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Efficacy and safety of the blood-based cardioplegia solution Huaxi-1 in cardiopulmonary bypass surgery: protocol for a multicentre randomised controlled trial

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ABSTRACT

Introduction Cardioplegia during cardiopulmonary bypass is essential for ensuring a surgical field free of blood and cardiac movement. Numerous cardioplegia solutions are available, but consensus guidelines about the safest or most effective do not exist. The present trial will compare the Huaxi-1 cardioplegia solution, which has been used since 2006 with good results at a major Chinese cardiac centre not involved in this trial, with the widely used Custodiol histidine-tryptophan-ketoglutarate (HTK) solution in terms of safety and efficacy at inducing cardiac arrest and protecting the myocardium during bypass.

Methods and analysis A total of 160 adult patients undergoing elective cardiac surgery requiring cardiopulmonary bypass and cardioplegic arrest will be recruited at four medical centres in China. Recruitment is planned to begin on 1 November 2024, and is expected to conclude by 31 October 2025. Eligible patients will be randomly allocated 1:1 to receive either Huaxi-1 or HTK cardioplegia solution. The primary endpoint is the peak level of high-sensitivity cardiac troponin T (hs-cTnT) within 48 hours after surgery between the two groups. The secondary endpoints include levels of myocardial injury markers such as the creatine kinase-myocardial band (CK-MB) and cardiac troponin I at baseline and at 6, 12, 24 and 48 hours after surgery. The two groups will also be compared in terms of how left ventricular ejection fraction changes from baseline and in terms of the rate of spontaneous cardiac recovery. Data will be analysed using SAS V.9.4.

Ethics and dissemination This trial has been approved by the ethics committees at Guangdong Provincial People's Hospital (lead site) and the three other study sites. The results of the study will be published in peer-reviewed journals and presented at international conferences.

Trial registration number ChiCTR2400089689 (www.chictr.org.cn).

STRENGTHS AND LIMITATIONS OF THIS STUDY

⇒ The trial involves four major cardiac centres, which may ensure the robustness of the findings to variability in patients and surgical practices.
⇒ The study sites do not include the medical centre where Huaxi-1 was developed and incorporated into routine practice, which may increase the objectivity of assessments.
⇒ The trial comprehensively assesses myocardial protection by assaying multiple biomarkers of myocardial injury and by determining left ventricular ejection fraction.
⇒ Blinding to an investigator is not feasible in the trial, which may introduce performance bias.

INTRODUCTION

During cardiopulmonary bypass (CPB), cardioplegia is essential for ensuring a surgical field free of blood and cardiac motion as well as for protecting myocardial function from injury due to the global ischaemia during aortic cross-clamping and subsequent reperfusion.[1 2] Cardioplegia is designed to achieve these goals by maintaining electrolyte balance to prevent myocardial oedema and by providing energy substrates for critical metabolic processes while slowing overall metabolism.

Since the development of K^+-rich cardioplegia solutions in the 1950s, more than 160 different formulations have been developed and implemented in clinical practice worldwide.[3] Despite this variety, the choice of cardioplegia solution is driven by the preferences of the surgeon or medical centre rather than consensus guidelines.[4] Cardioplegia solutions currently used in clinical practice

are generally categorised into crystalloid-based solutions and blood-based solutions that are supplemented with various electrolytes.

Traditional crystalloid solutions, such as the St. Thomas formulation,[5] use elevated potassium levels to depolarise myocardial membranes, thereby inducing diastolic arrest. In contrast, histidine-tryptophan-ketoglutarate (HTK),[6] while also a crystalloid solution, employs low sodium concentrations to hyperpolarise the myocardial membrane. This hyperpolarisation reduces cellular activity and enhances the tolerance of cardiac tissue to ischaemia.

In contrast, so-called 'blood-based' cardioplegia solutions combine the electrolytes in crystalloid solutions with the oxygen and metabolic substrates in human blood in order to ensure adequate aerobic support while providing protection against injury. One of the widely used examples is del Nido cardioplegia solution, which was developed for paediatric surgeries and is now used in adults.[7] Since 2006, West China Hospital of Sichuan University, one of the largest cardiac centres in China, has routinely used a blood-based cardioplegia solution known as 'Huaxi-1', developed by its clinicians. Huaxi-1 consists of a 4:1 mixture of autologous blood and crystalloid solution, prioritising oxygen delivery and metabolic support, whereas del Nido employs a 1:4 ratio, optimised for prolonged myocardial arrest with minimal metabolic demand. Huaxi-1 contains 21 mmol/L K^+ to induce cardiac arrest as well as haemoglobin (70–80g/L) to provide oxygen; glucose, insulin and magnesium sulfate to promote glycogen and ATP synthesis; bicarbonate (31.1 mmol/L) to regulate pH; lidocaine (0.35 mmol/L) to block sodium channels and thereby protect the myocardium; and mannitol to reduce oxidative stress and myocardial oedema.

