CLI 00125

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STATISTICAL ANALYSIS PLAN

October 16, 2023

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STATISTICAL ANALYSIS PLAN

CLI 00125

Protocol Title:	A Randomized, Double-Blinded, Controlled, Parallel Group, Non-inferiority, Phase III Study to Evaluate the Efficacy and Safety of the INTERCEPT Blood System for Red Blood Cells in Patients undergoing Complex Cardiac Surgery Procedures (the ReCePI study)
Product:	Red Blood Cells treated with the INTERCEPT Blood System
Protocol Number:	CLI 00125 (Amendment 8.0)
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ABBREVIATIONS

AE	Adverse Event
AKI	Acute Kidney Injury
ATC	Anatomical-Therapeutic-Chemical
ATSP	Acute Transfusion Support Period
CMH	Cochran-Mantel-Haenszel
CI	Confidence Interval
Control	Conventional RBCs
CRO	Contract Research Organization
CSR	Clinical Study Report
eCRF	Electronic Case Report Form
IPCW	Inverse Probability of Censoring Weighting
MAR	Missing at Random
MCMC	Markov Chain Monte Carlo
MedDRA	Medical Dictionary for Regulatory Activities
MI	Multiple Imputation
mITT	Modified Intention To Treat
MMRM	Mixed Models for Repeated Measures
M&N	Miettinen and Nurminen
MNAR	Missing Not At Random
PMM	Pattern Mixture Modeling
PPS	Per-Protocol Set
PT	Preferred Term
RBC	Red Blood Cell
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SAS	Statistical Analysis System
sCr	Serum Creatinine
SOC	System Organ Class
SROS	Study RBC Only Set
TE	Treatment Emergent
TEAE	Treatment Emergent Adverse Event
Test	INTERCEPT RBCs
TR	Transfusion Reaction
TRUST	Transfusion Risk Understanding Scoring Tool
US	United States

1. INTRODUCTION

This statistical analysis plan (SAP) specifies the statistical methods to be implemented for the analysis of data collected from the ReCePI study within the scope of Cerus' Protocol CLI 00125, "A Randomized, Double-Blinded, Controlled, Parallel Group, Non-inferiority, Phase III Study to Evaluate the Efficacy and Safety of the INTERCEPT Blood System for Red Blood Cells in Patients undergoing Complex Cardiac Surgery Procedures (the ReCePI study)." It applies to the study protocol (version 7.0) dated 20 December 2021 and provides detailed instructions as to how each analysis will be performed.

Results obtained from the analyses specified in the final approved version of the SAP will become the basis of the clinical study report (CSR) for this study. Post-hoc exploratory analyses not necessarily identified in this SAP may be performed to further examine study data. Any post-hoc, or unplanned, exploratory analysis will be clearly identified as such in the final CSR.

2. CLINICAL BACKGROUND

The ReCePI study is based on the premise that renal impairment within 48 hours of complex cardiac surgery is an indicator of inadequate tissue oxygenation and is a prognostic indicator that correlates with day 30 post surgery mortality (Lassnigg 2004, Lassnigg 2008). Renal hypoxia is a major determinant of renal impairment, measured as a change in the subjects' serum creatinine (sCr). Renal hypoxia may be caused by impaired renal blood flow, anemia, or qualitative dysfunction of red blood cells resulting in inadequate renal oxygenation. RBCs transfused to correct anemia are hypothesized to prevent renal hypoxia and impairment both by augmenting blood volume and by providing increased oxygen carrying capacity resulting in improved tissue oxygenation. This study is designed to demonstrate non-inferiority of INTERCEPT RBCs (Test) compared with conventional RBCs (Control) in preventing renal tissue hypoxia, as measured by the change in sCr from baseline prior to surgery to sCr levels within the 48-hour period following the completion of surgery.

It is not possible to evaluate the efficacy of transfused Test and Control RBCs in isolation, as study RBCs are naturally diluted by the patients' own blood during transfusion, as well as by re-transfused cell saver blood (a mixture of autologous and transfused RBCs) collected and transfused during surgery. In addition, the study protocol recognizes that cardiac surgery patients may unpredictably require large numbers of RBC components that exceed the available supply of RBCs prepared in accordance with the study protocol, resulting in the transfusion of non-study conventional RBC components. RBC transfusion under these circumstances is "per protocol", however, every effort has been made to avoid non-study RBC transfusions. Prevention of non-study RBC transfusions has proven a challenge. We assume that the use of non-study RBC will be random with respect to treatment assignment into the Test and Control groups as the manufacture of Test and Control RBC inventories has been balanced.

The use of cell-saver and non-study RBC transfusions is potentially problematic, and both the classical mITT and PP analyses study are subject to potential biases, despite execution of the study in compliance with the protocol.

As expected from prior literature review and confirmed by examination of blinded study data, the risk of the primary endpoint (AKI) and increasing number of RBCs transfused during the acute study transfusion period are proportional to the complexity and duration of the cardiac surgery procedures performed and the patients underlying condition and disease prior to surgery. Patients who receive larger numbers of RBC transfusions are more likely to meet the primary efficacy endpoint and are also more likely to receive non-study RBC transfusions per protocol due to a requirement for large volume or massive transfusions. For these reasons, the mITT analysis set best represents an assessment of the efficacy of Test RBCs, despite the inclusions of subjects who receive non-study RBCs within the 48 hours post-surgery analysis period of the primary efficacy endpoint.

