Statistical Analysis Plan

Sponsor:	Biotest AG
Protocol No:	Biotest 995
PPD Project Id:	PPD
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Title:	A randomized, active-controlled, multicenter, phase III study investigating efficacy and safety of intra-operative use of BT524 (human fibrinogen concentrate) in subjects undergoing major spinal or abdominal surgery (AdFIrst)
CRF Version No./Date:	3.0 / 15-Oct-2020
Protocol No./Date:	Final 4.0 / 04-Dec-2019 and Final 4.3 UK / 06-Sep-2021

1.0 Approvals

Sponsor	
Sponsor Name:	Biotest AG
Representative/ Title:	PPD
Signature /Date:	
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Project Manager/Title:	PPD
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Biostatistician / Title (Owner):	PPD
Signature /Date:	

(NOTE: Electronic Signatures should only be used if all parties have the ability to eSign.)

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Change History

Version	Change Log
1.0	Created as new
2.0	Added further rules for imputation of missing date and time Updated definition for relative day Updated definition for prior and concomitant medication Updated analysis for transfusion products and laboratory data
3.0	Updates which are editorial in nature and do not impact the analyses are not listed. Updated version of protocol and electronic Case Report Form (eCRF). Added new treatment with cryoprecipitate for subjects undergoing cytoreductive pseudomyxoma peritonei (PMP) surgery. Added interim analyses. Added visit Surgery Day 1 (Prior 1st dose). Updated inclusion criteria. Updated calculation of sample size. Updated randomization dependent on surgery (spine vs. cytoreductive PMP). Updated definitions of full analysis set and per-protocol set (PPS). Updated protocol deviation categories based on PD Guidance Template v3.0. Added TFL specification documents to Appendix 1
4.0	Added new UK protocol version.
5.0	Update to section 6.2 Sample Size Considerations. Clarification of total sample size taking randomized subjects who do not meet the PPS criteria into consideration. Update to section 8.12 Correction of Fibrinogen Level. Clarification of “15 minutes after start of IMP administration” added. Update to section 9.3 Full Analysis Set. Definition supplement for an unambiguous assignment of subjects to the analysis set. Update to section 9.4 Per-Protocol Set. Definition supplement for an unambiguous assignment of subjects to the analysis set.

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	Update to section 12.3.1, adding number of IMP administrations in grams. Update to safety section: 12.6.1 Adverse Events. Clarification and update to the list of AESIs and the respective definitions. Update to the AE table section, including analysis by type of surgery (spinal and abdominal surgery) 12.6.3 Laboratory Data. Analysis by type of surgery (spinal and abdominal surgery).
6.0	New exploratory efficacy endpoint “Overall Mortality” added to section 5.1 and section 7.2.1 Update to section 8.6. New subgroups added. These subgroups are described in more detail in section 12. Update to the definition of correction of fibrinogen level in section 8.12 Definition of overall mortality added to section 8.19 Update to section 9.3 describing the full analysis set to be in line with the ICH E9 guidelines. The previous full analysis set is now described in section 9.3.1. Tabulation of prohibited medication added to section 12.3.2 In section 12.5 new subgroups are added and the word ANCOVA is now replaced with ANOVA, Section 12.5.1.3 and 12.5.1.4 describe checking of ANOVA assumptions and alternative strategies in case the assumptions are significantly violated. Section 12.5.3.1 updated to include Pearson’s correlation between fibrinogen level and MCF Clauss assay. Plasma derived drugs removed from section 12.5.3.2 describing consumption of transfusion products after IMP until end of surgery. Summary of subjects with total avoidance of transfusion products post IMP administration added. Section 12.5.3.7. Method to produce the confidence internal added. Section 12.5.3.8 added to describe the analysis of overall mortality. Section 12.6.1 updated to accommodate new adverse event analyses and subgroup analyses. In section 12.6.3 a shift summary of hematology for number of subjects with change from normal or not clinical significant to clinical significant has been added.

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Abbreviations

AE	Adverse event
AESI(s)	Adverse event(s) of special interest
ANCOVA	Analysis of Covariance
ANOVA	Analysis of Variance
aPTT	Activated partial thromboplastin time
ATC	Anatomic Therapeutic Classification
AT III	Antithrombin III
BDRM	Blind Data Review Meeting
CMH	Cochran-Mantel-Haenszel
CSP	Clinical Study Protocol
DSMB	Data Safety Monitoring Board
ETP	Endogenous Thrombin Potential
eCRF	electronic Case Report Form
EU	European Union
FAS	Full Analysis Set
FFP	Fresh frozen plasma
FII	Factor II
FV	Factor V
FVII	Factor VII
FVIII	Factor VIII
FIX	Factor IX
FX	Factor X
FXI	Factor XI
FXIII	Factor XIII
F_{1+2}	Prothrombin Fragments 1+2
IMP	Investigational medicinal product
INR	International Normalized Ratio
IV	Intravenous
IWRS	Interactive Web Response System
PC	Protein C
PD	Protocol Deviation
PPS	Per-Protocol Set
PMP	Pseudomyxoma peritonei

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PS	Protein S
PT	Prothrombin Time
RBC	Red Blood Cells
SAF	Safety Set
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SD	Standard deviation
SMQ	Standard MedDRA Queries
SOC	System Organ Class
TAT	Thrombin-antithrombin III complex
TEAE	Treatment-emergent adverse event
TEE	Thromboembolic event
TFL	Tables, Figures and Listings
TGT	Thrombin Generation Test
TT	Thrombin Time
vWF	von Willebrand Factor
WBC	White Blood Cells
AE	Adverse event
AESI(s)	Adverse event(s) of special interest
ANCOVA	Analysis of Covariance
ANOVA	Analysis of Variance
aPTT	Activated partial thromboplastin time
AT III	Antithrombin III
ATC	Anatomic Therapeutic Classification
BDRM	Blind Data Review Meeting
CMH	Cochran-Mantel-Haenszel
CSP	Clinical Study Protocol
CTR	Clinical Trial Report
DSMB	Data Safety Monitoring Board
eCRF	electronic Case Report Form
ETP	Endogenous Thrombin Potential
EU	European Union
F_{1+2}	Prothrombin Fragments 1+2
FAS	Full Analysis Set
FFP	Fresh frozen plasma

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FII	Factor II
FIX	Factor IX
FV	Factor V
FVII	Factor VII
FVIII	Factor VIII
FX	Factor X
FXI	Factor XI
FXIII	Factor XIII
IMP	Investigational medicinal product
INR	International Normalized Ratio
IV	Intravenous
IWRS	Interactive Web Response System
mFAS	modified Full Analysis Set
NTEAE	Non-treatment emergent adverse event
PC	Protein C
PD	Protocol Deviation
PMP	Pseudomyxoma peritonei
PPS	Per-Protocol Set
PS	Protein S
PT	Prothrombin Time
RBC	Red Blood Cells
SAE	Serious Adverse Event
SAF	Safety Set
SAP	Statistical Analysis Plan
SD	Standard deviation
SMQ	Standard MedDRA Queries
SOC	System Organ Class
TAT	Thrombin-antithrombin III complex
TEAE	Treatment-emergent adverse event
TEE	Thromboembolic event
TFL	Tables, Figures and Listings
TGT	Thrombin Generation Test
TT	Thrombin Time
vWF	von Willebrand Factor
WBC	White Blood Cells

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2.0 Purpose

The statistical analysis plan (SAP) describes the statistical methods to be used during the reporting and analyses of data collected under Biotest AG Clinical Study Protocol 995.

3.0 Scope

This plan is a living document that will be created during the study start-up. The first version of the SAP will be drafted within three months of the final electronic Case Report Form (eCRF), and maintained throughout the lifecycle of the study. If any updates of the SAP are required before an interim analysis, then this version has to be approved and signed before locking any data for the interim analysis. The SAP will be finalized prior to database lock. The SAP will require sign off from the Project Manager, Biostatistician and representative of the sponsor.

The SAP outlines the following:

- Study objectives
- Study design
- Variables analyzed and analysis sets
- Applicable study definitions
- Statistical methods regarding major protocol deviations (PDs), study drug exposure, efficacy analysis, concomitant medications, adverse events (AEs) handling, laboratory data, vital signs and physical examinations
- Tables, Figures, Listings (TFLs) (defined in a separate document)

4.0 Introduction

This SAP describes the statistical methods to be used during the reporting and analyses of data collected under Biotest AG Clinical Study Protocol 995.

This SAP should be read in conjunction with the Clinical Study Protocol (CSP) and eCRF. This version of the plan has been developed using the CSP version 4.0, dated 04DEC2019 and version 4.2 UK, dated 04AUG2020 and eCRF version 3.0, dated 15OCT2020. Any further changes to the CSP or eCRF may necessitate updates to the SAP.

The purpose is to finalize a SAP so that programming can start. Versions of the SAP up to initial sponsor approval will be known as a draft SAP. If any updates of the SAP are required, for example due to changes of the CSP or eCRF, then this version has to be approved and signed before locking any data for the interim or final analysis.

4.1 Changes from Protocol

An additional mFAS population has been defined to follow ICH guidance.

The full analysis set (FAS) as defined in section 10.1 in the protocol “The FAS comprises all subjects who received at least one dose of IMP prior to the ‘end of surgery’ and have at least one post dose efficacy assessment“, has been altered in section 9.03 of this SAP version to “All randomized subjects receiving IMP post randomization and with data collected post randomization will be included in the FAS. Exclusion from the FAS can be considered in special cases as described in ICH E9, section 5.2.1.”. In addition, the previous FAS definition stemming from the protocol and defined in Section 9.0 of SAP version 5.0 is renamed to modified FAS (mFAS). These changes are done to be compatible with ICH E9 section 5.2.1.

Additional steps have been added for the efficacy section to allow for the potential of non-normal data. These are data transformations and a non-parametric Van Elteren’s method as specified in Section 12.5.1. This is an extension to Section 10.5 of the protocol.

Overall Mortality has been added to the SAP in Section 12.5.3.8 as a secondary efficacy endpoint.

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Per Section 10.5 of the protocol, there will be no imputation of missing values. Imputation has since been deemed necessary from SAP version 2.0. These rules are now defined in Section 8.4.

5.0 Study Objectives

The main purpose of this phase III study is to demonstrate the efficacy of BT524 as a complementary therapy to management of uncontrolled severe hemorrhage in acquired hypofibrinogenemia in subjects undergoing elective major spinal or abdominal surgery.

