STUDY PROTOCOL WITH STATISTICAL ANALYSIS PLAN

Effect of the oXiris Hemofilter on Early Endotoxin Burden in Patients With Septic Shock Requiring Continuous Renal Replacement Therapy: A Single-Center, Prospective, Single-Arm Clinical Trial

Official Title	Effect of the oXiris Hemofilter on Early Endotoxin Burden in Patients With Septic Shock Requiring Continuous Renal Replacement Therapy: A Single-Center, Prospective, Single-Arm Clinical Trial
NCT Number	Not yet assigned
Protocol Version	4.2
Document Date	2 July 2026
Document Type	Study Protocol with Statistical Analysis Plan

This document contains the statistical analysis plan in Section 5.5.

Compliance Statement

This investigator-initiated clinical study will be conducted in accordance with applicable Good Clinical Practice requirements, the Chinese Measures for the Administration of Investigator-Initiated Clinical Research in Medical and Health Institutions (Trial), and the Declaration of Helsinki. All study personnel will be appropriately trained. The study may begin only after written approval has been obtained from the institutional ethics committee and written informed consent has been obtained from each participant or the participant's legally authorized representative. Any protocol amendment requiring ethics review must be submitted for approval before implementation.

1. Protocol Summary

Sepsis and septic shock are among the most life-threatening conditions encountered in critical care. Their pathophysiology may include uncontrolled systemic inflammation, endotoxemia, and multiple-organ dysfunction, with substantial use of health-care resources. Extracorporeal blood purification may have a role in selected sepsis phenotypes, but high-quality clinical evidence remains limited. The oXiris hemofilter combines continuous renal replacement therapy (CRRT) with adsorption of endotoxin and inflammatory mediators. Previous observational studies have suggested possible improvements in hemodynamics and inflammatory burden, but prospective evaluation under a standardized treatment and assessment framework is needed.

This is a single-center, prospective, open-label, single-arm interventional clinical study in adults with septic shock who require CRRT. All enrolled participants will receive CRRT using the oXiris hemofilter (oXiris-CRRT). CRRT prescription, antimicrobial therapy, fluid resuscitation, vasopressor therapy, and other organ support will be provided according to standardized institutional procedures and clinical judgment. The primary endpoint is the change in blood endotoxin concentration, measured using a kinetic turbidimetric Limulus amebocyte lysate (LAL) assay, from baseline to completion of the planned 24-hour oXiris treatment. Secondary endpoints include inflammatory biomarkers (IL-6, IL-8, IL-10, TNF-alpha, C-reactive protein, procalcitonin, and white blood cell count), hemodynamic measures, organ function and organ-support requirements, intensive care unit and hospital length of stay, and 28-day and 90-day all-cause mortality. Thirteen participants are planned. The study is intended to estimate the magnitude of change in the primary biological endpoint and the frequency of safety events, thereby informing future studies and clinical practice.

2. Introduction

2.1 Scientific Background and Rationale

2.1.1 Endotoxin- and Inflammation-Driven Pathophysiology of Sepsis

Sepsis is life-threatening organ dysfunction caused by a dysregulated host response to infection and is one of the most common and fatal conditions in the intensive care unit (ICU). Global burden estimates indicate approximately 48.9 million sepsis cases and 11 million sepsis-related deaths annually, accounting for nearly one fifth of deaths worldwide and creating a major clinical and socioeconomic burden [1]. Key pathophysiologic pathways include systemic inflammation triggered by endotoxin (lipopolysaccharide) and pro-inflammatory cytokines such as IL-6 and TNF-alpha, together with coagulation and endothelial dysfunction. These processes can cause microcirculatory injury, inadequate tissue perfusion, and multiple-organ dysfunction [2]. Endotoxin burden and a hyperinflammatory state are important contributors to hemodynamic instability.

Despite advances in antimicrobial treatment, fluid resuscitation, hemodynamic support, and organ support, important therapeutic challenges remain. Some patients have persistent hemodynamic instability and hyperlactatemia despite adequate resuscitation, suggesting ongoing endotoxin- or inflammation-driven circulatory and microcirculatory dysfunction. Pharmacologic interventions targeting single inflammatory mediators or endotoxin have repeatedly failed, reflecting the biological complexity and heterogeneity of sepsis [3]. In addition, many current treatments are supportive and do not directly remove circulating

harmful mediators. Some patients therefore progress to refractory shock or multiple-organ failure. More effective and biologically targeted treatment strategies remain an important unmet need.