We previously showed that Huaxi-1 was superior to HTK in promoting energy regeneration and reducing ischaemia-reperfusion injury in animals subjected to myocardial ischaemia for 90 min.[8] Clinical experience at West China Hospital suggests that a single Huaxi-1 infusion can provide effective cardiac arrest for at least 40 min. The compositions of Huaxi-1 and HTK solutions are compared in detail to highlight their differences (table 1). The cardioplegia formulation presents several advantages over many other formulations because it (1) it mimics the cytosolic environment and is absorbed into the bloodstream without disrupting electrolyte balance or causing blood dilution, due to its low crystalloid content; (2) it is suitable for both antegrade and retrograde perfusion; and (3) its slight hyperosmolarity relative to blood helps reduce the risk of myocardial oedema.

Currently, no blood-based cardioplegia solution has been approved by the National Medical Products Administration in China, necessitating that hospitals prepare these solutions in-house. This practice introduces variability and increases the risk of unpredictable clinical outcomes.

Table 1 Comparison of ion and molecule concentrations in HTK and Huaxi-1 solutions

Ion or molecule	Huaxi-1 solution (mmol/L)*	HTK solution (mmol/L)
K^+	20.77 (4.5)	9
Mg^{2+}	6.88 (0.9)	4
Na^+	146 (140)	15
Ca^{2+}	0.25 (1.24)	0.02
HCO_3–	31.1 (25)	0
Cl-	120.94 (102)	100
SO_4^{2}–	7.68 (2)	2
Mannitol	13.17	29.97
Glucose	6.52 (5)	0
Lidocaine	0.35	0
Histidine	0	198
Tryptophan	0	2
Ketoglutarate	0	1

*The concentrations of ions or molecules in the blood component of this solution are indicated in parentheses.
HTK, histidine-tryptophan-ketoglutarate.

In light of these clinical successes and the potential benefits of blood-based cardioplegia solutions, Chengdu Qingshan Likang Pharmaceuticals (Chengdu, China) has optimised and standardised the Huaxi-1 formulation and wishes to assess its safety and efficacy in a multicentre trial, with an eye toward regulatory approval as a commercial product for clinical use. The trial will randomise patients to receive either Huaxi-1 or HTK in order to benchmark the new product against an industry standard. Such benchmarking may help clinicians select the most appropriate cardioplegia solution for their patients, though comprehensive studies on the topic are still lacking in the literature.

METHODS AND ANALYSIS

The trial protocol has been reported in accordance with the Standard Protocol Items: Recommendations for Interventional Trials guidelines[9] (online supplemental appendix 1).

Study design

This randomised, non-inferiority trial with a parallel active control arm will involve patients from four cardiac centres in China and will be conducted in compliance with the 'Regulations on the Supervision and Administration of Medical Devices'[10] and 'Good Clinical Practice for Medical Device Clinical Trials'.[11] The objective of the trial is to assess the safety and efficacy of Huaxi-1 in inducing cardiac arrest and providing myocardial protection during surgery with CPB. The trial aims to demonstrate that the clinical performance of Huaxi-1 is not inferior

to a widely used approved product, HTK. Any differences in performance are expected to fall within clinically acceptable limits and therefore the trial will have a non-inferiority design.

The control arm in this study will receive HTK rather than, for example, the del Nido formulation because HTK has been approved by the National Medical Products Administration in China and is the most frequent control formulation in controlled trials registered in the US database ClinicalTrials.gov. Even though the del Nido formulation, like Huaxi-1, contains mannitol, magnesium sulfate, bicarbonate buffer and lidocaine, it is not commercially available as a standardised formulation and has not been tested extensively in adults.

The conduct of the trial will be monitored regularly by suitably qualified external experts (online supplemental appendix 2) to ensure strict adherence to the study protocol at all four sites. To ensure consistency and minimise inter-laboratory variability, uniform assays are used across all participating hospitals and all samples are analysed centrally.

Patient and public involvement

None.