The primary objective of this study is to demonstrate that BT524 is non-inferior that means not worse than fresh frozen plasma (FFP)/cryoprecipitate with a non-inferiority margin of 150 mL in reducing intra-operative blood loss by intravenous (IV) administration in subjects with acquired hypofibrinogenemia undergoing elective major spinal or abdominal surgery.

If therapeutic equivalence (non-inferiority) has been demonstrated, therapeutic superiority of BT524 compared with FFP/cryoprecipitate will also be assessed.

Secondary objectives are to demonstrate the efficacy of BT524 by assessing the correction of the fibrinogen level intra-operatively, the transfusion requirements, post-operative blood loss in the first 24 hours, the number of subjects with rebleeds, the hospital length of stay and in-hospital mortality. Secondary objectives also comprise the safety of BT524 by documenting the number of AEs including changes in laboratory parameters, the viral status, and the frequency and severity of thrombosis and of thromboembolic events (TEEs).

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5.1 Endpoints

- Efficacy	***Primary Endpoint:*** • Intra-operative blood loss after decision to treat the subject with IMP until the end of surgery as measured by amount of blood from the blood suction unit and amount of blood from swabs, surgical cloths and compresses. ***Secondary Endpoints:*** • Proportion (%) of subjects with successful correction of fibrinogen level 15 minutes after start of first IMP administration • Time to first successful correction of fibrinogen level • Total amount of transfusion products (allogenic blood products) or autologous blood transfusion infused after start of first IMP administration until end of surgery • Amount of red blood cells (allogenic and autologous RBCs) infused after start of first IMP administration until end of surgery • Post-operative blood loss in the first 24 hours • Proportion (%) of subjects with rebleeds after the end of surgery until Day 8 • Hospital length of stay after surgery • In-hospital mortality ***Exploratory Endpoints:*** • Overall Mortality
- Safety	***Secondary Endpoints:*** • AEs • Changes in vital signs • Changes in clinical laboratory assessments of hematology, clinical chemistry, and urinalysis • Changes in clinical laboratory assessments of markers of coagulation • Changes in clinical laboratory assessments of coagulation factors • Frequency and severity of thrombosis and of TEEs • Viral status

6.0 Study Design

This is a phase III, prospective, randomized, active-controlled, multicenter, non-inferiority clinical study in adult subjects (≥ 18 years) undergoing major spinal or abdominal surgery to demonstrate the efficacy and the safety of intra-operative use of BT524 as a complementary therapy to management of uncontrolled severe hemorrhage in acquired hypofibrinogenemia.

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At least 200 subjects will be enrolled to ensure data are available for at least 100 evaluable subjects per treatment arm (BT524 or FFP/cryoprecipitate). The multicenter, multinational study will be conducted at approximately 15-20 sites in the EU and Switzerland with subjects undergoing major spinal surgery, and at one site in the United Kingdom with subjects undergoing cytoreductive pseudomyxoma peritonei (PMP) surgery.

This study will be partially blinded; surgeon, surgical staff and subjects will be blinded to treatment allocation throughout the entire surgery. The anesthesiologist who will administer the investigational medicinal products (IMP) could not be blinded to treatment allocation because of the inherent characteristics of the IMP BT524 and FFP/ cryoprecipitate.

Eligibility of subjects is assessed during screening with the exception of the inclusion criterion #5, which is assessed intra-operatively. The intraoperative inclusion criterion is different in subjects undergoing major spinal surgery (EU and Switzerland) and in subjects undergoing cytoreductive PMP surgery (United Kingdom).

Randomization of subjects to treatment will occur intra-operatively (predose) when eligibility for the clinical study has been confirmed. Subjects undergoing spine surgery will be randomized on a 1:1 basis to receive either BT524 or FFP, and subjects undergoing cytoreductive PMP surgery are to be randomized separately on a 1:1 basis to receive either BT524 or cryoprecipitate. For subjects undergoing spine surgery there will be a stratified randomization according to the predictive blood loss: > 1,000 mL to ≤ 2,000 mL and > 2,000 mL. The predictive blood loss will be recorded prior to surgery. Measurement of intra-operative blood loss will continue after decision to treat until end of surgery, and will serve as the primary efficacy parameter.

The study comprises a screening visit within 42 days prior to surgery to assess subjects eligibility, a baseline visit on the day of the surgery prior anesthesia (Day 1), the surgery phase (including randomization and single or repeated intra-operative administration of IMP, Day 1) and the follow-up phase of at least 5 weeks with 4 follow-up visits on Days 2, 3, 5 and 8 and the closing visit, including the final safety examination, scheduled on Day 36* after the day of surgery (*+35, up to Day 71 if required). The duration of individual study participation for eligible screened subjects is at least 5 weeks.

Subjects are entitled to discontinue participation in the clinical study at their own request at any time without stating a reason.

The investigator can terminate a subject's participation in the study at any time if continuation could lead to disadvantages for the subject, which cannot be justified by the investigator. Withdrawn subjects will not be replaced.

After 40 subjects have completed the study, a data monitoring of blinded aggregate data of all 40 subjects without separating the subjects according to treatment will be performed to recalculate the sample size. See section 6.2.1 for details.

In this study, three interim analyses with an alpha-adjustment according to Haybittle/Peto (Haybittle, 1971; Peto et al., 1976; Schulz and Grimes, 2005) are planned for assessment of adjusting the sample size. See section 10.0 for details.

A Data Safety Monitoring Board (DSMB) will independently review and assess the unblinded safety data throughout the entire study at regular intervals. The DSMB members can propose to stop the study at any time after a scheduled or unscheduled meeting in case of major safety concerns related to study treatment.

Further details on the assessment schedule (including the follow-up period) that will be used for assessment of the efficacy and safety parameters in this study are presented in the following flowchart of study.

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Study Schedule / Assessments	Day / Visit	D-42 to D-1	D-2 to D1	D1	D1	D1	D1	D2	D3	D5	D8	D36 (+35)
		Screening	Prior surgery (Baseline)	Surgery: prior 1st dose	Surgery: pre-dose	Surgery: post-dose	Surgery: end of surgery	Follow-up	Follow-up	Follow-up	Follow-up	Closing
Informed Consent		•										
Check/re-check of inclusion / exclusion criteria		•	•[1]									
Demographic data		•										
Classification of type of surgery		•										
Recording expected blood loss			•									
Body weight		•	•[2]									
Physical examination		•	•[2]					•	•	•	•	•
Pregnancy test, only in females of childbearing potential		•	•[2]									
Medical and surgical history		•	•[3]									
Viral safety: Collection of retention sample		•										•
Virus serology (hepatitis B, hepatitis C, HIV)		•										•
Vital signs		•	•	•	•	•	•	•	•	•	•	•
Hematology and clinical chemistry		•	•[2]	•			•	•	•	•	•	•
Urinalysis		•	•[2]					•	•		•	•
Markers of coagulation (coagulation activation tests)		•	•[2]	•		•[4]	•[5]	•	•	•	•	•
Plasma concentration of fibrinogen activity (Clauss assay)		•	•[2]	•		•[4]	•[5]	•				
FIBTEM A10 (ROTEM)		•	•[2]	•	•	•[4]	•[5]	•				

[1] Diagnostic tests will be repeated at the investigator's discretion.
[2] Diagnostic tests have to be done prior to surgery. In case of short time-period between screening and baseline (≤ 2 days) these tests have only to be repeated based on medical judgment of the investigator. If not repeated, screening results will serve as baseline.
[3] Previous medication: change from screening.
[4] Tests have to be done only 15 and 90 minutes after start of 1st IMP administration.
[5] Tests for *markers of coagulation and plasma activity of fibrinogen (Clauss assay, FIBTEM A10, MCF)* have to be done '90 min after start of 1st IMP administration' and at the 'end of surgery'. In case of a short time-period between these two time-points (<30 min) these tests have only to be repeated based on medical judgment of the investigator.

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Study Schedule **Day**	**D-42 to D-1**	**D-2 to D1**	**D1**	**D1**	**D1**	**D1**	**D2**	**D3**	**D5**	**D8**	**D36** (+35)
Assessments **Visit**	**Screening**	**Prior surgery** (Baseline)	**Surgery**				**Follow-up**	**Follow-up**	**Follow-up**	**Follow-up**	**Closing**
			prior 1st dose	**pre-dose**	**post-dose**	**end of surgery**					
Maximum clot firmness (MCF) (ROTEM)	•	•[2]	•	•	•[4]	•[5]	•				
Coagulation factors (including vWF)		•	•		•[6]						
Intra-operative inclusion criteria			•								
Intravenous infusion(s) of IMP (BT524 or FFP/cryoprecipitate)			•[7]								
Recording start/end of surgery			•			•					
Continuous measurement of blood loss from start of surgery			•			•	•				
Calculation and recording of blood loss						•[8]	•[9]				
Recording time of decision to treat the subject with IMP			•								
Order of IMP			•	•							
Randomization			•								
Rebleeding episodes						•	•	•	•	•	
Concomitant medication or treatment			•			•	•	•	•	•	•
Transfusion products			•			•	•	•	•	•	•
Adverse events	•	•	•			•	•	•	•	•	•

[6] Test has to be done only 90 minutes after start of 1st IMP administration.
[7] Total volume and total infusion time (start and end of each infusion) to be recorded.
[8] Intra-operative blood loss from time-point of decision to treat the patient with IMP until end of surgery.
[9] Recording of blood loss (drainage volume) until 24 hours after end of surgery.

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6.1 Inclusion/Exclusion Criteria

Only subjects meeting all of the following **inclusion criteria** will be considered for study inclusion (will be considered for randomization to study treatment):

1. Written informed consent obtained from subjects indicating that they understand the purpose of and procedures required for the study and are willing to participate in it
2. Subjects scheduled for elective major spinal or cytoreductive PMP surgery with expected major blood loss
3. Male or female, aged ≥ 18
4. No increased bleeding risk as assessed by standard coagulation tests and medical history
5. Intra-operative trigger for treatment
    a. Subjects undergoing spinal surgery: Intra-operative clinically relevant bleeding of approximately 1 L, requiring hemostatic treatment during surgery.
    b. Subjects undergoing cytoreductive PMP surgery: Intra-operative prediction of clinically relevant bleeding of > 2 L (approximately 60 minutes after the start of cytoreductive PMP surgery), requiring hemostatic treatment during surgery.