The SAP presents the primary non inferiority analysis as performed on the mITT analysis set that will include all randomized subjects transfused with any type of RBC component during or within 48 hours of surgery, including those who receive non-study RBCs and autologous cell-saver RBCs in addition to study RBC transfusion(s). Various sensitivity analyses using the mITT analysis set are proposed to assess the robustness of the primary analysis.

Additional exploratory analyses on two subpopulations are presented for comparison purposes: First, a Per Protocol Set (PPS) that excludes important protocol deviations but includes cell saver and non-study RBC transfusions given as described in the protocol. Second, a Study RBC Only Set (SROS) that includes subjects who received only study RBCs (and cell saver RBCs) according to the randomization assignment within the 48-hour post-surgery primary endpoint assessment period. Analyses will be performed to determine the effect of including incremental numbers of non-study RBC transfusions. For the purposes of the statistical analysis plan, ≥4 RBC units related to an 8-hour surgery and ≥10 units of RBCs within 24 hours represents larger volume or massive transfusion, respectively.

3. OBJECTIVE(S) AND ENDPOINTS

3.1 Objectives

The objective of this study is to evaluate the efficacy and safety of red blood cell (RBC) transfusion for support of acute anemia in cardiovascular surgery patients based on the clinical outcome of renal impairment following transfusion of RBCs treated with the INTERCEPT Blood System for Red Blood Cells (INTERCEPT RBCs) compared to patients transfused with conventional RBCs.

3.2 Endpoints

3.2.1 Primary Efficacy Endpoint

The primary efficacy endpoint is:

- Any raised sCr level, occurring after transfusion of a study RBC, of ≥ 0.3 mg/dL (or 26.5 μmol/L) from the pre-surgery baseline within 48±4 hours of the end of surgery.

3.2.2 Secondary Efficacy Endpoints:

- The incidence of Stage I, II or III Acute Kidney Injury (KDIGO 2012) based on changes in sCr levels from pre-surgery baseline and the need for renal replacement therapy (RRT) post-surgery.
- Mortality or the need for RRT by 30 days post-surgery.

3.2.3 Safety Endpoints

The primary safety endpoints are:

- Proportion of patients with any treatment-emergent adverse events (TEAEs) possibly, probably, or definitely related to study RBC transfusion through 28 days after the last study transfusion.
- Proportion of patients with treatment emergent (TE) antibodies with confirmed specificity to INTERCEPT RBCs by end of study (i.e., 75±15 days after the last study transfusion).

Additional safety assessments will include:

- Treatment-emergent AEs through 28 days after the last study transfusion.
- Treatment-emergent SAEs through 28 days after the last study transfusion.
- Transfusion reactions (as defined by the CDC National Healthcare Safety Network [NHSN] Hemovigilance Module protocol) through 28 days after last study transfusion.
- Treatment-emergent immunization to RBC alloantigens through 28±3 days after the last study transfusion.
- Treatment-emergent immunization to HLA alloantigens through 28±3 days after the last study transfusion.

Please note no formal hypotheses will be tested on the primary safety endpoints.

4. STUDY DESIGN

4.1 General Study Design

The study is a prospective, multicenter, randomized, double-blinded, active controlled, parallel-design, non-inferiority study.

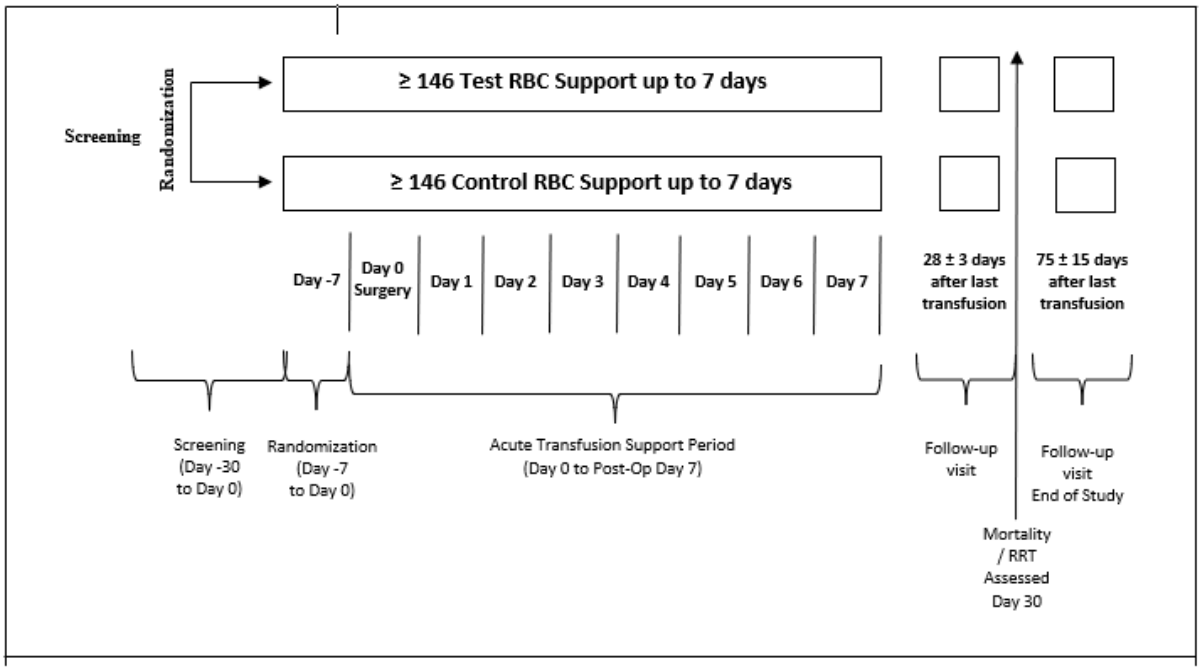
Subjects with high likelihood to received a transfusion as determined by the investigator and who meet eligibility criteria at Screening can be randomized up to 7 days before or on the day of surgery, but prior to the start of surgery.