2.1.2 Current Status of Endotoxin-Targeted Blood Purification

Extracorporeal blood purification (EBP) uses adsorption, filtration, and dialysis to remove circulating inflammatory mediators and endotoxin and thereby reduce the harmful molecular burden that contributes to inflammation and microcirculatory injury [4,5]. In patients with septic shock and acute kidney injury (AKI), infection control, resuscitation, and organ support remain central to management, and continuous or intermittent renal replacement therapy is initiated when clinically indicated [6]. Conventional CRRT filters such as AN69 and AN69ST are primarily used for fluid management and clearance of small solutes, including electrolytes and uremic toxins. Their principal role is organ support, and their capacity to remove inflammatory mediators is limited.

The oXiris hemofilter is a multifunctional membrane developed from the AN69ST platform. It provides endotoxin adsorption, cytokine adsorption, surface heparin grafting to improve local hemocompatibility, and simultaneous dialysis or filtration support. Unlike stand-alone hemoperfusion approaches such as polymyxin B hemoperfusion, oXiris can be integrated directly into a CRRT circuit, allowing renal support and adsorption of endotoxin and inflammatory mediators within the same treatment pathway. This approach is well aligned with the care of patients with septic shock who already require CRRT.

Previous clinical studies and meta-analyses have reported associations between oXiris treatment and reductions in norepinephrine dose, lactate concentration, Sequential Organ Failure Assessment (SOFA) score, inflammatory markers, and ICU length of stay [7,8]. However, the evidence base is derived mainly from retrospective cohorts, case-control studies, and small randomized studies, and the overall certainty of evidence remains limited. The magnitude of clinical benefit, the population most likely to benefit, and the optimal standardized treatment approach require prospective evaluation. The 2024 Asia-Pacific expert consensus suggested considering oXiris in patients with sepsis or septic shock who require CRRT and have hemodynamic instability, marked inflammation, severe lactic acidosis, or coagulation abnormalities, while emphasizing the need for additional prospective evidence [9]. A 2025 Chinese expert consensus on emergency hemoadsorption also supported early hemoadsorption in selected patients with severe sepsis phenotypes after completion of initial bundle care [10].

Accordingly, a rigorously conducted prospective study is warranted in patients with septic shock who require CRRT. Standardized CRRT procedures and prespecified sampling and assessment time points will be used to evaluate the effects of oXiris treatment on endotoxin burden, inflammation, hemodynamics, organ function, and clinical outcomes.

2.2 Risk-Benefit Assessment

2.2.1 Known and Potential Risks

- Filter or circuit clotting and treatment interruption. In a hyperinflammatory state, blood and proteins may adhere to the membrane and contribute to clotting, shortened filter life, repeated filter replacement, or interruption of CRRT. Although the oXiris membrane has a heparin-grafted surface, clotting remains possible.
- Bleeding and anticoagulation-related complications. Systemic heparin may cause bleeding or heparin-induced thrombocytopenia (HIT). The heparin-grafted oXiris surface does not eliminate risks associated with systemic anticoagulation. Regional citrate anticoagulation is preferred when appropriate, but it may cause metabolic alkalosis, citrate accumulation, or electrolyte disturbances.
- Adsorption of medications. Adsorptive membranes may nonselectively remove some antimicrobial agents or other clinically important drugs, potentially reducing drug exposure and requiring dose adjustment. Therapeutic drug monitoring will be used when clinically indicated.
- Filter-, access-, or circuit-related infection and mechanical complications. Risks associated with extracorporeal circulation include air embolism, vascular-access complications, pressure alarms, and infection. Strict aseptic technique and device management will be required.