Subjects having any of the following **exclusion criteria**, either at screening and/or at baseline will not be included in the study (will not be randomized to study treatment):

1. Pregnancy or unreliable contraceptive measures or breast feeding (women only)
2. Hypersensitivity to proteins of human origin or known hypersensitivity reactions to components of the IMP
3. Participation in another clinical study within 30 days before entering the study or during the study and/or previous participation in this study
4. Treatment with any fibrinogen concentrate and/or fibrinogen-containing product within 30 days prior to infusion of BT524
5. Employee or direct relative of an employee of the Contract Research Organization, the study site, or Biotest
6. Inability or lacking motivation to participate in the study
7. Medical condition, laboratory finding (e.g., clinically relevant biochemical or hematological findings outside the normal range), or physical exam finding that in the opinion of the investigator precludes participation
8. Presence or history of venous/arterial thrombosis or TEE in the preceding 6 months

6.2 Sample Size Considerations

The non-inferiority margin is defined as 150 mL blood loss, as such difference in blood loss after the decision to treat the subjects with IMP is considered as clinically not relevant.

It is assumed that BT524 is non-inferior that means not worse than FFP/cryoprecipitate with a non-inferiority margin of 150 mL in reducing intra-operative blood loss. Assuming a blood loss of about 500 mL in the FFP/cryoprecipitate treatment arm after the decision to treat the subject with IMP until end of surgery, a standard deviation (SD) of 375 mL, a non-inferiority margin of 150 mL, an alpha-level of 2.5% (1-sided) 100 evaluable subjects per treatment arm are needed to demonstrate the non-inferiority of BT524 by using a t-test (equivalence) with 80% power.

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Based on the results of the blinded sample size recalculation (see section 6.2.1) the power was reduced from 90% to 80% to allow an increase of the originally assumed SD of 325 mL to 375 mL.

The sample size will be recalculated at the interim analyses as defined in section 10.0. Sample size estimations will be performed using nQuery Advisor Version 4.0 or higher.

With 100 subjects per treatment arm superiority of BT524 can also be tested with a power of > 80% (t-test, alpha=0.05 2-sided, effect size Δ=0.5).

It is expected that approximately 10% of the randomized subjects will not fulfil the criteria to be included in the PPS. Therefore, approx. 220 randomized subjects are required to reach 200 evaluable subjects in the PPS.

6.2.1 Sample Size Recalculation

After 40 subjects had completed the study, the overall mean and SD for the primary efficacy variable intra-surgery blood loss after decision to treat the subject with IMP was derived using blinded aggregate data of all 40 subjects without separating according to treatment.

If the assumed mean and SD blood loss are not reflected in these subjects, then a sample size adjustment had to be considered to ensure that a sufficient number of subjects will be randomized to maintain a power of 90%.

If an adaption of the sample size is intended, this had to be documented in a protocol amendment.

The sample size recalculation was performed. The CSP (version 4.0) and the sample size considerations were updated (see section 6.2). The power was reduced to 80%.

6.3 Data Monitoring

This data monitoring is not an interim analysis because the analysis is performed with all subjects without separating the subjects according to treatment. Therefore, no alpha-adjustment is necessary.

6.4 Randomization

There will be a stratified randomization per surgery type.

Subjects undergoing spine surgery are to be randomized on a 1:1 basis to receive either BT524 or FFP, and subjects undergoing cytoreductive PMP surgery are to be randomized separately on a 1:1 basis to receive either BT524 or cryoprecipitate.

For subjects undergoing spine surgery the randomization will be stratified according to the predictive blood loss: > 1,000 mL to ≤ 2,000 mL and > 2,000 mL. The predictive blood loss will be recorded prior surgery.

Randomization of subjects to treatment will occur intra-operatively (predose) when eligibility for the clinical study has been confirmed.

<u>For subjects undergoing spine surgery the following applies:</u>

After an intra-operative blood loss of approximately 1 L, requiring hemostatic treatment (high risk for the need of fibrinogen supplementation with BT524 or FFP) during surgery, the randomization request will be sent to the pharmacy (if applicable). The pharmacy retrieves the randomization code via Interactive Web Response System (IWRS) and provides the prepared IMP to the unblinded anesthesiologist. In case the anesthesiologist retrieves the randomization code via IWRS, the pharmacy will be informed accordingly and can provide the IMP.

<u>For subjects undergoing cytoreductive PMP surgery the following applies:</u>

Randomization of PMP subjects to treatment will occur intra-operatively (pre-dose) when eligibility for the clinical study has been confirmed. Immediately after the prediction of clinically relevant bleeding > 2 L, requiring hemostatic treatment (high risk for the need of fibrinogen supplementation with BT524 or cryoprecipitate) during surgery, the person responsible for randomization will be informed and the randomization request will be sent. The person responsible for randomization retrieves the

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randomization code via IWRS and informs the blood bank/pharmacy. BT524 will be delivered to the operating room by the pharmacy. After randomization, BT524 will be prepared in the operating room in accordance with the manufacturer's instructions and administered immediately after reconstitution by the unblinded anesthesiologist.

After randomization, cryoprecipitate will be thawed and prepared for administration by the local blood bank, delivered to the operating room, and administered upon arrival.

In case the anesthesiologist retrieves the randomization code via IWRS, the blood bank/ pharmacy will be informed accordingly and can provide the IMP.

Subject Identification

For the coherent assignment of the study, documents all subjects having signed the informed consent and having entered the screening period will receive a subject number. The subject number comprises a five-digit number of which the first two digits define the investigational site and the last three digits the subject enrolled at the corresponding site. Subject numbers are assigned consecutively per site. Subject numbers are assigned unique and will not be replaced i.e., in case of a screening failure.

An interactive web/voice response system will be implemented and used for randomization and re-supply. Detailed instructions for the use of IWRS systems are provided in a separate document that will be filed in the Investigator Site File.

The random allocation of treatments to subjects will be done using a computerized randomization program. Subjects will receive a randomization number, which will be recorded along with the date of randomization in the eCRF.

7.0 Study Parameters and Covariates

7.1 Primary Efficacy Parameter

The primary efficacy parameter is determined from the intra-operative blood loss after the decision to treat the subject with IMP until the end of the surgery as measured by the amount of blood from the blood suction unit and the amount of blood from swabs, surgical cloths and compresses. The end of surgery is defined as time of last suture.

7.2 Secondary Parameters

7.2.1 Efficacy

Secondary efficacy will be determined using the following parameters:

- Proportion (%) of subjects with successful correction of fibrinogen level (FIBTEM A10) 15 minutes after start of first IMP administration.
  Successful correction of fibrinogen level in a subject is defined as restoring fibrinogen FIBTEM A10 baseline level to at least 95% measured by ROTEM 15 minutes after start of first IMP administration. A correction of at least 95% is considered successful, as measurement methods has a coefficient of variation of about 5% (Solomon et al., 2015).
- Time to first successful correction of fibrinogen level (15 minutes or 90 minutes after start of first IMP administration, end of surgery, not within surgery).
- Total amount (volume in mL and number of units [bags]) of transfusion products (allogeneic blood products) or autologous blood transfusion infused after start of first IMP administration until end of surgery. The end of surgery is defined as time of last suture.
- Amount (volume in mL and number of units [bags]) of red blood cells (RBCs) (allogeneic and autologous) infused after start of first IMP administration until end of surgery. The end of surgery is defined as time of last suture.

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- Post-operative blood loss in the first 24 hours.
- Proportion (%) of subjects with rebleeds after the end of surgery until Day 8. Rebleeds are defined as any bleed requiring hemostatic treatment (including reoperation) after the end of surgery until Day 8.
- Hospital length of stay after surgery, defined as the date of discharge minus the date of surgery.
- In-hospital mortality.

7.2.2 Exploratory Efficacy

Exploratory efficacy endpoints:

- Overall Mortality

7.2.3 Safety

Safety and tolerability in this clinical study will be addressed by the following safety parameters:

- Frequency, severity, seriousness and causality of AEs.
- Changes in vital signs (including pulse [heart rate], blood pressure, respiratory rate, body temperature).
- Change in clinical laboratory assessments of hematology, clinical chemistry and urinalysis.
- Change in clinical laboratory assessments of markers of coagulation:

  Prothrombin time (PT)/International Normalized Ratio (INR), activated partial thromboplastin time (aPTT), thrombin-antithrombin III complex (TAT), prothrombin fragments 1+2 (F1+2), D-dimer, protein S (PS), protein C (PC), antithrombin III (AT III), thrombin time (TT) and Endogenous Thrombin Potential (ETP) measured by Thrombin Generation Test (TGT) (TGT only for subjects undergoing cytoreductive PMP surgery).

- Change in clinical laboratory assessments of coagulation factors:

  Factor II (FII), factor V (FV), factor VII (FVII), factor VIII (FVIII), factor IX (FIX), factor X (FX), factor XI (FXI), factor XIII (FXIII) and von Willebrand factor (vWF) (vWF only for subjects undergoing cytoreductive PMP surgery).

- Frequency, severity, seriousness and causality of thromboses and TEEs, of relevant bleeding complications, of Hypersensitivity/ anaphylactic reactions, and of bleeding related ischaemic events.
- Change in viral status.

7.3 Predetermined Covariates and Prognostic Factors

In addition to treatment as a prognostic factor, there is a predetermined covariate for subjects undergoing spinal surgery only. The stratification factor predicted blood loss (> 1,000 mL to ≤ 2,000 mL and > 2,000 mL) is a covariate for the analysis of the primary efficacy variable and two secondary efficacy variables: amount of RBCs infused after IMP, post-operative blood loss in the first 24 hours.

Predicted blood loss will also be used as stratification factor in the analyses of two secondary efficacy variables: proportion of subjects with successful correction of fibrinogen level 15 minutes after start of first IMP administration, proportion of subjects with rebleeds after the end of surgery until Day 8.

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8.0 Definitions

8.1 Age

Age will be calculated as:

Age = (date of informed consent – date of birth) / 365.25 rounded to lowest integer

Only year of birth will be recorded. The date will be imputed as 01 January. The imputed date of birth will be used for age calculation.

8.2 Baseline

If not stated otherwise, the last non-missing valid observation prior to surgery will serve as the baseline measurement.

