period. A new requirement for postoperative dialysis may include hemodialysis, peritoneal dialysis, and any form of ultrafiltration.
9. Percentage of subjects with **arterial or venous thromboembolic events** defined as perioperative MI, stroke, mesenteric infarction, peripheral thromboembolism, acute

coronary graft thrombosis, intracardiac thrombosis, DVT, and pulmonary embolism, will be considered from the day of ICU admission and throughout the follow-up period.

10. Percentage of subjects with **infections** defined as presence of deep sternal-wound infections involving muscle, bone or mediastinum or bloodstream infections from the day of ICU admission and throughout the follow-up period.

    The diagnosis of infection must have at least one of the following conditions:

    a. Wound opened with excision of tissue or reexploration of the mediastinum
    b. Positive culture
    c. Treatment with antibiotics

11. Percentage of subjects with **prolonged mechanical ventilation** defined as the need for mechanical ventilation >24 hours from the day of ICU admission and throughout the follow-up period. Duration of mechanical ventilation is cumulative, i.e., summed if reintubation medically necessary during hospitalization.

12. Percentage of subjects with **all-cause postoperative mortality** defined as death during the same hospitalization as surgery or after discharge and throughout the follow-up period.

13. **ICU stay** duration (days) defined as the ICU discharge date minus the ICU admission date plus 1 day. If the discharge date is beyond the follow-up period and only '>1 month' is recorded then it will be reported as is in data listing; and date of last follow-up minus the ICU admission date plus 1 day or 34 days, whichever is longer, will be assigned for the statistical summary and analysis.

14. Percentage of subjects with **prolonged ICU stay** defined as >6 days will be considered from ICU admission.

15. **Length of hospital stay** (days) defined as the hospital discharge date minus the surgery date plus 1 day. If the discharge date is beyond the follow-up period and only '>1 month' is recorded then it will be reported as is in data listing; and date of last follow-up minus the surgery date plus 1 day or 34 days, whichever is longer, will be assigned for the statistical summary and analysis.

4.3.3 Safety Variables

1. Adverse events, serious adverse events (SAEs), and discontinuations due to AEs.
2. Risk for bleeding (will be assessed by monitoring hematocrit levels, urine dipstick tests, and any clinical adverse events for bleeding).
3. Physical examination and vital signs.
4. Clinical laboratory parameters: Hematology (RBC count, white blood cell count, hematocrit, Hb, and PLT, etc.) and clinical chemistry (creatinine, total bilirubin, alanine transferase [ALT], and aspartate transaminase [AST], etc.).

5. Viral retention samples:

   Blood samples for viral nucleic acid testing (NAT) and viral serology testing will be collected throughout the 30-day follow-up but will be tested only if the subject exhibits clinical signs and symptoms of viral infection during the one-month follow-up period.

5 General Statistical Considerations

All analyses will be conducted using SAS Version 9.4 or higher.

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

5.1 Data Handling

Unless otherwise noted, if an observation is missing at a specific scheduled visit/time point, the value at that visit/time point will not be imputed and will be set to missing.

Baseline in general will be defined as the last non-missing measurement taken prior to the start of the study treatment.

For ICU stay duration and length of hospital stay, if the discharge date is beyond the follow-up period and only '>1 month' is recorded then it will be reported as is in data listings; and the date of last follow-up minus the ICU admission date or surgery date, respectively, plus 1 day or 34 days, whichever is longer, will be assigned for the statistical summary and analysis.

If a patient was mis-stratified with regards to myocardial infarction, the patient will be analyzed according to the correct stratum (accurate stratum).

5.1.1 Adverse Events

Treatment emergent adverse events (TEAEs) will be characterized based on AE onset date/time relative to each subject's study treatment start date/time.

Adverse events with incomplete start dates will be considered TEAEs if:

- Day and month are missing and the year is equal to or after the year of study treatment start date;
- Day is missing, month is present and the year is after the year of study treatment start date;
- Day is missing and the year is equal to the year of study treatment start date and the month is equal to or after the month of study treatment start date;

- Day, month, and year are all missing.

5.1.2 Prior and Concomitant Medications

The following convention will be used for missing or partial end date information in order to determine whether a medication is prior or concomitant:

The unknown portions of a medication end date will be assumed to be as late as possible. If a medication end date is incomplete but the month/year of medication end date is prior to the month/year of the start of study treatment, then the medication will be considered a prior medication. If a medication end date is incomplete but the month/year of medication end date is the same as the month/year of the start of study treatment, then the medication will be considered a concomitant medication. All other incomplete medication end dates and all medications with missing end dates will be assumed to be concomitant medications.