Study setting

The four study sites are the Guangdong Provincial People's Hospital, The Second Xiangya Hospital of Central South University, The First Affiliated Hospital of Air Force Medical University and the First Affiliated Hospital of Nanjing Medical University. Each of these sites is a comprehensive tertiary medical centre with over 5000 beds and an annual outpatient volume exceeding 1 million. At each site, more than 1000 surgeries involving CPB are performed annually.

Study participants

Patients at the four study sites will be recruited starting from 1 November 2024, if they have been scheduled for elective open-heart surgery that requires CPB and cardioplegic arrest and that may or may not include thoracoscopic procedures. Other enrolment criteria (table 2) include an age of at least 18 years, ability to understand the trial's purpose and willingness to sign an informed consent form (online supplemental appendix 3).

Patients will not be enrolled if they present one of the following (table 3): need to undergo emergency surgery; prior cardiac surgery; EuroSCORE II≥10%; severe dysfunction of the liver or kidneys; acute myocardial infarction within 2 weeks, psychiatric disorders, substance abuse, myocarditis, cardiogenic shock or clinically significant left ventricular dysfunction, defined as a left ventricular ejection fraction (LVEF)<35%; known allergies to trial components or required drugs; presence of an implanted cardiac pacemaker or defibrillator; preoperative need for mechanical circulatory support; pregnancy; current participation in another trial; or any other condition that investigators deem incompatible with participation.

Subjects who have been enrolled in the trial but later withdraw at any time or are lost to follow-up will be treated as dropouts, as will subjects whom investigators exit from the study because of lack of compliance with the trial protocol or because of serious adverse events or complications. The reasons for all dropouts will be documented and, if appropriate and feasible, investigators should make every effort to contact dropouts and complete as many of the remaining assessments as possible. Investigators should also implement alternative arrangements for treatment.

Efforts will be made to ensure similar levels of participant enrolment across all four study sites. In any event, enrolment from any single centre should not exceed 50% of the total sample.

Randomisation and blinding

Randomisation will be conducted using a central randomisation system via the Randomizer software (V.1.0, 2022, Randomizer.org) by a colleague who is not involved in the study. This trial will not employ stratification by study site. Block randomisation will be used to ensure balanced allocation between groups. Patients are blinded to the treatment allocation. However, due to the marked differences in the appearance and administration techniques of the two solutions, it is not feasible to blind the investigators.

Outcomes

Primary and secondary endpoints

The trial will focus on the following outcomes of efficacy and safety, developed based on previous trials of cardioplegia solutions.[12–15] The primary endpoint is a peak level of high-sensitivity cardiac troponin T (hs-cTnT) in serum within 48 hours after surgery. Secondary endpoints include the levels of the creatine kinase-myocardial band (CK-MB) and of cardiac troponin I (cTnI) in serum before surgery (after anaesthesia induction), immediately after the end of CPB and at 6, 12, 24 and 48 hours after surgery. Levels of CK-MB, cTnI and hs-cTnT in serum are

Table 2 Inclusion criteria for enrolment in the trial

Criterion	Description or comment
Age	At least 18 years old, no restriction on gender
Scheduled for elective open-heart surgery	Can include thoracoscopic cardiac surgery
Cardiopulmonary bypass	Required with induced cardiac arrest
Informed consent	Voluntarily agrees to participate and signs consent form

Table 3 Criteria for exclusion from the trial

Criterion	Description or comment
Emergency surgery or heart transplantation	
History of previous cardiac surgery	
EuroSCORE II	≥10%
Severe organ dysfunction	Dysfunction of the liver or kidney, defined as a ratio of aspartate aminotransferase/alanine aminotransferase >3×upper limit of normal, serum creatinine >3×upper limit of normal or need for dialysis
Recent myocardial event	Acute myocardial infarction, myocarditis or infective endocarditis within the previous 2 weeks
Left ventricular dysfunction	Left ventricular ejection fraction <35%
Cardiogenic shock	Systolic blood pressure <90 mm Hg despite inotropic support
Allergies	Known or suspected allergy to components of the cardioplegia solutions or required drugs (eg, lidocaine, insulin)
Implanted device	Cardiac pacemaker or defibrillator
Mechanical circulatory support	Preoperative requirement for intra-aortic balloon pump or extracorporeal membrane oxygenation
Pregnancy and breastfeeding	
Severe psychiatric disorder	Disorders that interfere with the person's ability to provide informed consent or to adhere to study protocol
Substance abuse	History of alcohol or drug misuse
Concurrent clinical trials	Concurrent participation in other trials
Investigator judgement	Assessment of an individual as unsuitable for the trial