8.3 Change from Baseline

Change from baseline will be calculated by statistical programming as follows:

Change from baseline = Post-baseline measurement – baseline measurement

8.4 Missing Data Conventions

In this short study, not many missing values are expected. Therefore, in general, data will not be imputed.

In case there will be at least 10% missing data, these data will be checked at the Blind Data Review Meeting (BDRM) and imputation methods will be considered. It is assumed that these missing data will be at random and an imputation method which is based on the worst case of the respective treatment group will be used as a sensitivity analysis.

For binary endpoints, an observed case analysis (excluding missing data) will be considered to be the primary analysis method and a non-responder analysis (treating missing values as non-responders or the worst case) may be performed as a sensitivity analysis if deemed necessary.

Imputed values will be flagged in listings.

8.4.1 Missing Severity Assessment for Adverse Events

If severity is missing for an AE starting prior to the administration of IMP, then a severity of “Mild” will be assigned. If the severity is missing for an AE starting on or after administration of IMP, then a severity of “Severe” will be assigned. The imputed values for severity assessment will be used for incidence summaries, while the actual values will be used in data listings.

8.4.2 Missing Relationship to Investigation Product for Adverse Events

If the relationship to IMP is missing for an AE starting on or after the administration of IMP, a causality of “Related” will be assigned. The imputed values for relationship to IMP will be used for incidence summaries, while the actual values will be presented in data listings.

8.4.3 Missing Seriousness Assessment for Adverse Events

If no information about seriousness is available, the AE will be considered serious. The imputed values for seriousness assessment will be used for incidence summaries, while the actual values will be used in data listings.

8.4.4 Missing/under detection limit Laboratory values

Laboratory values that are "not measurable" or "under detection limit" should not be replaced and should not be included in the summary tables. Quantitative laboratory parameters reported as “< x” or “> y” will be imputed with x/2 and y, respectively, for inclusion in summary statistics. The reported value will be presented in listings.

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8.5 Handling of partial dates and missing/incomplete times

For prior or concomitant medications, incomplete (i.e., partially missing) start date and/or stop date will be imputed. When the start date and the stop date are both incomplete for a subject, impute the start date first.

For AEs, incomplete (i.e., partially missing) start dates and times and/or incomplete stop dates and times will be imputed.

For transfusion products, incomplete start and/or stop times will be imputed.

Imputation rules for incomplete dates for prior or concomitant medications, incomplete dates and times for AEs, and incomplete times for transfusion products are described in sections 8.5.1 and 8.5.2.

Imputed dates will be used for all further derivations (e.g., treatment-emergent adverse event [TEAE] definition, classification of medications as prior or concomitant, and study day calculation). Imputed dates will also be used and flagged in listings.

Imputed dates will be indicated in the listings by using ‘D’ if only day is imputed, and ‘M’ if day and month are imputed, Imputed times will be indicated in listings by using ‘m’ if only minutes are imputed and ‘h’ if minutes and hours are imputed. It is not expected that dates for any other parameter are missing. Should any other dates be missing or partially missing, then statistical programming will place a warning in the log file and inform the statistical lead should these dates be required for derivations.

8.5.1 Incomplete Start Date and Time

The following rules will be applied to impute the missing numerical fields. If the stop date is complete and the imputed start date is after the stop date, then the start date will be imputed using the stop date.

Missing Day and Month

- If the year of the incomplete start date is the same as the year of the date of the first dose of IMP, then the day and month of the date of the first dose of IMP will be assigned to the missing fields.
- If the year of the incomplete start date is before the year of the date of the first dose of IMP, then December 31 will be assigned to the missing fields.
- If the year of the incomplete start date is after the year of the date of the first dose of IMP, then 01 January will be assigned to the missing fields.
- If the subject was not treated (no IMP start date), then 01 January will be assigned to the missing fields.

Missing Month Only

- The day will be treated as missing and both month and day will be replaced according to the above procedure.

Missing Day Only

- If the month and year of the incomplete start date are the same as the month and year of the date of the first dose of IMP, then the day of the date of the first dose of IMP will be assigned to the missing day.
- If either the year is before the year of the date of the first dose of IMP or if both years are the same but the month is before the month of the date of the first dose of IMP, then the last day of the month will be assigned to the missing day.
- If either the year is after the year of the date of the first dose of IMP or if both years are the same but the month is after the month of the date of the first dose of IMP, then the first day of the month will be assigned to the missing day.
- If the subject was not treated (no IMP start date), then the first of the month will be assigned to the missing field.

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Missing/Incomplete Time

- If the time is missing and the date is complete and is the same as the date of the first dose of IMP, or the date is imputed to be this date, then the time will be set to the time of the first dose of IMP. Otherwise, missing times will be imputed as 00:00.
- If the minutes are given but the hour is not, then the time will be regarded as completely missing and handled as above.
- If the hour is given but the minutes are not, then if the hour is the same as the hour of the first dose of IMP, then the minutes of that dose of IMP will be assigned to the missing fields. Otherwise 00 will be assigned to the missing minutes.
- If the subject was not treated (no IMP start date), then missing times will be imputed as 00:00.

8.5.2 Incomplete Stop Date and Time

The following rules will be applied to impute the missing numerical fields. If the imputed stop date is before the start date (imputed or non-imputed start date), then the imputed stop date will be equal to the start date.

Missing Day and Month

- If the year of the incomplete stop date is the same as the year of the date of the study discontinuation/completion, the stop day and month will be set to the maximum of the date of study discontinuation/completion, as appropriate, or the date equivalent to 35 days after the last dose of IMP.
- If the year of the incomplete stop date is before the year of the date of study discontinuation/ completion, as appropriate, or the date equivalent to 35 days after the last dose of IMP, the day and month will be set to 31 December.
- If the year of the incomplete stop date is after the year of the date of study discontinuation/ completion, as appropriate, or the date equivalent to 35 days after the last dose of IMP, the day and month will be set to 01 January.

Missing Month Only

- The stop day will be treated as missing and both month and day will be replaced according to the above procedure.

Missing Day Only

- The stop day will be set to the last day of the month.

Totally Missing Stop Date

- The concomitant medication will be considered ongoing.

Missing /Incomplete Time

- If the time is missing, it will be imputed as 23:59.
- If the minutes are given, but not the hour, then the time will be regarded as completely missing and handled as above.
- If hours are given, but minutes are not, minutes will be imputed with 59.

8.6 Subgroups

The subgroup type of surgery (spinal and abdominal) is analyzed for the primary as well as for all secondary endpoints. Subgroups sex and race are analyzed for the primary endpoint.

Further subgroup analysis according to the predictive blood loss is planned for two secondary efficacy variables: total amount of transfusion products after IMP administration until end of surgery, total amount of RBC after IMP administration until end of surgery.

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For specific adverse events and other safety parameters, subgroup analyses are being conducted based on type of surgery, sex and race. For more information, please refer to section 12.6.1.

8.7 Pooling of Centers

In case of low number of subjects per center, summaries of data by center would be unlikely to be informative. Therefore, data from all centers per country/region (if applicable) and in total will be pooled prior to analysis. Only enrollment will be summarized by country and center.

8.8 Completed Study

A subject is considered to have completed the study when he/she is presumed to have followed the CSP (i.e., completed visits approximately 5 weeks after surgery).

8.9 End of Surgery

End of surgery is defined as time of last suture.

8.10 Study Visits

The labels of study visits in this study are as following:

- Screening
- Baseline
- Surgery Day 1 (Prior 1st dose)
- Surgery Day 1 (Pre-dose)*
- Surgery Day 1 (15 min after IMP start)
- Surgery Day 1 (90 min after IMP start)
- Surgery Day 1 (End of Surgery)
- Day 2, Follow-up
- Day 3, Follow-up
- Day 5, Follow-up
- Day 8, Follow-up
- Closing Visit
- Early Discontinuation

*only applicable in case of repeated intra-operative IMP administration

For a subject who did not complete the study, date of early discontinuation is the last visit, when the subject discontinued from the study. As subjects are expected to attend a Closing Visit whether they complete or discontinue early from the study, subjects who complete the study will have their Closing Visit data summarized under 'Closing Visit', while subjects who discontinue early from the study will have their Closing Visit data summarized under 'Early Discontinuation'. If a subject terminates the study prematurely without having a closing visit, the data from the last regular visit will be summarized under this visit.

8.11 Amount of Blood Loss

During surgery, the blood loss will be quantified by measuring the continuous bleeding mass with a blood suction unit (and/or a cell saver) and by calculation of the amount of blood from swabs, surgical cloths and compresses. For the primary endpoint, the amount of blood will be measured for the time between decision to treat and end of surgery.

Calculation of blood loss:

Blood loss (mL) = Volume of blood on swabs, surgical cloths and compresses (mL) + Volume of blood in the suction container (mL) + Total volume of blood loss from other sources (mL)

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Volume of blood on swabs, surgical cloths and compresses (mL) = Weight of used/unused surgical cloths and compresses (g) - Weight of dry swabs, surgical cloths and compresses (g). 1 g weight represents 1mL volume of blood.

Volume of blood in the suction container (mL) = Total volume of fluid collected in suction container (mL) - Total volume of anticoagulant solution in blood suction container (mL)* - Total volume of irrigation solution (mL)

* Not applicable for abdominal surgery

Total volume of irrigation solution = Capacity of irrigation syringe (mL) * Number of times irrigation syringe used

Handling of missing data:

For the calculation of **Blood loss**, Volume of blood on swabs, surgical cloths and compresses and Volume of blood removed from the surgical field by the blood suction is required. If there is at least one missing value (see above for single items needed) the blood loss cannot be calculated, and the volume will be missing.

For the calculation of blood in the suction container: Total volume of anticoagulant solution in blood suction container is not applicable for abdominal surgery and will not be considered for the calculation.

For the calculation of Total volume of irrigation solution: If Capacity of irrigation syringe is missing and Number of times irrigation syringe used > 0 then volume is missing. If Capacity of irrigation syringe is provided and Number of times irrigation syringe used is missing, then volume is missing. If Capacity of irrigation syringe is missing and Number of times irrigation syringe used = 0 then volume is 0.