2.2.2 Potential Benefits

- Improved hemodynamics and reduced vasopressor requirements. Removal of circulating endotoxin and pro-inflammatory cytokines may reduce inflammation-driven vasodilation and capillary leakage, potentially improving mean arterial pressure and reducing vasopressor requirements.
- Reduced endotoxin and inflammatory burden. Small prospective studies and case series have reported reductions in IL-6, TNF-alpha, C-reactive protein, and endotoxin activity after oXiris treatment, supporting a plausible biological effect.
- Reduced need for organ support. Earlier stabilization of hemodynamics and improved oxygen delivery may reduce dependence on vasopressors, mechanical ventilation, or prolonged CRRT and may shorten ICU stay.
- Potential survival benefit in selected patients. Prior studies of endotoxin adsorption suggest that benefit may be concentrated within specific endotoxin ranges. The present study is not powered to establish a survival benefit but may inform future controlled studies.

2.2.3 Overall Risk-Benefit Evaluation

The biological rationale for removing endotoxin and pro-inflammatory mediators during CRRT is plausible, and available evidence supports the potential for short-term improvements in inflammatory and hemodynamic measures. Important risks include filter or circuit clotting, medication adsorption, anticoagulation-related complications, infection, and mechanical complications. These events are clinically observable and will be actively monitored and managed using standardized procedures. In adults with septic shock who already require CRRT, prospective evaluation of oXiris within a controlled research framework is considered ethically acceptable.

3. Study Objectives and Endpoints

3.1 Objectives

3.1.1 Primary Objective

To evaluate whether CRRT using the oXiris hemofilter reduces short-term blood endotoxin burden in adults with septic shock who require CRRT. This objective is intended to determine whether the proposed biological mechanism of oXiris produces a measurable short-term effect and to inform subsequent evaluation of medium- and long-term clinical outcomes.

3.1.2 Secondary Objectives

To describe and evaluate changes after oXiris-CRRT in the following domains:

- Inflammatory biomarkers: IL-6, IL-8, IL-10, TNF-alpha, C-reactive protein (CRP), procalcitonin (PCT), and white blood cell (WBC) count.
- Hemodynamic and perfusion measures: mean arterial pressure (MAP), blood lactate, and capillary refill time (CRT).
- Organ-support requirements and organ function: norepinephrine-equivalent dose (NEE) and SOFA score.
- ICU length of stay, hospital length of stay, and all-cause mortality at Days 28 and 90.

3.2 Endpoints and Definitions

3.2.1 Primary Endpoint

Change in blood endotoxin concentration from baseline to completion of the planned 24-hour oXiris treatment. Venous blood will be collected before initiation of CRRT and immediately after completion of the planned 24-hour treatment. Endotoxin concentration will be measured using a kinetic turbidimetric LAL assay. The change will be calculated as the post-treatment value minus the baseline value; a negative value indicates a reduction in endotoxin concentration.

3.2.2 Secondary Endpoints

- Inflammatory cytokines: IL-6, IL-8, IL-10, and TNF-alpha measured before treatment and immediately after completion of the planned 24-hour oXiris treatment.
- Other inflammatory markers: CRP, PCT, and WBC count measured before treatment, immediately after the planned 24-hour treatment, and during ICU follow-up according to the study schedule.
- Norepinephrine-equivalent dose: a composite measure of vasopressor intensity, calculated in micrograms/kg/minute as follows:

$$NEE = norepinephrine + epinephrine + (dopamine \times 0.01) + (phenylephrine \times 0.01) + (vasopressin \times 0.4)$$

All component doses will be converted to µg/kg/minute before calculation. NEE will be calculated at baseline and after completion of the planned 24-hour treatment.

- SOFA score: total score from 0 to 24 across respiratory, cardiovascular, renal, hepatic, coagulation, and central nervous system domains. Higher scores indicate more severe organ dysfunction. SOFA will be assessed at baseline and after completion of the planned 24-hour treatment.
- ICU length of stay: days from enrollment to ICU discharge or death in the ICU.
- Hospital length of stay: days from enrollment to hospital discharge or in-hospital death.
- All-cause mortality at Day 28 and Day 90 after enrollment.

3.2.3 Safety Endpoints

All adverse events (AEs) and serious adverse events (SAEs) occurring during the study will be recorded, and the investigator will assess the possible relationship of each event to the study intervention.