Randomized subjects will receive as clinically indicated and determined by the treating physician only study RBCs during the Acute Transfusion Support Period (ATSP), starting from surgery till post-operative Day 7, hospital discharge or death, whichever is first.

Following the ATSP, Subjects will return for Day 28 and Day 75 Visits after last study transfusion.

At least 292 eligible subjects will be randomized and transfused with study RBC(s) (Figure 1).

Figure 1: Study Design Diagram



4.2 Randomization and Blinding

Subject will be randomized with a 1:1 ratio to receive either INTERCEPT RBCs (Test) or conventional RBCs (Control). Randomization will be stratified by site, pre-existing renal impairment (sCr ≥1.2 mg/dL vs. < 1.2 mg/dL), and surgery procedure based on less vs. more risk for renal complications due to surgical complexity. The randomization schedule is generated and implemented by an independent CRO.

Randomized subjects who do not receive any study RBC transfusions from randomization to within the first 48 hours after completion of surgery will be discontinued from study and replaced.

This is a double-blinded study. The subjects, operating room staff, surgical staff, ICU staff and others caring for participating patients, as well as the sponsor (and delegates) will be blinded to treatment assignment. Only appropriate blood bank staff and unblinded delegates who monitor the production of the RBC components will be able to access the treatment arm assignment. Selected Blood Center staff will not be blinded due to their role required for collection, preparation, and releasing of Test and Control RBC components for transfusion to study subjects.

4.3 Sample Size Estimation

By assuming the proportion of patients with ΔsCr ≥ 0.3mg/dL to be 30% in the Control arm and no more than a 50% increase from the Control rate (15%) as the non-inferiority margin, a sample size of ≥292 patients (146 per arm) will provide approximately ≥80% power to declare non-inferiority at the two-sided 0.05 alpha level, assuming the true treatment difference is zero.

Under the same assumptions, sample sizes of 334 patients (167 per arm) and 392 patients (196 per arm) will provide 85% and 90% power, respectively.

In order to maintain sufficient study power to establish non-inferiority for the primary efficacy endpoint, Cerus plans to transfuse a minimum of 320 subjects and may go up to 400.

4.4 Visits and Assessments

Assessments at each visit and the time/visit window for each post-baseline assessment are specified in the Assessment Schedule (Table 1). For subjects whose study participation is terminated prior to Day 75 Visit, to the extent possible, physical exam, including general neurological examination, should be performed, concomitant medications and adverse events should be recorded. Standard laboratory tests should be collected, and other blood samples should be performed at the Exit Visit.

Table 1: Assessment Schedule

Study Period	Screening / Randomization		Acute Transfusion Support Period (Surgery to Post-operative Day 7, hospital discharge or death, whichever is first)		Follow-up Visits		
	Screening visit	Baseline (Pre-surgery)					
Window / Assessment	Day -30 to Day 0	pre-surgery	Day 0 Surgery	Days 1 – 7, hospital discharge or death	28±3 days After last study transfusion or Early Termination	30 days After Surgery	End of Study (75±15 days After Last Study Transfusion)
Informed Consent /Assent	X						
Demographics	X						
Indication / Type of scheduled surgery	X						
Medical, surgical, transfusion and medication history including radiographic contrast media within 7 days of surgery / the need for irradiated RBC products	X						
Number of prior pregnancies (females)	X						
Physical examination including height and weight	X						
TRUST score components	X						
Randomization	X[#]						
Vital signs (HR, BP, RR)	X[1]	X[3]	X	X[6]	X		
Concomitant medications	X	X[3]	X[7]	X[6]	X		
Weekly telephone calls							
EKG	X[1]	X[3,4]	X[4]	X[4]			
Pregnancy test (if applicable)	X[1,2]	X[2,3]					
Hematology panel	X[1,2]	X[2,3]	X[2]	X[2,6]	X[2]		
Blood chemistry	X[1,2]	X[2,3]	X[2]	X[2,6]	X[2]		
Blood type	X[1]						
Serum Creatinine at 48±4 hours after end of surgery				X			
Hemodynamic Parameters		X[4,5]	X[3,5]	X[3,6]			
Fibrinogen, Troponin		X[4]	X[4]	X[4]			
Urine output			X[8]	X[6,8]			
Procedure details			X				
Study RBC crossmatch and Study RBC transfusion(s)/data			X	X			