8.12 Correction of Fibrinogen Level

The fibrinogen level is assessed by FIBTEM A10 test, MCF and Clauss assay at the following visits: screening, prior surgery (baseline), Surgery Day 1 (Prior 1st dose), Surgery Day 1 (Predose)*, Surgery Day 1 (15 minutes after start of first IMP administration), Surgery Day 1 (90 minutes after start of first IMP administration), Surgery Day 1 (End of surgery), and Day 2, Follow-up Visit. (* only applicable in case of repeated intra-operative IMP administration).

Rate of fibrinogen restoring = fibrinogen level at a post-baseline visit / fibrinogen level at baseline*100

If the rate of fibrinogen restoring is ≥ 95%, then the fibrinogen level is considered to be successfully corrected at that post-baseline visit for the subject.

The above rate will be calculated for the following visits: Surgery Day 1 (15 minutes after start of first IMP administration), Surgery Day 1 (90 minutes after start of first IMP administration), Surgery Day 1 (End of surgery), and Day 2, Follow-up.

The time to first successful correction of fibrinogen level is classified into the following categories: "Within 15 minutes after IMP start", "Greater than 15 and less than or equal to 90 minutes after IMP start", "Greater than 90 minutes after IMP start" and "Unsuccessful correction".

At every visit, the time point relative to the start time of IMP is calculated in minutes as Collection Time of sample minus Start Time of IMP. Any corrections <15 minutes after IMP start are not considered for classification.

The earliest time point, when the rate is ≥ 95%, is the time to first successful correction of fibrinogen level and will be used to categorize the subjects into the respective categories.

Subjects with time to first successful fibrinogen correction on Follow-up Visit Day 2 will be assigned to the category "Greater than 90 minutes after IMP start". If the rate of fibrinogen restoring is < 95% at all the time points, correction of fibrinogen level is reported as "Unsuccessful correction".

A sensitivity analysis adds a time window of ±3 minutes to the category "Within 15 minutes after IMP start", so that time points ≥12 and ≤18 minutes fall into this category. To the category "Greater than 15 and less

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than or equal to 90 minutes after IMP start" a time window of +3 minutes is added, so that time points between >18 and ≤93 minutes fall into this category. Corrections >93 minutes are categorized as "Greater than 90 minutes after IMP start".

In addition, and independent from the above categories, the time to first successful correction is classified into "Until end of surgery" or "Not within surgery". Subjects with time to first successful correction at the time point Surgery Day 1 (End of surgery) are assigned to "Until end of surgery". Subjects with time to first successful correction on Follow-up Visit Day 2 are assigned to "Not within surgery". For the visits Surgery Day 1 (15 minutes after start of first IMP administration) and Surgery Day 1 (90 minutes after start of first IMP administration) the time point is compared with the End time of Surgery and subjects are assigned to "Until end of surgery" or "Not within surgery" accordingly. If the rate of fibrinogen restoring is < 95% at all the time points, correction of fibrinogen level is reported as "Unsuccessful correction".

The whole analysis for the correction of fibrinogen levels is also done based on results obtained from MCF and Clauss assay to perform sensitivity analyses.

8.13 Amount of Transfusion Products

All types of transfusion products administered, including RBCs will be counted. The volumes and units (bags) of all these products will be summarized by type of transfusion product, and by treatment arm.

8.14 Amount of Red Blood Cells

The volumes and units (bags) of allogeneic RBCs and autologous blood transfusion/cell salvage will be counted separately.

8.15 Post-operative Blood Loss in the First 24 hours

The plasma drainage tube and disposable drainage bag will be emptied 24 hours after the end of surgery. The post-operative drainage volume in the first 24 hours is collected in eCRF.

8.16 Rebleeds after the End of Surgery until Day 8

Rebleeds are defined as any bleed requiring hemostatic treatment (including reoperation) after the end of surgery until Day 8, which are recorded in eCRF.

8.17 Hospital Length of Stay after Surgery

Length of stay after surgery (days) = date of discharge – date of surgery

Where date of discharge is the date of discharge following the IMP treated surgery.

8.18 In-hospital Mortality

In-hospital mortality is defined as death occurring during the hospital stay.

8.19 Overall Mortality

Overall mortality is defined as death occurring during study duration.

8.20 Relative Day

If the assessment date is prior to the date of IMP administration, the relative day is date of assessment minus date of IMP administration. If the assessment date is on the date of IMP administration or after that, then the relative day is the date of assessment minus date of IMP administration plus 1.

8.21 Time to Start of First Investigational Medicinal Product Infusion

Time to start of first IMP infusion = Start time of first IMP administration - Time of decision to treat subject with IMP.

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8.22 Total planned Investigational Medicinal Product Volume

Total planned IMP volume = Sum of all planned IMP volume.

8.23 Total actual Investigational Medicinal Product Administered

Total actual IMP administered = Sum of all actual IMP administered.

8.24 Duration of Investigational Medicinal Product Infusion

Duration of XXX IMP administration = End time of XXX IMP administration - Start time of XXX IMP administration

Where XXX is the first, second … administration.

8.25 Relative Time in Surgery

Duration of Surgery = End time of surgery - Start time of surgery

Time until decision to treat = Time of decision to treat subject with IMP - Start time of surgery

Time to end of surgery after decision to treat = End time of surgery - Time of decision to treat subject with IMP

Time of IMP exposure during surgery = End time of surgery - Time of start of first IMP administration

9.0 **Analysis Sets**

9.1 All Subjects Enrolled Set

The All Subjects Enrolled Set includes all subjects who have given informed consent to this study.

9.2 Safety Analysis Set

The Safety Analysis Set (SAF) comprises all subjects who have received at least one dose of IMP. Subjects will be analyzed according to the treatment received.

9.3 Full Analysis Set

All randomized subjects receiving IMP post randomization and with data collected post randomization will be included in the FAS. Exclusion from the FAS can be considered in special cases as described in ICH E9, section 5.2.1. For the purpose of this trial, there are two FAS: FAS and mFAS. All subjects will by default be included and analyzed according to the randomized treatment allocation in both definitions.

The decision to exclude subjects from FAS will be done at a data review meeting after reviewing the data as described in the Data Review Meeting Plan.

9.3.1 Modified Full Analysis Set

As per the section above, all randomized subjects will be included in the modified full analysis set (mFAS) who meet the following conditions.

1. Subjects who received at least one dose of IMP prior to the ‘end of surgery’ and
2. have at least one post-dose efficacy assessment.
3. This includes all subjects whose IMP infusion started prior to the end of surgery, irrespective of the amount of IMP infused.

Subjects will be analyzed as randomized. The decision to exclude subjects from mFAS will be done at a data review meeting after reviewing the data as described in the Data Review Meeting Plan.

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9.4 Per-Protocol Set

The Per-Protocol Set (PPS) is a subset of FAS and includes all subjects who are compliant with the CSP without any major PDs thought to have the potential to impact the results of the efficacy analysis, e.g., no treatment with IMP, incomplete treatment with IMP (administration of the 1st IMP dose is not completed if the end of the 1st IMP administration is after the end of surgery), treatment with IMP after the 'end of surgery', no post-dose efficacy assessment for the primary endpoint. Classification of PDs as major or minor will be agreed upon at the BDRM prior to database lock and also prior to each Interim Analysis for eligible subjects.

Subjects will be analyzed according to the treatment received.

10.0 Interim Analyses and Data Monitoring

To ensure subject safety, the safety data will be evaluated by a DSMB at regular intervals. A DSMB charter and list of required tables and listings is available (see Appendix 1).

After 40 subjects have completed the study, a blinded sample size recalculation will be performed. See section 6.2.1 for details. A specification of required table and listing is available (see Appendix 1).

In this study, three interim analyses of the observed blood losses are planned to have the option of adjusting the sample size needed.

For this purpose, an alpha-adjustment according to Haybittle/Peto (Haybittle, 1971; Peto et al., 1976; Schulz and Grimes, 2005) is planned. This leads to local alpha levels of 0.001 for each interim analysis and a significance level of 0.05 for the final analysis to reach a global significance level of 5%.

Past interim analyses were based on the PPS, and past sensitivity analyses were based on the mFAS.

The first interim analysis is planned with approximately 50 spine subjects, the second one with at least 40 cytoreductive PMP subjects and all other evaluable spine subjects at that time point. The third interim analysis is planned after approximately 80% of subjects of the total sample size.

Aim of all interim analyses is to adapt the sample size according to the observed blood losses and the SDs:

a) Early termination due to non-inferiority of BT524 in comparison with the used standard therapies.
b) Continuation with the sample size as initially planned.
c) Adjustment of sample size to take into account changes from the previous assumptions on the additional blood loss.
d) Stopping the study early due to futility if the sample size re-estimation indicates a much higher number than planned before.

As soon as the approximate required number of subjects for an interim analysis has received at least one dose of IMP prior to the end of surgery, completed treatment, the relevant data points (including measurement and documentation of blood loss between decision to treat and end of surgery) have been satisfactorily reviewed and the corresponding items/forms have been frozen, a data snapshot will be taken. Further details are specified in the Planned Data Deliverables section of the Data Management Plan. Classification of PDs as major or minor will be agreed PD review meeting and then reviewed during the BDRM meeting based on the snapshot data.

At each interim analysis, the tables and listings which are specified in the document: Biotest 995 TFL Specifications for Interim Analysis of Sample Size (see Appendix 1) will be produced.

For the interim analysis, the sample size will be recalculated according to the observed blood losses and the SDs using nQuery Advisor version 4.0 or higher.

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The interim analysis will be based on unblinded data. The analysis will be performed by an Independent Interim Analysis Team. The interim analysis outputs will be created in a secure, restricted-access storage area on the PPD SAS file-server. All derived datasets, analysis outputs, email communications, etc. created as part of an interim analysis and containing the potential to unblind or result in operational bias, must be handled in a manner such that the Project Team has no access to them. The Independent Reporting Statistician files the final datasets, and any email communication in a secure, restricted-access storage area.

The Independent Reporting Statistician shares only the newly calculated sample size and the recommendation (see rules above) from the interim analysis results with the Sponsor and PPD study team.

In the final BDRM meeting the need for additional subgroup definitions related to length between decision to treat and end of surgery will be evaluated. If required, additional subgroups will be added to the SAP and shells in a new SAP version prior to database lock and the final evaluation.

11.0 Data Review

11.1 Data Handling and Transfer

Please see the Data Management Plan.

11.2 Data Screening

Beyond the data screening built into the PPD Data Management Plan, the PPD programming of analysis datasets and TFLs provides additional data screening. Presumed data issues will be output into SAS logs identified by the word “Problem” and extracted from the logs by a SAS macro and sent to Data Management.