4. Study Population

4.1 Inclusion Criteria

- Age 18 years or older.
- Septic shock according to Sepsis-3 criteria: known or suspected infection with acute organ dysfunction (increase in SOFA score of at least 2 points), and persistent hypotension requiring vasopressors to maintain MAP of at least 65 mmHg with blood lactate greater than 2 mmol/L despite adequate fluid resuscitation.
- AKI classified as Kidney Disease: Improving Global Outcomes (KDIGO) stage 2 or stage 3.
- A clinical decision by the treating team to initiate CRRT. Indications may include AKI, fluid overload, severe metabolic acidosis, refractory hyperkalemia, or use as circulatory support.

4.2 Exclusion Criteria

- The decision to initiate CRRT was made 24 hours or more before enrollment.
- A documented decision to limit life-sustaining treatment or not to pursue invasive treatment.
- Active major bleeding, a major bleeding event within the previous 48 hours, or a severe uncorrected bleeding tendency.
- Confirmed or suspected HIT, or a known severe allergy to heparin or the filter materials.
- Severe irreversible end-stage organ failure with an expected survival of 48 hours or less.
- Pregnancy or breastfeeding.
- Participation within the previous 6 months in another interventional study that may affect the outcomes of this study.
- Written informed consent not obtained from the participant or legally authorized representative.

4.3 Lifestyle and Concomitant-Treatment Considerations

The following restrictions and management rules are intended to support safety and consistent interpretation of the intervention. They will be explained in the informed consent process and documented after enrollment.

- Smoking. Smoking, including electronic cigarettes, is not permitted during hospitalization for the study. Pre-existing smoking history will be recorded. Any smoking during the study will be documented and assessed for possible effects on respiratory or oxygenation measures.
- Alcohol. Alcohol use is not permitted during hospitalization. Patients with acute alcohol intoxication or recent heavy alcohol use causing significant organ dysfunction will be evaluated for delayed enrollment or exclusion.
- Activity. Activity will follow routine ICU or ward practice. Strenuous activity is not encouraged except for clinically indicated early mobilization or rehabilitation. No additional restrictions apply after discharge beyond usual rehabilitation advice.
- Diet. Nutrition will follow routine ICU or ward practice. Special nutritional products or high-dose supplements that could affect inflammatory markers will be documented.
- Restricted or reportable treatments. Other hemoadsorption therapies or adsorptive devices, such as polymyxin B hemoperfusion or CytoSorb, are not permitted concurrently unless clinically necessary. Systemic immunosuppressive treatment not authorized by the study team and medications with major interactions with the CRRT filter must be reported. Clinical care and participant safety take priority. If a restricted intervention becomes necessary, it will be recorded as a co-intervention, including indication, start and stop times, and potential effect on study outcomes. The study intervention may be paused or discontinued if the co-intervention creates a safety conflict or makes the planned intervention uninterpretable; follow-up and safety assessments will continue whenever possible.

4.4 Screen Failures

4.4.1 Definition

A screen failure is a candidate who undergoes preliminary screening or provides consent but is not ultimately enrolled and does not start the study intervention. Reasons may include failure to meet eligibility criteria, refusal of consent by the patient or legally authorized representative, an urgent clinical change before baseline assessment or intervention, inability to obtain the primary endpoint samples, or another investigator-determined reason that makes enrollment inappropriate.

4.4.2 Documentation

A screening log will be maintained and will include an anonymized identifier, screening date, screening outcome, and the specific reason for screen failure.

4.5 Recruitment and Retention

- Recruitment site. The study will be conducted at Sun Yat-sen Memorial Hospital, which has CRRT capability, access to oXiris filters, and laboratory capacity for endotoxin and cytokine testing.
- Recruitment method. ICU admissions will be screened daily by study physicians for Sepsis-3 septic shock and a possible need for CRRT.
- Planned recruitment. Based on ICU capacity and CRRT use, approximately 10-20 participants may be screened or recruited per year. The target enrollment is 13 participants over approximately 12 months, subject to feasibility observations during the pilot phase.
- Retention. At enrollment, multiple contact methods will be collected, including the participant and family or emergency contacts. Follow-up reminders may be sent 7 days and 24 hours before scheduled post-discharge follow-up. Telephone, outpatient, home-based, electronic questionnaire, or secure online communication may be used when appropriate.