5.1.3 Clinical Laboratory

For the summaries of post-baseline results only measurements from scheduled visits will be considered, although all values will be reported in data listings.

5.1.4 AT Levels

The lower limit of quantification (LLOQ) for AT Level in serum using an antigenic content assay is 20%. All AT Level values obtained in the study are expected to be above LLOQ considering they reflect both endogenous AT and infused AT-III (Human) exogenously.

For summary of AT levels, any missing or not valid concentration values of AT level will be treated as missing without imputation.

5.2 Analysis Populations

Intention-to-treat (ITT) population will include all subjects who are randomized. It will be used for the analyses of inclusion/exclusion criteria and protocol deviation.

Modified intention-to-treat (mITT) population will include all subjects who are randomized, receive any amount of study drug, and are operated on. It will be used for listings and analyses of operative assessment and efficacy data.

Per-protocol (PP) population will include all mITT subjects with no major protocol deviations that might have impact on the primary efficacy assessment (to be defined in a blinded data review meeting prior to database lock), including postoperative AT administration per investigator's discretion. It will be used for the sensitivity analysis of the primary efficacy variable.

Safety population will include all subjects who receive any amount of study drug. It will be used for listings and analyses of demographic and baseline characteristic, serum pregnancy test, medical history, prior and concomitant medication (except heparin use which is considered efficacy data), study drug infusion, treatment compliance, and safety data. Difference between each subject's planned and actual treatments will be summarized in the CSR should there be any.

5.3 Sample Size

Assuming that percentage of subjects with major morbidity composite is 22.0% in Placebo group and 14.3% in AT-III treatment group (i.e., AT-III treatment group has a 35% relative reduction comparing to the Placebo control group), 192 subjects per treatment arm (total 384 subjects) are needed to have 70% power to detect a trend at alpha = 0.15. Considering 5% dropout rate, it is estimated that 404 subjects (202 subjects per treatment arm) need to be randomized.

5.4 Randomization and Blinding

Subjects will be randomized into one of two treatment groups: AT-III (Human) treatment group and Placebo control group. Subjects will be stratified by their previous history of MI. Unblinded pharmacist or designee will obtain the treatment assignment based on a computer-generated randomization schedule.

To minimize the risk of unblinding, all subjects will receive the same total volume for all treatments with no visible differences in the external appearance of treatments, infusion bags will be covered with a non-transparent sleeve, and opaque intravenous (IV) tubing will be utilized. The unblinded study pharmacist, or designee, will prepare investigational product (i.e., AT-III [Human] and placebo) at each site. Monitoring of activated clotting time (ACT) for subject safety can potentially unblind an investigator if performed immediately before AND immediately after investigational product (IP) administration. Thus, to avoid potential unblinding, investigators should restrain from monitoring ACT values immediately before AND immediately after IP administration. Individual ACT determinations before OR after IP administration is not considered as potentially unblinding and is therefore permitted. However, if ACT monitoring is done prior to IP administration care must be taken to ensure blinded study personnel are not unblinded. Grifols Clinical Trial Materials personnel and Grifols Pharmacovigilance personnel will have access or be privy to treatment assignments.

Emergency unblinding instructions will be located in the Pharmacy Manual (to be used only if required).

Investigators must not draw any blood samples for AT levels outside the requirements of the protocol. Results from protocol-specified AT, thrombin-antithrombin complex (TAT), and prothrombin fragment 1+2 (F1+2) measurements will not be provided to the investigator or blinded sponsor personnel in order to ensure maintenance of the treatment blind.

6 Subject Disposition

Subject disposition will include the number and percentage of all subjects screened, in the populations of ITT (randomized), safety, mITT, and PP, and completing the study by treatment group and overall.

The number and percentage of subjects discontinuing from the study will be summarized by reason for discontinuation by treatment group and overall. The number and percentage of screening failures will be summarized for reasons of ineligibility.

Disposition status will be listed for all subjects.

7 Protocol Deviations

Protocol deviations will be identified during the study and evaluated before database lock. The type/category of protocol deviations and severity (i.e., minor or major) will be listed and summarized for ITT population by treatment group and overall.

8 Demographics and Medical History

Demographic (age, age categories [<65 versus >=65 years], gender, race, and ethnicity, previous MI) and baseline characteristics such as weight, height, body mass index (BMI), smoking status, Ejection Fraction, serum pregnancy test, baseline AT activity level (i.e., last available AT activity level before study treatment) (central laboratory) will be listed and summarized using descriptive statistics for safety population by treatment group and overall. AT activity levels (local laboratory) from pre-screening, screening, and pre-randomization for all screened subjects will only be listed.