Study Period	Screening / Randomization		Acute Transfusion Support Period (Surgery to Post-operative Day 7, hospital discharge or death, whichever is first)		Follow-up Visits		
	Screening visit	Baseline (Pre-surgery)					
Window / Assessment	Day -30 to Day 0	pre-surgery	Day 0 Surgery	Days 1 – 7, hospital discharge or death	28±3 days After last study transfusion or Early Termination	30 days After Surgery	End of Study (75±15 days After Last Study Transfusion)
DAT, IAT	X[1]		X[4,9]	X[4,9]	X		
INTERCEPT RBC antibody screen (S-303-specific antibody screen)	X		X[4,9]	X[4,9]	X		X
Sample for HLA antibodies	X				X		
All other blood components (including non- study RBCs) transfused, if any			X	X	X		
Estimated blood loss from surgery			X				
Estimated blood loss from other source including chest tube(s)				X[6]			
AEs/SAEs/TRs	X*	X*	X*	X*[,6]	X^		
Documentation of vital status and need for RRT					X	X[11]	X

1. For Screening/Randomization purposes, data from medical records within 30 days prior to their surgical procedure may be used.
2. See protocol Table 7.2: Hematology and Chemistry Testing for details on required labs.
3. These assessments can be combined with screening assessments if screening visit is also on Day -1 or Day 0 Pre-Surgery.
4. Data collected on eCRFs only as available from medical records if performed as Standard of Care.
5. Hemodynamic Parameters: Peripheral O2 Saturation, Mean Arterial Pressure, Central Venous Pressure.
6. These assessments are required to be collected daily on Day 1 through Day 7 post surgery or until discharge. If subject returns after discharge for a postoperative standard of care visit, assessments will be collected in eCRFs on the appropriate study day.
7. Any drugs given to induce anesthesia or drugs given as a prophylactic in combination with anesthesia prior to surgery do not need to be recorded. Medications given during anesthesia to treat an AE/SAE need to be recorded.
8. Urine output data collected daily while a urinary catheter is in place. The date entered should reflect the start of the collection and match the visit date.
9. S-303 screen must be performed whenever a routine RBC alloantibody screen (IAT) is performed during the transfusion period.
10. Laboratory results recorded in the eCRFs during this period will only be those assessed as clinically significant by the Investigator.
11. Assess at Day 30 post surgery via clinic visit, medical record, or phone call. Any RRT that is provided prophylactically during surgery while the subject is on a bypass machine does not meet this endpoint.
12. # Eligible patients will be randomized up to 7 days before or on the day of surgery, but prior to the start of surgery (i.e., induction of anesthesia).
13. * The adverse event, serious adverse event and transfusion reaction collection period will begin at the start of surgery or the first study RBC transfusion, whichever is first.
14. ^ The adverse event, serious adverse event and transfusion reaction collection period will begin at the start of surgery or the first study RBC transfusion, whichever is first. AEs including protocol specified AEs, TRs and SAEs (see protocol Section 7.3) are collected through 28 days after the last study transfusion; also collected daily during acute transfusion support period.

5. DEFINITIONS

5.1 Time-Related Terms

5.1.1 Baseline Visit

The *Baseline visit* is on the day before or on the day of study cardiac surgery prior to start of surgery (whichever comes last).

5.1.2 Study Day

The *study day* describes the relative day of an observation starting with the first study transfusion date designated as Study day 1. It will be calculated as:

- For days prior to the first study transfusion date, Study Day = Date – Date of first study Transfusion
- For days on/after the first study transfusion date, Study Day = Date – Date of first study Transfusion + 1

5.1.3 RBC Exposure

The *extent of exposure* to RBC transfusions will be assessed by units and volume of treatment exposure, derived as:

- Units of treatment exposure = RBC Units transfused from surgery till both 48 hours post-surgery and ATSP
- Volume of treatment exposure = RBC volume transfused from surgery till both 48 hours post-surgery and ATSP
- For the purposes of the statistical analysis plan, ≥4 Total RBC units related to an 8-hour surgery and ≥10 units of RBCs within 24 hours represents larger volume or massive transfusion, respectively.

5.2 Efficacy Related Definitions

5.2.1 Baseline Score

The *baseline score* is the last measurement taken before the first study RBC transfusion and before the start of the study cardiac surgery.

5.2.2 Change and Percent Change from Baseline

The change and the percent change from baseline in a measure at a post-baseline visit will be derived as:

- Change = (Score at the Post-Baseline Visit) – (Baseline Score).

If there are multiple measurements at a post-baseline visit, the maximum value will be used.

For the analysis of primary efficacy endpoint, maximum serum creatinine within 48 ±4 hours post-surgery will be used.