5. Study Design and Statistical Analysis Plan

5.1 Overall Design

This is a single-center, prospective, open-label, single-arm interventional clinical study. Adults with septic shock who require CRRT will be enrolled consecutively. Each participant will receive CRRT using the oXiris hemofilter, with biological measures, medication exposure, clinical assessments, and outcomes collected at prespecified time points.

5.1.1 Primary Study Hypothesis

Blood endotoxin concentration will decrease after 24 hours of oXiris-CRRT compared with baseline.

5.1.2 Secondary Study Hypotheses

- oXiris-CRRT may improve hemodynamic measures and reduce early organ-support requirements, including vasopressor intensity.
- ICU and hospital length of stay and 28-day and 90-day mortality will be described to estimate event rates and effect sizes for future controlled studies.
- The safety profile of oXiris-CRRT will be acceptable with respect to filter life, bleeding or clotting complications, medication adsorption, and device-related events.

5.2 Study Procedures and Schedule

5.2.1 Study Procedures

- Patient identification and preliminary screening. ICU patients will be screened daily to determine whether they meet the definition of septic shock and are expected to require CRRT.
- Eligibility confirmation and informed consent. Inclusion and exclusion criteria will be assessed, and written informed consent will be obtained from the participant or legally authorized representative. The candidate will be entered in the screening log.
- Baseline assessment. Within 24 hours after enrollment, the following will be collected: age, sex, height, weight, smoking and alcohol history; relevant medical history, including cardiovascular disease, hypertension, diabetes, dyslipidemia, peripheral vascular disease, liver and kidney disease, pulmonary disease, and malignancy; source of infection; microbiologic information; SOFA score; Simplified Acute Physiology Score II (SAPS II); vasopressor use and calculated NEE; arterial blood gas measures including lactate; complete blood count; electrolytes; and liver and kidney function tests. Before CRRT begins, venous blood will be collected through the central venous access established for CRRT to measure endotoxin and inflammatory biomarkers and to avoid unnecessary additional venipuncture. CRRT should begin promptly after baseline sampling. MAP and capillary refill time will be assessed at the bedside.
- Intervention and treatment-period data collection. oXiris-CRRT will be initiated and continued for a planned 24 hours. Immediately after completion, venous blood will be collected for endotoxin, inflammatory biomarkers, and therapeutic drug monitoring when applicable. MAP, vasopressor use and NEE, capillary refill time, SOFA score, filter performance, CRRT prescription, AEs, and SAEs will be recorded.
- In-hospital observation. Clinical status and selected laboratory measures will continue to be collected while the participant remains hospitalized.
- Long-term follow-up. Survival status will be determined at Days 28 and 90 after enrollment.

5.2.2 Schedule of Assessments

Study time points include screening on Day 1; the treatment period on Day 2 at 0 hours and 24 hours; in-hospital follow-up on Days 3, 5, and 7; and long-term follow-up on Days 28 and 90. Study personnel will use standardized procedures for data collection, sample collection, and outcome assessment.

- Screening period. Written informed consent and eligibility confirmation will occur on the day of enrollment. Baseline information will be collected within 24 hours after enrollment.
- Treatment period. The oXiris hemofilter is planned to run continuously for 24 hours. Before treatment and immediately after treatment, venous blood will be collected for endotoxin and inflammatory biomarkers; MAP, capillary refill time, vasopressor use, NEE, SOFA score, arterial blood gas results, blood counts, filter parameters, CRRT prescription, AEs, and SAEs will be recorded.
- In-hospital follow-up. For participants who remain hospitalized, clinical status and scheduled measures will be collected on Days 3, 5, and 7.
- Long-term follow-up. Survival status will be collected at Days 28 and 90 by telephone, WeChat, outpatient contact, or another appropriate method.

Data analysis will begin within approximately 1 month after the final participant completes follow-up. Analysis and preparation of the study report are expected to require an additional 2-3 months.