Medical case and relevant medical history, surgical history, topical hemostats medication history will also be summarized for safety population by treatment group and overall.

9 Treatments and Medications, Operative Assessments

9.1 Prior and Concomitant Medications

Concomitant medications and those taken within the last month (including transfusions of blood or any blood-derived product) prior to the Screening visit must be recorded in the electronic case report form (eCRF). Prior medications are defined as any medication which ended before the start of study treatment. Concomitant medications are defined as any medication which is started on or after the start of study treatment or any medication taken prior to the study treatment and continued after the start of study treatment.

Summaries of all medications taken during the course of the study will be presented in tabular form and coded using Anatomical Therapeutic Chemical (ATC) classification codes via the World Health Organization Drug classification (WHO-Drug) Dictionary. All medications will be summarized for safety population by treatment group and overall and sorted alphabetically by medication class (i.e., ATC Level 2) and medication sub-

class (i.e., ATC level 4). If the ATC Level 4 term is missing, the ATC Level 3 term will be used in the medication summary table and data listing. For the summary tables, if a subject has taken a medication more than once, the subject will be counted only once at the ATC level. All medications will be listed by subject.

Heparin use recorded on the separate Heparin Administration eCRF form will be listed and summarized separately from other medications based on the mITT population, since it is closely associated with efficacy and is not a safety variable in this study.

9.2 Extent of Exposure

For each treatment, total AT-III actual dose (IU), total actual volume infused (mL), study drug infusion duration (min) will be summarized. Actual AT-III Dose (IU) equals to calculated AT-III dose (IU) * total volume infused (mL) / total volume prepared (mL). Duration of infusion (min) is the difference between the infusion end date/time and the start date/time [(infusion end date/time – infusion start date/time].

9.3 Treatment Compliance

The treatment compliance is calculated as [total volume infused / total volume prepared]*100%. The total volume prepared is calculated based on the subject's weight at screening. Treatment compliance will be listed and summarized by treatment. The numbers of subjects with compliance <80%, >120%, and >=90% will be calculated.

9.4 Operative Assessments

Type of surgical procedure and duration of CPB and aortic clamp will be listed and summarized for mITT population.

10 Efficacy Analysis

10.1 Primary Efficacy Analyses

Number and Percentage of subjects with any component of a major morbidity composite will be summarized by treatment and analyzed using Cochran Mantel Haenszel test adjusting for history of MI based on mITT population. Sensitivity analyses using PP population or mITT population minus subjects with postoperative AT administration per investigator's discretion will also be provided.

Subgroup analyses by age group, gender, and surgery type (isolated CABG vs. all other surgeries per medical review of all surgical procedures) will be performed for both mITT and PP populations, as well as mITT population minus subjects with postoperative AT administration.

10.2 Secondary Efficacy Analyses

All secondary efficacy analyses will be performed by treatment group based on the mITT population. No multiplicity adjustment will be performed.

All AT activity level data will be presented in listings. Postoperative AT levels at the ICU admission will be summarized. The analysis of covariance (ANCOVA) will be used to compare the treatment difference. ANCOVA model will include the AT level as the dependent variable, treatment as the fixed effect and baseline AT level (last available AT level from central laboratory before study drug administration) as the covariate. The same analyses will also be conducted for AT levels at other time points.

The number and percentage of subjects with AT levels of 58% or higher at the ICU admission will be summarized and compared with Chi-square test (or Fisher Exact test as appropriate).

Postoperative chest-drain blood loss in the first 12 and 24 hours after surgery will be listed, and will be summarized and analyzed using group t-test.

All data about transfusion requirements will be listed and summarized for the mITT population. Each blood product (e.g., RBCs, FFP, PLTs, cryoprecipitate, fibrinogen concentrate, PCCs, plasma, and whole blood) will be summarized separately and analyzed using group t-test. Cell saver blood use will also be analyzed. If the assumptions for parametric test are not met, the non-parametric test (Wilcoxon signed rank test) will be used. Shapiro-Wilk test will be used to test the normality.

Duration of ICU stay and the length of hospital stay will be listed and summarized. The non-parametric Wilcoxon rank sum test will be used to compare the treatment difference, and the Hodges-Lehmann estimate and 95% confidence interval (CI) will be provided.