5.3 Other Definitions

5.3.1 Prior and Concomitant Medications

Medications will be categorized into prior medications and concomitant medications which coded using WHO Drug Global B3, March 2018. Specifically, prior medication is defined as any medication taken and ended prior to the study RBC start date. Concomitant medication is defined as any medication taken concurrently while receiving study RBC, i.e., the treatment period (period of time from first unit to last unit) of a concomitant medication taken by a subject must overlap with the treatment period.

5.3.2 Non-Study RBC

In this study, non-study RBC (conventional RBC) may be transfused according to the protocol to provide the patient with an appropriate and necessary treatment when study RBCs are unavailable or patient's need for RBC transfusions exceeds the quantity of study RBCs in inventory at the hospital blood bank (e.g., during a Massive Transfusion Protocol). Unlike Test and Control RBCs, conventional RBCs may be sourced from any blood collection center and are not manufactured using study RBC production protocols. The use of cell saver blood is not included as part of non-study RBCs.

The non-study RBC use will be summarized for Test and Control in terms of units and proportions for both within 48 hours and whole ATSP.

6. STUDY POPULATION

6.1 All Enrolled Patients Set (ALL)

The ALL set includes all subjects who signed informed consent and were randomized.

6.2 Modified Intent-to-Treat (mITT) Population

The mITT includes all randomized subjects who received at least one study transfusion. Efficacy analyses will be performed using the mITT and summarized by assigned treatment as randomized. Safety analyses will be performed using the mITT and summarized by actual treatment received.

6.3 Per-Protocol Set (PPS)

The PPS is a subset of the mITT. It will be the analysis population for exploratory and sensitivity analyses and will be summarized using treatment as randomized. Any subject affected by an important protocol deviation that might affect the primary efficacy analysis will be excluded from the PPS. The PPS will be identified before the unblinding of treatment assignments.

Before the database lock and before unblinding the treatment assignments, Cerus' study team will review all protocol deviations, identify subjects with any protocol deviation that could impact the efficacy outcome, and determine whether or not to exclude the subject from the PPS.

The following protocol deviations may be considered as important and will lead to an exclusion of subjects form the PPS:

- Randomized to Test but received Control treatment within 48 hours of the end of surgery.
- Randomized to Control but received Test treatment within 48 hours of the end of surgery.
- Subjects who received non study RBC transfusions within 48 hours completion of surgery due to a blood bank error when appropriate study RBC components were available.
- Subjects who did not receive any study RBCs within 48 hours of the end of surgery.
- Subjects who received a study RBC transfusion but have no assessment of sCr concentrations after the transfusion and within 48 ±4 hours of the end of surgery.

Additional protocol deviations may be considered important at the blinded data review meeting and will be documented appropriately. If a subject is randomized incorrectly or is administered the incorrect study treatment, analyses of the efficacy endpoints will be based on the assigned treatment, whereas all other analyses will be based on the actual treatment received.

Note that the study protocol allows for the transfusion of non-study RBCs when study RBCs are unavailable or the patient's need for RBC transfusions exceeds the quantity of study RBCs in

inventory at the hospital blood bank (e.g., during a Massive Transfusion Protocol). These non-study RBC components are therefore considered to be transfused per protocol.

6.4 Study RBC Only Set (SROS)

The SROS is a subset of the PPS. Subjects who received non study RBC within 48 hours of the completion of study surgery will be excluded from the SROS. It will be the analysis population for the exploratory analysis with sensitivity analyses and will be summarized using treatment as randomized.

7. GENERAL CONSIDERATIONS

All measures will be summarized by treatment (planned or actual received) descriptively. Continuous variables will be summarized using descriptive statistics such as number of observations (n), mean, standard deviation, median, minimum, and maximum. Categorical variables will be tabulated using frequency (n) and percent (%).

The statistical testing will be conducted at a significance level of 0.05 (two-sided) and the 95% confidence interval will be shown, unless specified otherwise. No statistical testing will be conducted for safety measures.

All data manipulations, descriptive summaries, and statistical hypothesis testing will be performed using Statistical Analysis System (SAS) Version 9.4 or later.

7.1 Handling of Missing Data

7.1.1 Efficacy Measure

For the primary analysis of serum creatinine (sCr) increase within 48 hours of surgery start, no imputation is needed for the analysis on observed cases using the Miettinen and Nurminen (M&N). In this case, the maximum observed post-baseline sCr value will be used to calculate the increase from baseline. If a subject has no post-baseline sCr within 48 ±4 hours post-surgery at all, this subject will be treated as missing the primary endpoint. These missing values will be imputed via multiple imputation in the sensitivity analysis with logistic regression approach.

In the sensitivity analysis with Pattern-Mixture Model (PMM) approach, for subjects who received non-study RBC within 48 hours post-procedure, any sCr values collected after the transfusion of non-study RBC will be censored (i.e., treated as missing). Missing sCr data will be imputed using the Multiple Imputations (MI) approach.

Missing secondary efficacy endpoints will not be imputed.

7.1.2 Safety Measures

Descriptive summaries of safety measures will be based on observed data only. No imputation of missing scores will be implemented.