5.3 Methods to Reduce Bias

- Consistency of enrollment and intervention delivery. Consecutive enrollment will be used. CRRT procedures, anticoagulation, and sample collection will follow standardized operating procedures. Study staff will be trained before study initiation, with periodic quality-control review.
- Open-label design and objective endpoints. The study is open label. Samples for the primary biological endpoint will be coded with anonymized identifiers and analyzed by the central laboratory or trained laboratory personnel. Mortality is an objective endpoint. Study-site and participant identifiers will be removed from analysis datasets before database lock.

5.4 Definition of Study Completion

The study will be considered complete when the final participant has completed the Day 90 follow-up assessment.

5.5 Statistical Analysis Plan

5.5.1 Sample Size and Rationale

The sample-size calculation is based on the primary endpoint, defined as the within-participant change in endotoxin concentration from baseline to completion of the planned oXiris treatment:

$$\Delta = X_{post} - X_{pre}$$

A value of $\Delta < 0$ indicates a reduction in endotoxin concentration. Let $\mu\Delta$ denote the population mean change and μC the prespecified historical-control mean change. If the upper limit of the two-sided 95% confidence interval for $\mu\Delta$ is below μC, the observed change will be considered greater than the historical-control benchmark. The corresponding hypotheses used for sample-size planning are:

$$H0: \mu\Delta \geqslant \mu C,\quad H1: \mu\Delta < \mu C$$

Because the criterion is based on the upper limit of a two-sided 95% confidence interval, the calculation corresponds to a one-sided alpha of 0.025. AN69ST was used as the historical benchmark. A previous study reported a mean reduction of 0.08 EU/mL with AN69ST [11], giving μC = -0.08 EU/mL under the present definition of change.

A case series of 60 patients with sepsis reported a baseline endotoxin concentration of 0.55 [0.44-0.68] EU/mL and a post-oXiris concentration of 0.25 [0.13-0.41] EU/mL [12]. Because the results were reported as medians and interquartile ranges, the median was used as an approximation to the mean and the standard deviation was approximated as:

$$SD \approx \frac{IQR}{1.35}$$

The expected mean change is:

$$\mu_1 = 0.25 - 0.55 = -0.30$$

The approximate baseline and post-treatment standard deviations are:

$$\sigma_{pre} \approx \frac{0.68 - 0.44}{1.35} = 0.1778$$

$$\sigma_{post} \approx \frac{0.41 - 0.13}{1.35} = 0.2074$$

For the paired design, the standard deviation of the change is estimated as:

$$\sigma_\Delta = \sqrt{\sigma_{pre}^2 + \sigma_{post}^2 - 2\rho\sigma_{pre}\sigma_{post}}$$

Using a conservative within-participant correlation of ρ = 0.40 gives σΔ ≈ 0.2124. The normal-approximation sample-size expression is:

$$n \approx \left(\frac{(Z_{1-\alpha} + Z_{1-\beta})\sigma_{\Delta}}{\mu_C - \mu_1} \right)^2$$

With α = 0.025 and power (1-β) = 0.80, the final calculation was performed using an exact one-sample t-distribution approach because of the small anticipated sample. Ten evaluable participants are required. Allowing for approximately 20% attrition or non-evaluable primary-endpoint data, the target enrollment is 13 participants.

5.5.2 Analysis Populations

- Full Analysis Set. All enrolled participants who receive at least one oXiris treatment and have a baseline and at least one post-baseline assessment of the primary endpoint. Primary and secondary efficacy analyses will use this set.
- Per-Protocol Set. Participants in the Full Analysis Set who complete the planned intervention and primary-endpoint assessment without a major protocol deviation that could materially affect efficacy evaluation. This set will be used for sensitivity analysis.
- Safety Analysis Set. All participants who receive any part of the study intervention. Safety analyses will use this set.

5.5.3 General Statistical Principles

Analyses will emphasize descriptive statistics and interval estimation. The prespecified 95% confidence-interval criterion used in sample-size planning will be reported for the primary endpoint; no additional confirmatory hypothesis testing is planned. The primary analysis will use the Full Analysis Set, the Per-Protocol Set will be used for sensitivity analysis, and the Safety Analysis Set will be used for safety outcomes. Analyses will be performed using R software.