Review of dry run TFL allow for further data screening prior to lock. The PPD statistician and the sponsor must approve database lock.

12.0 Statistical Methods

All analyses will use SAS® version 9.4 or higher.

Subject data listings will be ordered by randomized or actual treatment (‘Not randomized’ / ‘Not treated’, ‘BT524’, ‘FFP’, ‘Cryoprecipitate‘), subject number (numerically, ascending) and visit of assessment respectively where applicable. All tables will be presented by treatment arm (‘BT524’, ‘FFP/Cryoprecipitate‘) and overall. Where applicable tables will also be presented by treatment (‘BT524’, ‘FFP’, ‘Cryoprecipitate‘) or by surgery (spinal, abdominal).

Unless otherwise noted, categorical variables will be summarized using number of observations (n), counts and percentages within category. Percentages will be rounded to one decimal place; except 100% will be displayed without any decimal places and percentages will not be displayed for zero counts.

Unless otherwise noted, continuous variables will be summarized using the number of observations (n), arithmetic mean, SD, minimum, Q1, median, Q3 and maximum. The minimum and maximum values will be displayed to the same level of precision as the raw data, the mean, median, Q1 and Q3 to a further decimal place and the SD to two additional decimal places. Unless stated otherwise the calculation of percentages will be based on the total number of subjects in the population of interest. Thus counts of missing observations will be included in the denominator and presented as a separate category.

The significance level will be 2.5% (one-sided), confidence intervals will be 95% (two-sided). All statistical tests (p-values) will be two-sided, unless otherwise stated.

12.1 Subject Disposition

The number of subjects screened and, the number and percentage who failed screening prior to surgery or during surgery, together with the main reasons, will be summarized for the All Subjects Enrolled Set. The number of subjects randomized, number and percentage of subjects treated with IMP, number and

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percentage of subjects whose treatment was unblinded during the surgery will be summarized by treatment, treatment arm and overall for the All Subjects Enrolled Set. The subject disposition summary will be repeated for spinal and abdominal surgery.

The number of subjects included in each subject analysis set will be summarized by treatment arm and overall for the SAF. The number and percentage of subjects who completed the study or prematurely withdrew/discontinued from the study with the reason for withdrawal/discontinuation will be summarized by treatment, treatment arm and overall for the SAF.

The number and percentage of subjects treated at each country and center will be summarized by treatment, treatment arm and overall for the SAF.

A listing of details of whether the patients completed or discontinued study prematurely will be presented for the SAF. The informed consent and eligibility will be listed for the All Subjects Enrolled Set. The randomization data will be listed for the All Subjects Enrolled Set. A listing of analysis set allocation will also be created for the All Subjects Enrolled Set.

12.2 Protocol Deviations

Deviations from the CSP will be documented on an on-going basis during conduct of the clinical study based on monitoring reports (e.g., failure of eligibility criteria), data management checks and statistical programming (e.g., prohibited medications based on drug codes). Protocol deviations will be discussed and agreed in the BDRM to find PDs with major impact on subject safety or the validity of the study data. Subjects with major PDs will be excluded from the PPS under the assumption that the deviation may have an impact on the efficacy analysis.

In general, PDs will be considered according to the following general categories:

- Inclusion criteria
- Exclusion criteria
- Study drug
- Assessment – safety
- Laboratory/ endpoint data
- Visit window
- Prohibited concomitant medications
- Overdose/ misuse
- Informed consent
- Other

Protocol deviations will be summarized by treatment arm and overall and listed for the FAS. The protocol deviation summary will be repeated for spinal and abdominal surgery.

12.3 Treatments

12.3.1 Extent of Study Drug Exposure

After a bleeding (risk) assessment only subjects requiring hemostatic treatment (high risk for the need of fibrinogen supplementation with BT524 or FFP/cryoprecipitate) during surgery, will be randomized and treated with BT524 or FFP/cryoprecipitate. If the bleeding remained unchanged and an ongoing relevant blood loss will be confirmed or the hemostatic control is not considered sufficient and requires further intervention or a new major blood loss occurred in the course of the surgery, subjects can be treated with repeated IMP administration according to their randomized treatment group.

As the IMP (BT524 or FFP/cryoprecipitate) will be administered intravenous (IV) to each subject under the supervision of the unblinded anesthesiologist, the compliance is expected to be 100%. In addition, the assessment of plasma fibrinogen concentrations may also serve as an adherence measure.

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Exposure of IMP will be summarized by treatment separately for the SAF and repeated by surgery type (spinal or abdominal).

Descriptive statistics will be provided by treatment for the

- number and frequency of IMP administration(s) during surgery
- planned volume of IMP for each (1st, 2nd, …) infusion
- actual volume of IMP administered for each (1st, 2nd, …) infusion
- total planned volume of IMP administered during surgery
- total actual volume of IMP administered during surgery
- time to start of first IMP infusion
- duration of each (1st, 2nd, …) IMP infusion
- number of interruption(s) during surgery for those subjects whose treatment administration was interrupted for the SAF
- actual gram of IMP (BT524) received by type of surgery and total

A listing will be created by subject number for infusion administration of IMP for the SAF.

12.3.2 Prior and Concomitant Medications or Treatment

The most updated version of the World Health Organization drug dictionary will be used to classify prior and concomitant medications by preferred term. Medications will also be coded with the lowest available Anatomical Therapeutic Chemical (ATC) classification system (level 4 to level 2).

Prior medication is defined as any medication with start date prior to IMP start date, irrespective of the stop date. Prior medications will be summarized by ATC classification and preferred term in treatment arms and overall for the SAF. The number and percentage of subjects using each medication will be displayed together with the number and percentage of subjects using at least one prior medication within each medication group and subgroup. This will be repeated by surgery type.

Concomitant medication is defined as any medication taken on or after the start date of IMP, irrespective of start date. Any concomitant treatment (surgeries or procedures) will be coded as “All Other Therapeutic”. Concomitant medications/treatment categorized by ATC classification and preferred term will be summarized by treatment arm and overall for the SAF. The number and percentage of subjects using each medication will be displayed together with the number and percentage of subjects using at least one medication within each medication group and subgroup. This will be repeated by surgery type.

A medication can be both prior and concomitant if it was taken both prior and on or after the date of IMP.

In addition, prohibited medications will be tabulated.

All prior and concomitant medications will be listed separately for the SAF.

12.3.3 Medical and Surgical History, and Concomitant Disease

Medical and surgical history, and concomitant disease will be coded using the most updated version of the MedDRA® dictionary. Medical and surgical history until signature of informed consent, and concomitant disease will be summarized separately by MedDRA system organ class and preferred term in treatment arms and overall for the SAF. These will also be repeated by surgery type.

A listing of medical and surgical history, and concomitant disease will be created for the SAF.

12.3.4 Transfusion Products

The type of transfusion products includes autologous blood transfusion/cell salvage, allogenic platelet concentrates, allogeneic RBCs, allogeneic FFP, cryoprecipitate and other.

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The number and percentage of subjects, using any transfusion products will be summarized by treatment and treatment arm for the SAF. This will be repeated by surgery type. The number and percentage of subjects will also be presented for each type of transfusion product. The volume (mL) of administration for each type of transfusion product will be summarized as well. This will also be repeated by surgery type. These summaries will also be performed separately for RBCs taken before start of first IMP (SAF) and for each type of transfusion product after start of first IMP (SAF). Transfusion products will be listed for the All Subjects Enrolled Set.

12.4 Demographic and Baseline Characteristics

Descriptive summaries of demographic and baseline characteristics will be presented by treatment, treatment group and overall, for FAS, mFAS, SAF and PPS. The following demographic characteristics will be summarized in the following order in the tables: sex, race, ethnicity, age, weight, height, body mass index, childbearing potential, and predictive blood loss. The summary will also be repeated for surgery type.

A listing will be created to show all the demographic and baseline characteristics for the All Subjects Enrolled Set.

A listing will be created to show results for pregnancy test of female subjects with childbearing potential for the SAF.

12.4.1 Surgery

The detailed information of surgery, including e.g., type of surgery (categorized by spinal and abdominal), predictive blood loss, duration of surgery, time until decision to treat subject, time to end of surgery after decision to treat, and overall vital signs monitoring during surgery will be listed for the All Subjects Enrolled Set. Number and percentage of subjects for each type of surgery, descriptive statistics for duration of surgery, time until decision to treat (relative to start of surgery), time to end of surgery (relative to time of decision to treat) and time of IMP exposure during surgery will be presented by treatment group and overall for FAS. The surgery summary, without type of surgery will be repeated for spinal and abdominal surgery.

12.5 Efficacy Analyses

12.5.1 Primary Efficacy Parameter

The primary efficacy parameter is the intra-operative blood loss after decision to treat the subject with IMP until the end of surgery as measured and calculated by amount of blood from the blood suction unit and amount of blood from swabs, surgical cloths and compresses.

The primary objective of this study is to demonstrate that BT524 is non-inferior that means not worse than FFP/cryoprecipitate with a non-inferiority margin of 150 mL in reducing intra-operative blood loss by intravenous (IV) administration in subjects with acquired hypofibrinogenemia undergoing elective major spinal or abdominal surgery. The primary analysis of this endpoint is a non-inferiority test in the PPS. The null hypothesis for the primary analysis is that the degree of inferiority of BT524 compared to standard treatment (FFP/cryoprecipitate) is greater than or equal to the non-inferiority margin. The alternative hypothesis is that the degree of inferiority of BT524 compared to FFP/cryoprecipitate is less than the non-inferiority margin.

$$H_0: \mu_1 - \mu_2 \geq \delta$$
$$H_1: \mu_1 - \mu_2 < \delta$$

where

μ_1 = mean intra-operative blood loss after the decision to treat the subject with IMP in the BT524 treatment arm

μ_2 = mean intra-operative blood loss after the decision to treat the subject with IMP in the FFP/cryoprecipitate treatment arm

δ = non-inferiority margin = 150 mL

The final analysis will be performed using a two-way analysis of variance (ANOVA) with the intra-operative blood loss after the decision to treat the subject with IMP until the end of surgery as the dependent variable

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and the predictive blood loss (> 1,000 mL to ≤ 2,000 mL and > 2,000 mL) as a covariate. The least square means and treatment difference (BT524 versus FFP/cryoprecipitate) will be presented with the corresponding 2-sided 95% confidence intervals and p-value. Non-inferiority will be demonstrated if the upper confidence limit of the 95% confidence interval for the difference in the least square means is less than the non-inferiority margin (150 mL). Assumptions for the ANOVA model will be evaluated by diagnostic (e.g., Shapiro-Wilk test for normality) and graphical methods such as residual plots, including distribution of residuals, and Q-Q plots of residuals. The ANOVA assumptions are reviewed during the BDRM and in case the assumptions are not fulfilled, data transformation and alternative methods are discussed (see sections 12.5.1.3 and 12.5.1.4).