7.1.3 Dates for Medical Events and Medications

Completely or partially missing onset and resolution dates of medical history (MH), AE, and concomitant medications (CM) will be imputed as follows:

Table 2: Handling of Missing Data for Medical Events and Medications

Date	Type of Missing Date	Handling of Missing Date
Event onset date (e.g., YYYY-MM-DD)	Completely missing	No imputation will be applied: For AE, the event will be considered treatment emergent. For CM, the event will be considered concomitant. For MH, the event will be considered to occur prior to Inform Consent date.
	Only YYYY is available	Use the first day of YYYY (Jan. 1st) to impute the missing month and date parts of the onset date.
	YYYY and MM are available, but DD is missing	Use the first day of MM (1st) to impute the missing date part of the onset date.
Event resolution date (e.g., YYYY-MM-DD)	Completely missing	No imputation will be applied. The event will be considered ongoing (i.e., not resolved) at the last visit date.
	Only YYYY is available	Use the last day of YYYY to impute the missing month and date parts of the resolution date.
	YYYY and MM are available, but DD is missing	Use the last day of MM to impute the missing date part of the resolution date.

7.2 Multi-Center Studies

This is a multi-center study enrolling subjects from approximately 15-20 U.S sites. The number of subjects per site might be small. Separate analyses by site are not meaningful and not desirable. Therefore, there are no analyses adjusting for sites. All subjects from all sites will be pooled for statistical analysis.

7.3 Multiple Comparisons / Multiplicity

No multiplicity adjustment is needed for this study. The superiority of the primary efficacy endpoint will be tested only after the non-inferiority is met.

7.4 Interim Analysis

A blinded interim analysis was performed to re-estimate the sample size in October 2021 which resulted in the reduction in sample size from 600 to ≥ 292. This blinded interim analysis was discussed with the FDA on October 6, 2021, in Q-Sub BQ150260/6.

No other interim analysis is planned for this study.

8. DATA SAFETY AND MONITORING BOARD

The study data and safety will be monitored on a regular basis by the Data and Safety Monitoring Board (DSMB) to ensure patient safety and review protocol compliance. The DSMB will review group-blinded data generated by an independent statistician. All DSMB members are independent of the sponsor.

9. SUMMARY OF STUDY POPULATION DATA

9.1 Subject Disposition

The disposition of ALL (i.e., all randomized subjects) will be summarized by treatment and overall. The summary will include the number and percentage of ALL subjects in the mITT, PPS and SROS. The disposition summary will also include the number and percentage of completers and non-completers, as well as the number and percentage of discontinuations of the study and of the study RBC by the primary discontinuation reason.

9.2 Demographics and Baseline Characteristics

Subject demographics and baseline characteristics will be descriptively summarized for the mITT population by planned treatment and overall. Specifically, for subject demographics, the following variables will be summarized:

- Age at randomization (continuous and categorical: < 65 years or ≥ 65 years)
- Sex (categorical: Male or Female)
- Ethnicity (categorical: Hispanic/Latino or Not)
- Race (categorical: American Indian or Alaska Native, Asian, Black, Native Hawaiian or Pacific Islander, White, or Other)

For baseline characteristics, the following variables will be summarized:

- Trust Score
- Serum Creatinine (continuous and categorical: < 1.2 or ≥ 1.2 mg/dL)
- Blood Type (A, B, O and AB)
- RH factor (Positive or Negative)

9.3 Medical, Surgical and Transfusion History

For this study, medical history and adverse events will be coded using MedDRA 21.1, 2018. Each medical event will be classified into a SOC and mapped to a Preferred Term (PT).

The medical and surgical history will be summarized for the mITT. Subjects reporting any medical and surgical history at baseline will be tabulated by SOC and PT for each planned treatment and overall.

9.4 Protocol Deviations

In this study, protocol deviations are categorized as follows:

- Informed Consent
- Inclusion/Exclusion Criteria and Concomitant Treatment
- Study Procedures/Treatment, including Study Tests/Assessments/ Laboratory
- Visit Scheduling
- Withdrawal Criteria
- Other

A protocol deviation is considered important if it may affect the study primary endpoints and the subject's rights, safety, or well-being, and/or the completeness, accuracy, and reliability of the study data. Cerus' study team will review all protocol deviations and determine the list of important protocol deviations prior to database lock. All randomized subjects with any important protocol deviation(s) will be tabulated by deviation category for each planned treatment and overall. In addition, protocol deviations will also be listed.

9.5 Prior and Concomitant Medications

For this study, prior and concomitant medications will be coded using WHO Drug Global B3 (March 2018). Each medication will be classified using the Anatomical-Therapeutic-Chemical (ATC) classification system and mapped to a preferred drug name, summarized for the mITT population.

Subjects taking any prior medications will be tabulated by ATC level 3, level 4, and preferred drug name for each actual treatment received and overall. A subject will be counted at most once for each prior medication, even if the subject took the same prior medication on multiple occasions. Subjects taking any concomitant medications will be tabulated similarly. In addition, prior medications and concomitant medications will also be listed, separately.