- Baseline characteristics. Continuous variables will be summarized using mean and standard deviation or median and interquartile range, depending on distribution. Minimum and maximum may also be reported. Categorical variables will be summarized using counts and percentages.
- Primary endpoint. Individual participant changes in endotoxin concentration will be displayed. The mean change and its two-sided 95% confidence interval will be reported. The median change and interquartile range will also be reported when informative.
- Secondary endpoints. Inflammatory biomarkers, NEE, SOFA score, and other continuous endpoints will be summarized at each time point using mean and standard deviation or median and interquartile range. Changes from baseline will be reported with means and two-sided 95% confidence intervals when appropriate. Categorical endpoints will be summarized using counts and percentages. Repeated measures will be displayed in tables and longitudinal plots.
- Length-of-stay and survival outcomes. Day 28 and Day 90 all-cause mortality will be summarized as counts, percentages, and two-sided 95% confidence intervals. A Kaplan-Meier survival curve will also be presented. ICU and hospital length of stay will be summarized using median and interquartile range.
- Safety outcomes. AEs and SAEs will be summarized by event type and participant incidence and stratified by severity and assessed relationship to the intervention.
- Bias control. Consistent eligibility criteria, standardized intervention procedures, fixed sampling windows, and consistent endpoint definitions will be used to reduce selection and measurement bias.
- Sensitivity analysis. The primary endpoint will be reanalyzed descriptively in the Per-Protocol Set and compared with the Full Analysis Set to assess consistency and robustness.
- Missing data. The amount and pattern of missing data will be described. Where the assumptions are supportable, multiple imputation may be used for data that are not missing completely at random. Sensitivity analyses will be performed for the primary endpoint.

6. Study Intervention

6.1 Intervention and CRRT Prescription

All participants will receive CRRT using the oXiris hemofilter. No concurrent control group is included. If a standard filter must be substituted for safety or technical reasons, the reason will be documented as a protocol deviation. To reduce treatment heterogeneity, all participants will receive continuous venovenous hemofiltration (CVVH) using a Prismaflex system and an oXiris hemofilter. The planned oXiris treatment duration is 24 hours. Whether CRRT continues beyond the study treatment period, and the choice of any subsequent filter or modality, will be determined by the treating physician according to the participant's evolving clinical condition. CRRT prescription, anticoagulation, vascular access, and replacement-fluid composition will follow standardized institutional procedures.

Standard treatment parameters are:

- Anticoagulation: regional citrate anticoagulation is preferred. If contraindicated or unsuitable, another clinically appropriate strategy or no anticoagulation may be used.
- Prescribed effluent dose: 25-30 mL/kg/h.
- Blood flow rate: 100-150 mL/min.
- Replacement-fluid rate: 1,000-1,500 mL/h, with a 50/50 pre-dilution and post-dilution distribution.
- Replacement-fluid composition: the institutional standard CRRT solution, adjusted as clinically necessary for electrolyte and acid-base status.
- Treatment duration: continuous operation for 24 hours. The filter may be replaced earlier because of clotting, increased transmembrane pressure, suspected adsorption saturation, or another clinical or technical indication.

Clinical decisions to discontinue or transition renal replacement therapy will follow applicable standards and clinical judgment. CRRT may be discontinued when vital signs and hemodynamics are stable, nonrenal organ function has recovered, and fluid, electrolyte, acid-base, and volume abnormalities have been corrected. Participants whose overall condition has stabilized but whose kidney function has not recovered may transition to intermittent renal replacement therapy. CRRT may be paused when urine output and renal recovery are adequate for fluid and nutritional requirements. If kidney function does not recover but systemic inflammation and volume status stabilize, intermittent hemodialysis or peritoneal dialysis may be used according to clinical need.

6.1.1 Timing of Study Assessments

Sampling and assessment time points will be standardized. Post-treatment venous blood samples will be obtained immediately after completion of the planned 24-hour oXiris treatment.