In addition, the following analyses will be presented:

- Sensitivity analysis of the same analysis repeated for the FAS and mFAS
- Sensitivity analysis of spinal surgery subjects with no tumors.

12.5.1.1 Assessment of Superiority

If the non-inferiority is demonstrated by the primary analysis, then superiority will be assessed in the FAS and mFAS and repeated for surgery type, sex and race. For superiority analysis, the null hypothesis is that the blood loss in BT524 treatment arm is greater than that in FFP/cryoprecipitate treatment arm. The alternative hypothesis is that the blood loss in BT524 treatment arm is less than that in FFP/cryoprecipitate treatment arm.

$$H0: \mu1 - \mu2 \geq 0$$
$$H1: \mu1 - \mu2 < 0$$

where μ1 and μ2 are the same as above.

The same ANOVA model as the non-inferiority analysis will be used. The least square means and difference of least square means will be presented together with the 95% confidence intervals and 2-sided p-value. The superiority of BT524 is demonstrated if the 2-sided p-value is less than 0.05.

12.5.1.2 Sub-Group Analyses: Predicted Blood Loss

In addition to the primary analysis, a subgroup analysis of the primary endpoint will also be performed in the mFAS. Subgroups are predictive blood loss, as well as spinal and abdominal surgery. These subgroups will be further splitted by sex and race.

Primary endpoint will be analyzed using a one-way ANOVA in each subgroup. The least square means and difference of least square means together with 95% confidence intervals and p-values in each subgroup will be presented.

The intra-operative blood loss after decision to treat with IMP will be summarized descriptively overall and for subgroups predictive blood loss and surgery (spinal, abdominal) and will be listed for the FAS. In addition, intra-operative blood loss after decision to treat with IMP will be summarized as continuous data as a box plot for both treatment arms for the mFAS and the PPS and subgroups predictive blood loss and surgery (spinal, abdominal) for the mFAS.

12.5.1.3 Data Transformation

In the case that the statistical assumptions for the ANOVA model are shown to be violated via the diagnostic tests then the primary efficacy parameter will be log-transformed. The ANOVA will then be re-run and checked via the diagnostic tests. If the ANOVA assumptions now hold this will be used. If they do not hold then the alternative method shall be used on the non-log-transformed data.

12.5.1.4 Alternative Method

The statistical assumptions for the ANOVA model will be tested in regards to the primary endpoint. Primarily the outcome of a Shapiro-Wilk test and graphical quantile-quantile (QQ) plots will used to assess if the

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assumptions are significantly violated. If deemed necessary other test may be performed as well. The methods used, and the conclusions will be discussed in the clinical trial report (CTR).

In case there is a significant violation to the assumptions for the ANOVA model for the primary endpoint, a non-parametric alternative method will be implemented, proposed by van Elteren for stratified or blocked continuous response variables.

This alternative method will be used as a sensitivity analysis to the primary non-inferiority analysis for the PPS, FAS and mFAS. It will also be used as a sensitivity to the superiority analysis for the FAS and mFAS.

12.5.2 Multiplicity

The primary variable is only tested for superiority when the non-inferiority is demonstrated by the primary analysis, which will not lead to inflation of the Type-I error.

The statistical analyses on secondary variables are of exploratory nature. No adjustment for multiplicity is needed.

12.5.3 Secondary Efficacy Parameters

All analyses of the secondary efficacy parameters will be performed on the FAS overall and for spinal surgery and abdominal surgery.

12.5.3.1 Correction of Fibrinogen Level

The proportion (%) of subjects with a successful correction of fibrinogen level (by FIBTEM A10) 15 minutes after start of first IMP administration will be compared between the treatment arms using a Cochran-Mantel-Haenszel (CMH) approach stratified by predictive blood loss (> 1,000 mL to ≤ 2,000 mL and > 2,000 mL). The number and percentage of subjects with a successful correction of fibrinogen level 15 minutes after start of first IMP administration will be presented for both treatment arms with corresponding 95% confidence intervals. The estimated treatment effect (i.e., the difference in response rate between the treatment arms, BT524 – FFP/cryoprecipitate), corresponding 95% confidence interval, and 2-sided p-value for the difference will be presented.

The time to first successful correction of fibrinogen level (by FIBTEM A10) is assigned to the following categories: "Within 15 minutes after IMP start", "Greater than 15 and less than or equal to 90 minutes after IMP start", "Greater than 90 minutes after IMP start" and “Unsuccessful correction”. The 4 categories will be compared between the two treatment arms using a Chi-square test.

In an additional analysis the time to first successful correction of fibrinogen level (by FIBTEM A10) is classified relative to the end of surgery into "Until end of surgery", "Not within surgery" or “Unsuccessful correction”. The 3 categories will be compared between the two treatment arms using a Chi-square test.

For the above Chi-square tests, the number and percentage of subjects in each category will be presented for each treatment arm, together with an overall p-value for the difference between the two treatment arms.

The absolute values and change from baseline in fibrinogen levels by FIBTEM A10, together with the rate of fibrinogen restoring will be summarized with descriptive statistics over time by treatment arm. The absolute values at screening and change from screening at baseline will also be summarized for fibrinogen levels by FIBTEM A10.

In addition, the test results of MCF and Clauss assay will be summarized in the same way. The results of these three assays will be listed for the All Subjects Enrolled Set.

Mean with SD, median with interquartile ranges will be presented graphically over time for both treatment arms for FIBTEM A10, MCF and Clauss assay.

Pearson’s Correlation will be tabulated between the FIBTEM A10 and Clauss assay for baseline and prior first dose values in the Fibrinogen FIBTEM A10 summary tables.

Analysis will be repeated for spinal and abdominal surgery.

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12.5.3.2 Consumption of Transfusion Products after IMP until End of Surgery

The type of transfusion products includes autologous blood transfusion/cell salvage, allogenic platelet concentrates, allogeneic RBCs, allogeneic FFP, cryoprecipitate and other. Consumption of transfusion products is measured by the total amount of transfusion product (volume in mL and units [bags]) infused after start of first IMP and until the end of surgery.

This secondary efficacy endpoint will be descriptively summarized for volume (mL) and units (bags) of transfusion product for each product type by treatment arm. The number and percentage of subjects, using any transfusion product, together with the number and percentage of subjects using each transfusion product type will also be summarized by treatment arm.

Analysis will be repeated for spinal and abdominal surgery.

Two box-whisker plots will be created to present the amount of each type of transfusion product after start of first IMP until end of surgery by product type. One plot will show volume in mL and the other will show units (bags). Analysis will be repeated for spinal and abdominal surgery.

All the consumption of transfusion products in this study will be listed for the All Subjects Enrolled Set.

Summaries of subjects will also be provided for subjects with total avoidance of transfusion products post IMP administration. This will be repeated by surgery type.

12.5.3.3 Amount of RBCs infused after IMP until End of Surgery

The amount (volume in mL and units [bags]) of RBCs (allogeneic and autologous) infused after start of first IMP administration until the end of surgery will be summarized by treatment arm.

An ANOVA analysis will be performed with the amount of RBCs required as the dependent variable and the predictive blood loss (> 1,000 mL to ≤ 2,000 mL and > 2,000 mL) as a covariate. The least square means and difference in least square means will be presented with the corresponding 95% confidence intervals and 2-sided p-value. This will be repeated by surgery type.

In addition, a subgroup analysis of the amount of RBCs will be performed for the FAS. Subgroups are predictive blood loss as well as spinal and abdominal surgery. The amount of RBCs will be analyzed using an ANOVA for each subgroup. The least square means and difference of least square means together with 95% confidence intervals and p-values for each subgroup will be presented.

12.5.3.4 Post-operative Blood Loss

The post-operative blood loss in the first 24 hours is the blood loss from end of surgery until 24 hours after end of surgery. This secondary efficacy endpoint will be descriptively summarized by treatment arm.

An ANOVA analysis will be performed with the post-operative blood loss in the first 24 hours as the dependent variable and the predictive blood loss (> 1,000 mL to ≤ 2,000 mL and > 2,000 mL) as a covariate. The least square means and difference (BT524 versus FFP/cryoprecipitate) in least square means will be presented with the corresponding 95% confidence intervals and 2-sided p-value. This will be repeated by surgery type.

A box-whisker plot for post-operative blood loss will be generated for both treatment arms.

In addition, a subgroup analysis of the post-operative blood loss in the first 24 hours will be performed for the FAS. Subgroups are spinal and abdominal surgery. The post-operative blood loss will be analyzed using an ANOVA for each subgroup. The least square means and difference of least square means together with 95% confidence intervals and p-values for each subgroup will be presented.

Post-operative blood loss will be listed for the FAS.

12.5.3.5 Proportion of Subjects with Rebleeds

The proportion of subjects with rebleeds will be compared between the treatment arms using a CMH approach stratified by predictive blood loss (> 1,000 mL to ≤ 2,000 mL and > 2,000 mL). The number and

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percentage of subjects with a rebleed will be presented with corresponding 95% confidence intervals. The estimated treatment effect (i.e., the difference in rebleed rate between the treatment arms, BT524 – standard treatment (FFP/cryoprecipitate)), corresponding 95% confidence interval, and 2-sided p-value for the difference will be presented. The descriptive summary of the number of rebleeds will also be presented by treatment arm.

Analysis will be repeated for spinal and abdominal surgery.

Post-operative rebleeds will be listed for the FAS.

12.5.3.6 Length of Hospital Stay after Surgery

Subjects without a date of discharge (i.e., hospitalization ongoing) at Day 36 will be classified into > 36 days. The frequency of subjects whose length of hospital stay falls in the following categories are presented: 1-7, 8-14, 15-21, 22-28, 29-36, > 36 days.