9.6 Surgical and Clinical Characteristics

The following surgical related parameters will be summarized for the mITT population using descriptive statistics:

- Duration of study cardiac surgery
- Time on pump

- Time to discharge
- Volume of cell saver blood
- Use of cell saver blood, platelets, plasma and cryoprecipitate in frequency and percentages

9.7 Exposure to Study RBC and Other Blood Product

The extent of exposure to a study RBC is measured by the number and volume of study components as derived in Section 5.1.3. For subjects in the mITT Population, the units of exposure will be summarized using descriptive statistics, and frequency and percentage of subjects will be tabulated by number of study RBC components and total study plus non-study RBC components (1, 2, 3, 4, 5, 6, 7, 8, 9, or ≥ 10 units) for subject. Descriptions will be provided for both within the first 48 hours of post-surgery and the entire ASTP period.

10. EFFICACY ANALYSES

The primary efficacy analyses will be performed on the mITT population, with subjects classified by planned treatment, irrespective of the actual treatment received.

It may be necessary to complete additional exploratory analyses after the planned analyses are completed. Full details of additional analyses will be presented in the CSR, and any such analyses will be clearly identified as post-hoc.

10.1 Analyses of Primary Endpoint

10.1.1 Primary Analyses

The primary efficacy endpoint is the proportion of patients with a diagnosis of renal impairment defined as any raised sCr level, occurring after transfusion of a study RBC, of ≥ 0.3 mg/dL (or 26.5 µmol/L) from the pre-surgery baseline within 48±4 hours of the end of surgery.

The corresponding hypothesis is:

$$H_0: \; P_{Test} - P_{Control} \geq 50\% \times \hat{P}_{Control}$$

versus

$$H_A: \; P_{Test} - P_{Control} < 50\% \times \hat{P}_{Control},$$

where P_{Test} and $P_{Control}$ are the event rates for Test (INTERCEPT) and Control groups, respectively, and $\hat{P}_{Control}$ is the observed Control rate.

The primary analysis of the primary efficacy endpoint will be based on Miettinen and Nurminen (M&N) method stratified by baseline sCr (sCr ≥1.2 mg/dL vs. < 1.2 mg/dL) and cardiac surgery group performed (more at risk for renal complications vs. less at risk) based on the observed cases.

The maximum observed post-baseline sCr value will be used to calculate the increase from baseline. If subjects have no post-baseline sCr within 48 hours post-surgery at all, this subject will be treated as missing the primary endpoint.

The common risk difference in proportions and the Mantel-Haenszel score 95% CI for the treatment difference (Test – Control) will be calculated using the stratified M&N method adjusting for baseline sCr and cardiac surgery group type. The estimate of the common risk difference will be calculated based on the stratum risk differences across all strata (Agresti, 2002; p. 231). Non-inferiority will be claimed if the upper bound of the 2-sided M&N Score 95% CI for the treatment difference (Test – Control) is less than 50% of the observed Control rate. Further, if the upper bound is less than 0, then superiority is achieved.

10.1.2 Sensitivity Analyses

To assess the robustness of the results from the primary analysis of the primary efficacy endpoint, two sensitivity analyses will be performed (Table 3). For subjects who received non-study RBC within 48 hours post-procedure, any sCr values collected after the transfusion of non-study RBC will be censored (i.e., treated as missing) in the sensitivity analysis with PMM approach.

Table 3: Overview of Primary and Sensitivity Analysis Methods

Primary or Sensitivity	Statistical Method	Analysis Population	Handling of Missing Data
Primary Analysis	Miettinen and Nurminen	mITT	Observed cases
Sensitivity Analyses	Logistic Regression	mITT	Multiple Imputation (MI)
	Pattern Mixture Modeling (PMM)	mITT	Multiple Imputation (MI)

While the Logistic regression with MI is under missing at random (MAR) assumption, a second sensitivity analysis is to assess the sensitivity to departure from MAR assumption. Specifically, a Pattern-Mixture Model (PMM) with delta-adjustment of imputations based on missing not at random (MNAR) assumption will be implemented as follows:

a. Use MCMC to create monotone missingness first. Then obtain standard multiple imputations under MAR assumptions for missing sCr values using monotone regression. The missing data are filled in and 1000 complete data sets are created.

b. For each sCr value that had been missing because of censoring after having received non-study RBC within 48 hours post-procedure, regardless of treatment group, adjust these values by adding λ to the imputed values, where λ = 0.1 mg/dL.

c. Each of the 1000 complete data sets will then be analyzed separately using the same M&N method as used for the primary analysis.

d. The estimates obtained from the M&N method of each complete data set are combined for inference purposes.

e. Repeat step b, c, d, with λ = 0.2, 0.3, ..0.5 mg/dL or until the conclusion is reversed from the primary analysis, whichever comes first. The larger the value of λ that is required to reverse the conclusion of the primary analysis, the more robust the conclusion is considered to be.

10.1.3 Exploratory Analyses

To correct for the impact due to switching from study RBCs to non-study RBCs in patients who receive non-study RBCs, an inverse probability censoring weighting (IPCW) method will be used. In this approach, in order to derive weights adjusting for the confounding effects due to use of non-study RBCs, both baseline and post baseline covariates will be considered including, but not restricted to, age (< 65, ≥ 65), race (white, non-white), gender, serum creatinine, previous MI, surgery risk type, surgery duration, blood loss, cell saver use.