well-established indicators of perioperative myocardial injury.[16] CK-MB is quite sensitive but less specific, so cTnI and hs-cTnT will also be assayed. The levels of both cardiac troponins begin to rise 4–8 hours after myocardial injury and peak around 12–24 hours,[17] while elevated levels of hs-cTnT beyond 24 hours are associated with postoperative complications.[18] Secondary endpoints include the change in LVEF from baseline (post-anaesthesia induction and pre-surgery) to immediately after CPB, as well as the rate of spontaneous cardiac recovery, defined as the proportion of participants exhibiting restoration of spontaneous cardiac rhythm. Spontaneous cardiac recovery is characterised by the restoration of autonomous cardiac rhythm following the release of aortic cross-clamping, without the necessity for interventions such as electrical defibrillation. The LVEF will be measured using transoesophageal echocardiography (TEE) by measuring the internal diameter of the left ventricle at the level of the papillary muscle in the short-axis view during end-systole and end-diastole, then converting these measurements to volumes using the Teichholz formula[19] and finally to the ejection fraction as described.[20]

Other secondary endpoints include the rate of all-cause mortality up to discharge or postoperative day 30, and the re-operation rate, defined as the percentage of participants requiring a second or subsequent open-chest or thoracoscopic surgery for any reason during follow-up. Additionally, the need for an intra-aortic balloon pump (IABP) or extracorporeal membrane oxygenation (ECMO) will be monitored, along with the time points of intensive care unit (ICU) admission and discharge. Lastly, the duration of the initial ICU stay will be evaluated.

Safety monitoring

Adverse events from enrolment through the end of follow-up will be recorded and classified according to severity. Adverse events will be considered 'treatment-emergent' if they occur after the first administration of cardioplegia solution. The analysis will focus on these adverse events, especially if they are severe and/or lead to an exit from the study. The following adverse events related to CPB will also be recorded: low cardiac output syndrome, new-onset myocardial infarction, malignant arrhythmia, acute kidney injury and new-onset cerebral infarction (online supplemental appendix 4). Malignant arrhythmias will be recorded both post-cross clamp removal while still on CPB and post-separation from CPB, but they will be reported and analysed separately. All study sites will report and classify adverse events consistently, following a standardised scheme that applies throughout all phases of the trial, from screening to discharge or 30 days post-surgery.

Group allocation and minimal sample size

Participants will be randomly assigned in 1:1 to undergo cardioplegia with the Huaxi-1 or HTK formulation. Huaxi-1 will be defined as equivalent to HTK if the difference in levels of myocardial injury markers between the

two trial arms does not exceed 50%. This threshold is supported by prior evidence showing that differences below 50% in biomarker levels between groups are not associated with clinically significant adverse outcomes.[21–23] Given this margin of non-inferiority and assuming a coefficient of variation of 0.95 in assayed marker levels,[24] we estimate that at least 126 trial participants will be required to provide a statistical power of 80% at a one-sided significance level of 0.025, based on the module 'Non-inferiority tests for the ratio of two means (log-normal data)' in PASS 2021 software (NCSS, Kaysville, Utah, USA). This minimal size will be increased to 158 to counteract a dropout rate of 20% and increased again to 160 to accommodate block randomisation. For missing data on biomarkers, sensitivity analyses will employ various imputation methods, such as mean imputation and regression imputation, to validate the robustness of the trial results.

Interventions and assessments

The trial will comprise nine assessments (table 4). During the first assessment, potential participants will be fully assessed for enrolment and informed about the purposes of the trial. If they sign the informed consent form, their medical history, including age, sex, height, weight and ethnicity, along with demographic characteristics such as history of major diseases, surgeries, allergies and lifestyle

Table 4 Procedures and timing of assessments during the trial

Procedure	Visit 1	Visit 2	Visit 3	Visit 4	Visit 5,6,7	Visit 8	Visit 9	Early withdrawal
Phase of trial	Screening	Baseline	Intraoperative	30 min after CPB	6 hours, 12 hours, 24 hours after surgery	48 hours after surgery	Discharge or 30 days after surgery	
Informed consent	X							
Medical history and demographics	X							
NYHA classification	X							
EuroSCORE II	X							
Vital signs	X	X	X	X	X	X	X	X
ECG	X							
Laboratory tests	X					X	X	X
Pregnancy testing	X							
TTE	X							
TEE		X		X				
Check of eligibility	X	X						
Randomisation		X						
Markers of myocardial injury		X		X	X	X		X
Surgical data			X					
Duration of CPB			X					
Duration of aortic cross-clamp			X					
Analysis of arterial blood gases		X	X	X				
Time point of ICU transfer in/out					X	X	X	X
IABP/ECMO					X	X	X	X
Adverse events	X	X	X	X	X	X	X	X
Notes	*	†	‡					§