Number and percentage of subjects with each following procedure or morbidity will be separately summarized and analyzed using Chi-square test (or Fisher Exact test as appropriate). Separate listings will be provided, except all-cause postoperative mortality which will be included in the listing of deaths. A separate eCRF page for Stroke was added while the study was underway, including questions of ‘Had a stroke?’, ‘stroke number’, ‘stroke date/time’, and ‘stroke type’. For subjects without the Stroke page, all AEs will be evaluated for stroke events by the medical review.

- Need for surgical reexploration
- Low cardiac output syndrome
- Myocardial infarction (MI)
- Stroke
- Acute kidney injury (AKI)
- Arterial or venous thromboembolic events
- Infections
- Prolonged mechanical ventilation (>24 hours)
- All-cause postoperative mortality
- Prolonged ICU stay (>6 days)

11 Safety Analysis

All safety analyses will be performed by treatment, based on the safety population.

11.1 Adverse Events

All reported AEs will be coded and summarized by system organ class (SOC) and preferred term (PT) according to Medical Dictionary for Regulatory Activities (MedDRA). The investigator verbatim term and SOC/PT will be shown simultaneously on the data listings of all AEs.

AEs will be classified as TEAEs or non-TEAEs depending on the comparison of AE onset date/time with the study treatment start date/time, and they will be summarized separately. A TEAE is an AE with an onset at or after the start of study treatment, otherwise it is a non-TEAE.

AE causality will be classified and assessed by the investigator. If the causality is definitive, probable, possible, or doubtful/unlikely, the event will be defined as a suspected adverse drug reaction (ADR). If the causal relationship is labeled as "Unrelated", it will be considered that the AE is not imputable to the investigational medicinal product and it is not a suspected ADR. In the framework of this study, a suspected ADR with a causal relationship of "definite" is named as adverse reaction (AR).

The incidence of non-TEAEs will be summarized. An overall summary of TEAEs, incidences of TEAEs, suspected ADRs, ARs, TEAEs with outcome of deaths, TEAEs leading to premature discontinuation from the study will be provided. The summary of treatment emergent bleeding events will be provided through the follow-up period (i.e., all treatment emergent bleeding events recorded), through 1 week post surgery (Day 7), and through 2 weeks post surgery (Day 14). Incidences of TEAEs will also be summarized by seriousness, severity, and causal-relationship to the study drug. Each subject is only counted once at each level of SOC and PT. For the summary by severity or by causality, each subject is only counted once per SOC or PT using the most severe or highest causality event. Summary tables will be sorted alphabetically by SOC and PT.

All AEs, deaths, SAEs, AEs leading to premature discontinuation from the study will be listed separately.

11.2 Clinical Laboratory Evaluations

All clinical laboratory test results will be listed.

The absolute values and change from baseline will be summarized using descriptive statistics by parameter and visit. Shift tables, based on the low/normal/high flags, will also be summarized by parameter and visit. Laboratory results out of the normal range judged by the investigators as clinically relevant will be reported, analyzed, and discussed as AEs.

11.3 Vital Signs

All vital sign data for systolic blood pressure, diastolic blood pressure, heart rate, temperature, and respiratory rate will be listed.

The absolute values and change from baseline will be summarized using descriptive statistics by visit.

Abnormal vital signs judged by the investigators as clinically significant will be reported, analyzed, and discussed as AEs.

11.4 Physical Examinations

All physical examination data will be listed by subject.

Findings at the Screening Visit will be summarized with numbers and percentages by body system. Entries for 'Other' body systems will be grouped together; a subject with 2 or more 'Other' entries will be counted only once. Physical exam change findings after the Screening Visit will be summarized with numbers and percentages for each response category to the question ‘Were there any new findings or existing findings that worsened?’ (‘No’, ‘Yes’).

Abnormal findings judged by the investigators as clinically significant will be reported, analyzed, and discussed as AEs.

12 Interim Analysis

No interim analysis is planned.

13 Changes in Planned Analysis

Modified ITT (mITT) is added as a new analysis population and will be used for efficacy analyses that were planned to be conducted on ITT. The reason of this change is that the number of subjects who were randomized but not administered study drug is more than expected and the efficacy variables are complications and sequelae of complex cardiac surgery. Only subjects who are dosed and operated on should be evaluated.

Per protocol (PP) population will be based on mITT, not just ITT as originally planned.

Safety population will be used for demographic and baseline summary analyses which were planned to be conducted on ITT population. The reason of this change is that the number of subjects who were randomized but not administered study drug is more than expected.