The final criteria for selected covariates would need to statistically have a p-value of less than or equal to 0.1 in the multivariate logistic regression models for weight calculations.

Exploratory analysis of the primary efficacy endpoint will also be conducted on PPS and SROS on observed cases. Additionally, primary efficacy endpoint will also be explored between Test and Control on SROS plus subjects with 1, 2, 3 and ≥ 4 non-study RBCs, sequentially. For the purposes of the statistical analysis plan, ≥4 RBC units related to an 8-hour surgery and ≥10 units of RBCs within 24 hours represents larger volume or massive transfusion, respectively.

Table 4: Overview of Exploratory Analyses

Statistical Method	Analysis Population
IPCW	mITT
Miettinen and Nurminen	PPS
Miettinen and Nurminen	SROS
MMRM	mITT
Miettinen and Nurminen	ALL

A mixed model repeated measures (MMRM) of the change from baseline in continuous sCr data will also be explored. This model will include fixed effects for treatment, visit, baseline sCr, surgery type preformed and treatment*visit interaction assuming an unstructured covariance matrix.

ALL subjects between mITT and non-transfused subjects who were randomized but not transfused any study RBC will also be compared with Miettinen and Nurminen approach.

Additional exploratory analysis may be conducted as suggested by the data including a Bayesian approach.. Any P values presented from exploratory efficacy analyses will be interpreted in an exploratory sense only.

10.2 Analyses of Secondary Efficacy Endpoints

Secondary efficacy outcome measures include:

- The proportion of patients with a diagnosis of stage I, II or III Acute Kidney Injury (KDIGO 2012) based on changes in sCr levels from pre-surgery baseline and the need for renal replacement therapy (RRT) post-surgery.
- Mortality or the need for RRT by 30 days post-surgery

CMH test will be used for assessment of treatment differences for the secondary efficacy endpoints. No formal statistical inferences will be drawn from secondary efficacy analyses. Any P values presented from secondary efficacy endpoints will be interpreted in an exploratory sense only.

10.3 Subgroup Analyses

To assess the homogeneity of treatment effects among subgroups, subgroup analyses with descriptive summaries will be performed by age groups (< 65 vs ≥ 65 years), sex (males or females), race (White or Non-white), baseline sCr (sCr ≥1.2 mg/dL vs. < 1.2 mg/dL), baseline TRUST score (< 5 vs ≥ 5) and cardiac surgery group performed (more at risk for renal complications vs. less at risk) will be reported for sCr and primary efficacy endpoint.

Other subgroup analyses may be performed as suggested by the data.

11. SAFETY ANALYSES

The mITT population will be used for all safety summaries, and subjects will be classified by actual treatment received. Descriptive summary of each safety-related measure will be performed. In the event that a subject received both Test and Control RBC during the study, the subject will be summarized with those receiving Test.

The primary safety endpoints (Related TEAEs and TE antibodies specificity to INTERCEPT RBCs) will be compared and tabulated by treatment using frequency (n) and percent (%). P values from a Cochran-Mantel-Haenszel (CMH) test stratified by baseline sCr (sCr ≥1.2 mg/dL vs. < 1.2 mg/dL) and cardiac surgery group performed (more at risk for renal complications vs. less at risk) will be presented in an exploratory sense only.

Other safety endpoint including alloantigens and HLA alloantigens will be carried out in the same manner.

Subjects with any AE(s), SAE(s), TRs and AEs leading death will be tabulated by type of AE(s) for each actual treatment received and overall.

Besides the overall summary, subjects with any AE(s), any SAE(s) or TR(s) will be tabulated by SOC, preferred term, severity, and relatedness. A subject who experienced multiple events within a SOC or preferred term will be counted only once for that SOC or preferred term.

AEs, SAEs, TRs, AEs leading to death, AEs leading to study transfusion discontinuation, AEs leading to death, if any, will be listed separately.

12. CHANGES TO PROTOCOL AND/OR SPECIFIED ANALYSES

The following changes to the data analysis pre-specified in the protocol are made after the study started.

1. Use of M&N Score 95% CI for treatment difference of the primary efficacy endpoint stratified by baseline sCr category and cardiac surgery group.

   Protocol states: CMH

   Justification: Stratified M&N Score 95% CIs will be used to control for baseline sCr category and cardiac surgery group. It has a higher coverage probability of the true risk difference for situations with a mixture of both small and large strata sample sizes, unbalanced treatment allocation or rare events (Klinhenberg, 2014).

2. Add an exploratory analysis using IPCW approach.

   Justification: Per FDA's request to provide a more sophisticated statistical adjustment to mitigate the bias from the use of non-study RBCs.

3. Add a PMM (Pattern Mixture Modeling) sensitivity analysis.

   Protocol states: As a sensitivity analysis, logistic regression will be utilized.

   Justification: PMM will be used to assess the sensitivity to departure from MAR assumption.

4. Add an exploratory analysis using Bayesian approach.

   Justification: to provide an alternative approach to aid the interpretation of the study results.

13. REFERENCES

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