The length of hospital stay after surgery will be summarized descriptively by treatment arm for those subjects, who had a day of discharge until closing visit. The details of surgery date, discharge or ongoing hospital stay, as well as length of hospital stay will be listed for the FAS.

Analysis will be repeated for spinal and abdominal surgery.

12.5.3.7 In-hospital Mortality

The number and percentages of subjects who died during their hospital stay will be presented with corresponding 95% confidence intervals of death rate by treatment arm using the Clopper-Pearson Method.

Analysis will be repeated for spinal and abdominal surgery.

The details of mortality during hospital stay will be listed for the FAS.

12.5.3.8 Overall Mortality

The number and percentages of subjects who died will be presented with corresponding 95% confidence intervals of death rate by treatment arm using the Clopper-Pearson Method.

The details of overall mortality will be listed for the FAS.

12.6 Safety Analyses

All safety analyses will be based on the SAF.

12.6.1 Adverse Events

Adverse events will be coded using the most updated version of the MedDRA® dictionary.

Adverse events occurring during or after the administration of study medication are TEAEs. If a partially missing AE start date is imputed, the imputed date will be used to assess if the AE is treatment emergent. If AE start date is completely missing and AE end date/time is after IMP start date/time or if it is completely missing, then it is a TEAE. All other AEs are regarded non-treatment emergent.

The search criteria for the adverse events of special interest are specified in the table below:

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AESI	Search Criteria
Thrombosis or TEE	SMQ: Embolic and thrombotic events (narrow)
Relevant bleeding complication	All AESIs ticked as AESI, but not falling under categories “Thrombosis or TEE” or “Suspicion of transmission of infective agents (viral safety)”
Suspicion of transmission of infective agents (viral safety)	Preferred terms: “transmission of infectious agent via product” and “suspected transmission of infectious agent via product”

Adverse events related to Hypersensitivity/anaphylactic reactions/anaphylactic shock will be selected by using the following criteria: SMQ: Hypersensitivity, (narrow), Anaphylactic reaction (narrow), and Anaphylactic shock conditions (narrow).

Adverse events related to bleeding related ischaemic events will be selected by using the following criteria: SMQ: Ischaemic heart disease: Myocardial infarction (narrow), Other ischaemic heart disease (narrow); Ischaemic colitis (broad); Ischaemic central nervous system vascular conditions (narrow); Embolic and thrombotic events, arterial (narrow); Embolic and thrombotic events, venous (narrow); PT: Peripheral ischaemia; Ischaemia; Gastric ischaemia; Hepatic ischaemia; Ischaemic contracture of the left ventricle; Dry gangrene; Ischaemic gastritis; Ischaemic limb pain; Necrosis ischaemic; Spleen ischaemia; Uterine ischaemia.

If a serious adverse event (SAE) occurs in a subject after the period of observation, i.e., after the last study visit, and is considered by the investigator to be related to the study medication, this should still be recorded as a SAE. If the eCRF has been closed for the subject, the investigator should contact the sponsor to determine how to report the SAE.

The total number and percentage of subjects reporting at least one AE and the absolute count of AEs will be tabulated for each treatment, treatment arm and overall once for the overall trial population and separately by type of surgery (spinal and abdominal surgery), sex and race. The initial summary will also provide a breakdown of the following:

- Any AE
- Any non-severe AE
- Any TEAE
- Any NTEAE
- Any non-serious TEAE
- Any non-severe TEAE
- Any treatment-related TEAE
- Any severe AE
- Any severe TEAE
- Any severe treatment-related TEAE
- Any SAE
- Any serious TEAE
- Any serious treatment-related TEAE
- Any AESI
- Any treatment-emergent AESI
- Any treatment-emergent treatment-related AESI
- Any AE leading to discontinuation from study
- Any TEAE leading to discontinuation from study

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- Any treatment-related TEAE leading to discontinuation from study
- Any AE with outcome of death
- Any TEAE with outcome of death
- Any treatment-related TEAE with outcome of death
- Any TEAE of special interest
    - Thrombosis or TEE
    - Suspicion of transmission of infective agents
    - Relevant bleeding complication
- Any TEAE of Hypersensitivity/ anaphylactic reactions/ anaphylactic shock
- Any TEAE of Bleeding related ischaemic events

The number of events, number and percentage of subjects reporting AEs will be tabulated (as well for causality and maximum severity) in the following way once for the overall population, and separately by type of surgery (spinal and abdominal surgery) and sex in the following:

- Treatment-emergent AEs by system organ class and preferred term
- Serious treatment-emergent AEs by system organ class and preferred term
- Causally related TEAE by system organ class, preferred term and maximum severity
- Not Causally related TEAE by system organ class, preferred term and maximum severity
- Causally related Serious TEAE by system organ class, preferred term and maximum severity
- Not Causally related Serious TEAE by system organ class, preferred term and maximum severity

The number of events, number and percentage of subjects reporting AEs will be tabulated (as well for causality and maximum severity) in the following way once for the overall population, and separately by type of surgery (spinal and abdominal surgery) in the following:

- Non-treatment emergent AEs by system organ class and preferred term (will not be tabulated for maximum severity or causality)
- Treatment-emergent AEs by system organ class and preferred term of Hypersensitivity/ anaphylactic reactions/ anaphylactic shock
- Treatment-emergent AEs by system organ class and preferred term of Bleeding Related Ischaemic Events
- Treatment-emergent AEs by system organ class and preferred term leading to study discontinuation (will not be tabulated for maximum severity, causality or separately by type of surgery)
- Treatment-emergent AEs by system organ class and preferred term leading to death (will not be tabulated for maximum severity or separately by type of surgery)
- Treatment-emergent AEs by system organ class and preferred term leading to hospitalization (will not be tabulated for maximum severity or causality)

The following AESI categories (only treatment-emergent adverse events) will be presented by system organ class and preferred term for the overall population, and separately by type of surgery and sex. They are repeated for maximum severity and causality for overall and surgery type only.

- Adverse events of special interest:
    - Thrombosis or TEE
    - Suspicion of transmission of infective agents (presented only for the overall population and for causality)

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- Relevant bleeding complication

Listings will be produced for all AEs (including non-treatment emergent adverse events) reported in the eCRF for the All Subjects Enrolled Set.

In the listing, time to onset will be presented. Time to onset of the adverse event is defined as start date/time of adverse event – date/time of first study drug administration.

12.6.2 Deaths and Serious Adverse Events

Listings for SAEs and subjects with AEs with the outcome of death will also be provided for the SAF.

12.6.3 Laboratory Data

The following clinical laboratory variables will be summarized for the overall population, and separately by type of surgery (spinal and abdominal surgery):

Hematology	Red blood cells (RBC), white blood cells (WBC), platelet count, hemoglobin, hematocrit, neutrophils (absolute and %), lymphocytes (absolute and %), monocytes (absolute and %), eosinophils (absolute and %) and basophils (absolute and %)
Biochemistry	Aspartate aminotransferase, alanine aminotransferase, creatinine, creatinine clearance (Cockcroft and Gault), blood urea nitrogen (BUN), urea, gamma-glutamyltransferase (γ-GT), alkaline phosphatase (AP), total bilirubin, direct bilirubin, indirect bilirubin, potassium, sodium, calcium and chloride
Urinalysis	pH, blood, WBC, protein, glucose, ketone bodies, nitrite, bilirubin and urobilinogen
Markers of Coagulation	PT, INR, aPTT, TAT, F_{1+2}, D-dimer, PS, PC, AT III, TT and ETP(TGT)
Coagulation factors	FII, FV, FVII, FVIII, FIX, FX, FXI, FXIII and vWF

Note: Protein C (PC) is also referred to as Factor XIV. Direct bilirubin and indirect bilirubin are only assessed in case of an elevated bilirubin value and are thus not generally included in statistical summaries.

Quantitative laboratory parameters reported as “< x” or “ y” > will be imputed with x/2 and y, respectively, for inclusion in summary statistics. The reported value will be presented in listings.

Descriptive statistics for clinical laboratory values (in SI units) and changes from baseline at each scheduled assessment visit (as stated in Flowchart of study) with the exception of urinalysis parameters and direct/indirect bilirubin will be presented by treatment and treatment arm.

Laboratory results of all hematology, biochemistry, urinalysis parameters and PT/INR, aPTT will be categorized with respect to the laboratory specific reference ranges as normal/abnormal (i.e., high [H], low [L], where applicable). Abnormal values will be further classified with respect to clinical relevance by the investigator. The results and percentages will be summarized by visit and overall for each treatment and treatment arm.

Shifts from baseline to each post-baseline visit for all laboratory variables (except direct and indirect bilirubin) will be summarized by treatment and treatment arm using number and percentage of subjects.

In addition, a shift summary of hematology for number of subjects with change from normal or not clinical significant to clinical significant will be presented.

All the laboratory data will be listed for the SAF.

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12.6.4 Vital Signs

Descriptive statistics for vital signs (systolic and diastolic blood pressure, pulse rate, respiratory rate and body temperature) and their changes from baseline at each scheduled study visit will be presented by treatment and treatment arm. This will be repeated by surgery type.

The overall vital signs monitoring results during surgery will be tabulated by number and percentage of subjects in each treatment, treatment arm and overall. This will be repeated by surgery type.

All vital signs will be listed for the SAF.

12.6.5 Physical Examinations

All the physical examination parameters will be listed for the SAF.

12.6.6 Viral Status

A shift table from screening to closing visit for virus serology parameters (hepatitis B, hepatitis C and HIV) will be summarized by treatment and treatment arm using number and percentage of subjects.

Listings for viral status will be created respectively for the SAF.

13.0 Validation

PPD goal is to ensure that each TFL delivery is submitted to the highest level of quality. Our quality control procedures will be documented separately in the study specific programming quality control plans.

14.0 References

PPD

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Appendix 1 Tables, Figures, Listings, and Supportive SAS Output Appendices

Clinical Study Report:

Please see the separate document: Biotest 995 TFL Specifications.

DSMB:

Please see the separate documents for the DSMB Charter and a List of DSMB TFL Specifications.

Blinded Sample Size Recalculation:

Please see the separate document: Biotest 995 TFL Specifications for Sample Size Recalculation.

Interim Analyses for Sample Size Assessment:

Please see the separate document: Biotest 995 TFL Specifications for Interim Analysis of Sample Size.

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