*Tests will not be performed at screening if results are available from equivalent testing within the preceding 7 days.
†Conducted within 1 week of screening. Medical history (illnesses, surgeries, allergies, smoking, alcohol or substance use) and demographics (age, gender, height, weight, ethnicity). Transoesophageal echocardiography, assays of myocardial injury markers and blood gas analysis will be performed after anaesthesia induction but before surgery. Time until cardiac arrest is defined as the interval from cardioplegia administration to the onset of the first arrest (only the first arrest will be analysed). Time until cardiac recovery is defined as the interval from aortic unclamping until spontaneous rhythm restoration.
‡Blood gases will be analysed at 10 min after cardioplegia infusion and 10 min after aortic unclamping.
§If a subject withdraws after 48 hours, myocardial injury markers do not need to be assessed.
CPB, cardiopulmonary bypass; ECMO, extracorporeal membrane oxygenation; IABP, intra-aortic balloon pump; ICU, intensive care unit; NYHA, New York Heart Association; TEE, transoesophageal echocardiography; TTE, transthoracic echocardiography.

habits like smoking, alcohol use and substance abuse will be collected. Functional classification according to the New York Heart Association scale[25] will be determined, and the EuroSCORE II will be calculated as described.[26]

Respiration, heart rate, non-invasive blood pressure and body temperature will be measured. A complete blood analysis will be conducted, including red and white blood cell counts, platelet counts, haemoglobin concentration, haematocrit and routine blood biochemistry tests, such as aspartate aminotransferase, alanine aminotransferase, albumin, total bilirubin, direct bilirubin, blood urea nitrogen/urea and serum creatinine. Coagulation activity will be assessed through assays of activated partial thromboplastin time, prothrombin time, thrombin time and fibrinogen. A serum-based pregnancy test will be performed for all premenopausal women. Additionally, subjects will undergo 12-lead ECG and transthoracic echocardiography (TTE) to determine LVEF.

If tests equivalent to those described above were performed within 7 days prior to the first visit, those results will be used instead of conducting new tests.

During the second assessment, conducted within 1 week of the first one, participants will be randomised into the Huaxi-1 or HTK group using a central randomisation system via the Randomizer software (V.1.0, 2022, Randomizer.org). Patients will be blinded to their allocation to minimise bias. After anaesthesia induction and before surgery, the heart rate, invasive blood pressure and body temperature will be measured; baseline LVEF will be determined using TEE; levels of CK-MB, cTnI and hs-cTnT will be assayed; and blood and blood gases will be analysed (pH, pCO_2, excess base, standard and actual bicarbonate, lactate, Na^+, K^+, Ca^{2+}, Cl^-, haemoglobin, haematocrit, glucose).

The participants will undergo open-heart surgery involving CPB and their allocated cardioplegia. Thoracoscopic procedures may be performed at the surgeon's discretion and according to hospital procedures.

The third assessment is intraoperative and comprises analysis of arterial blood gases within 10 min after initial cardioplegia and 10 min after aortic cross-clamp removal in order to assess acid-base balance, electrolytes, blood dilution and glucose levels. Details will be recorded about the surgical approach (eg, open thoracotomy, thoracoscopy), type of surgery (eg, coronary artery bypass grafting, valve surgery), type of transfusion (autologous blood, red blood cells, plasma, platelets) and volume, CPB (duration of CPB, duration of aortic cross-clamping), cardioplegia administration (mode of delivery, volume infused, number of infusions and intervals and time until first cardiac arrest), the time from release of aortic cross-clamping until spontaneous cardiac recovery, whether resuscitative interventions such as defibrillation will be needed to restore cardiac rhythm. The fourth assessment, also intraoperative, will be conducted within 30 min after weaning off CPB. Heart rate, invasive blood pressure and body temperature will be measured, LVEF will be determined using TEE; levels of CK-MB, cTnI and hs-cTnT will be assayed; and arterial blood gases will be analysed.

Postoperative follow-up

Each participant will be followed until hospital discharge or postoperative day 30, whichever occurs first. The longest follow-up is expected to be 37 days.

At 6 hours, 12 hours, 24 hours and 48 hours after surgery (fifth through eighth assessments), respiration, heart rate, invasive blood pressure and body temperature will be recorded; and levels of CK-MB, cTnI and hs-cTnT will be assayed. If invasive arterial pressure is not being performed at these times, blood pressure will be measured non-invasively. Participants will be transferred to the intensive care unit after chest closure in the operating room for further treatment and monitoring. Subsequently, they will be moved to the general ward for continued care. The time points of ICU transfer in and out will be recorded. If a participant is readmitted to the ICU after transferring out, the time points of readmission and subsequent transfer out will also be documented.

At the eighth assessment, laboratory tests will be assayed the same as in the first assessment.

The ninth assessment will be performed at discharge for patients discharged before day 30, or on day 30 for patients who remain hospitalised. Respiration, heart rate, non-invasive blood pressure and body temperature will be recorded and laboratory tests will be assayed the same as in the first assessment. Use of an IABP or ECMO at any time after surgery until the end of follow-up will be recorded. Adverse events will be comprehensively documented from the first assessment through the end of the ninth assessment.

Data collection

Data in this trial will be collected using electronic case report forms, which only authorised investigators will be able to access in order to ensure data quality and patient privacy. To the extent possible, data will be recorded in accordance with Good Clinical Practice for Medical Device Clinical Trials.[11] Throughout data management, logical checks will be performed on the database by trial monitors to help ensure timely and accurate data collection (online supplemental appendix 2). Any data that appear inconsistent or implausible will be addressed through a query form sent to the investigators. Each site will be responsible for verifying its own data, and corrections will be made as necessary before the database is locked for statistical analysis.

Statistical analysis

Centralised statistical analyses will be conducted using SAS V.9.4 (SAS Institute, Cary, North Carolina, USA). Continuous data will be summarised by the number of observations, mean, SD, minimum, maximum, median, first quartile and third quartile.

Primary endpoint

Analysis of covariance will be used for the primary endpoint, with baseline hs-cTnT as a covariate. Adjustments will include study centre, surgical approach, procedure type and perfusion strategy to control for potential confounders.

Secondary endpoints

For longitudinal data, repeated measures analysis of variance or mixed-effects models will be employed, depending on data characteristics and underlying assumptions. These models will evaluate group, time and group×time interactions while adjusting for potential confounders if necessary.

For non-longitudinal continuous data, intergroup differences will be assessed using t-tests or Wilcoxon rank-sum tests and within-group differences will be evaluated using paired t-tests or Wilcoxon signed-rank tests.

Categorical data will be expressed as n (%) with intergroup differences analysed using χ^2 tests or Fisher's exact tests. Differences in stratified categorical data will be assessed using the Cochran-Mantel-Haenszel test. Type 1 error will be controlled as per established statistical practices. Unless otherwise stated, differences will be considered significant at a two-sided p value<0.05.

Ethics and dissemination

This trial was approved in May 2024 by the Ethics Committee at Guangdong Provincial People's Hospital (Approval No. QX2024-021-02). Subsequent approvals were obtained from the Ethics Committees at the other participating sites: The Second Xiangya Hospital of Central South University (2024 Ethics Review No. 075), The First Affiliated Hospital of Air Force Medical University (Approval No. QX20241016-C-1); and The First Affiliated Hospital of Nanjing Medical University (Approval No. 2024-D-173). Written approval from all four ethics committees is required prior to any amendments to the trial protocol or the informed consent form. If the trial is suspended at any study site, its resumption must receive written approval from the corresponding ethics committee.

The conduct of the trial will be closely monitored by external experts in accordance with Good Clinical Practice guidelines for Medical Devices,[11] ensuring the accuracy and completeness of data collection, including the reporting of adverse events and any technical or procedural errors.

We aim to publish the trial findings in reputable international peer-reviewed journals to reach a broader audience. Cardiovascular surgeons, anaesthesiologists, perfusionists and other healthcare professionals from the four study sites will disseminate the findings at both national and international conferences. This trial is expected to provide robust validation of the efficacy and safety of a standardised, commercially available blood-based cardioplegia solution, potentially benefiting diverse patient populations with cardiovascular diseases worldwide.

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Contributors WY, ZJ, YfS, CZ and YX conceived and designed the study. WY, CW, TZ, and YW will recruit patients, and WY, GX, ZW, LZ, FY, LW, TC, JY, YfS, YhS, and JD will implement the trial. WY wrote this protocol, which YX critically revised. YX is the guarantor of the article. All authors approved the submitted version of the protocol.

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