6.2 Storage, Dispensing, and Study Responsibilities

- Storage. oXiris filters and any clinically required standard replacement filters will be stored by the institutional device or pharmacy management unit at 10-30°C, protected from moisture, compression, and direct sunlight. Expiration dates will be checked regularly, and device numbers and use will be traceable.
- Dispensing. Filters will be issued by the study nurse or device manager using the participant study number. Dispensing time, lot number, and duration of use will be recorded.
- Responsibilities are summarized below:

Role	Responsibilities
Principal/Sub-Investigator	Clinical decisions and medical management
Research Nurse	Enrollment coordination, specimen collection, and treatment documentation

Role	Responsibilities
CRRT Technician	CRRT operation, settings, and monitoring of filter performance
Laboratory Personnel	Endotoxin and inflammatory-biomarker testing
Data Manager	Data entry, verification, and follow-up of missing data

6.3 Intervention Adherence

The following measures will support consistent intervention delivery:

- A standardized CRRT operating procedure will be used.
- All study personnel will receive standardized training.
- The research nurse will review CRRT settings for protocol consistency.
- Filter replacement will follow the manufacturer's instructions and institutional procedures.
- Periodic monitoring will be performed to verify adherence.

Adherence will be assessed using CRRT treatment logs, actual versus prescribed treatment time, filter selection, and premature treatment interruptions.

Adequate adherence is defined as actual oXiris-CRRT operating time of at least 80% of the prescribed time during the first 24 hours and no concurrent use of another adsorptive blood-purification device. Use of a standard filter or early termination for safety reasons will be documented and classified as a protocol deviation.

6.4 Concomitant Treatments

The following treatments are permitted according to clinical guidelines and must be recorded, including type, dose, and timing:

- Antibacterial, antifungal, and antiviral agents.
- Vasopressors and other vasoactive medications.
- Corticosteroids.
- Nutritional support.
- Blood products, including red blood cells, plasma, and platelets.
- Intravenous fluids.
- Sedation, analgesia, and mechanical ventilation.
- Therapeutic drug monitoring for clinically important medications, including beta-lactam antibiotics, when indicated.

The following are prohibited or should be used with caution because they may interfere with interpretation of the blood-purification effect:

- Concurrent use of another adsorptive blood-purification device, such as polymyxin B hemoperfusion or CytoSorb.
- Nonstandard supratherapeutic anticoagulation not required for clinical care.
- Experimental treatments that conflict with the study intervention. If required for emergency care, their use must be reported and documented.

6.4.1 Emergency Management

Immediate clinical rescue measures will be initiated and documented if any of the following occur:

- Hemodynamic deterioration with persistent MAP below 60 mmHg.
- Severe bleeding.
- Complete filter clotting resulting in treatment interruption.
- Severe electrolyte disturbance, such as serum potassium greater than 6.5 mmol/L.
- Device failure that prevents maintenance of life support.

Additional vasopressors, urgent restart of CRRT, blood-product transfusion, or replacement of the oXiris filter may be used as clinically necessary. If a standard filter is required because of clotting, device failure, or a contraindication, the event will be documented as a rescue measure, co-intervention, and protocol deviation as applicable.

7. Intervention Discontinuation, Participant Withdrawal, and Loss to Follow-up

7.1 Temporary or Permanent Discontinuation of the Study Intervention

The study intervention will be paused or discontinued if any of the following occur:

- An SAE that the investigator considers possibly or probably related to the study intervention, including sustained deterioration directly attributed to filter use, severe bleeding requiring at least 4 units of red blood cells, severe hypotension preventing adequate CRRT blood flow, or strongly suspected filter-related HIT with thrombocytopenia and a positive anti-PF4 antibody test.
- A technical problem involving the filter or CRRT system, including at least two filter-clotting episodes within 24 hours, severe vascular-access failure without a prompt alternative, or inability to maintain a prescribed dose of at least 18 mL/kg/h.
- A clinical emergency that takes priority, such as urgent surgery or the need for a higher level of organ support, including extracorporeal membrane oxygenation, requiring a change in the CRRT approach.
- The investigator determines that continued intervention poses an unacceptable risk.

Management after interruption or discontinuation will include:

- Continuation of scheduled clinical data collection whenever possible, even if the intervention is paused.
- Possible restart of CRRT after resolution of the cause, such as stabilization of hemodynamics, correction of clotting, or re-establishment of vascular access, if judged appropriate by the investigator.
- Retention in the Safety Analysis Set if the CRRT plan is changed, the filter is changed, or CRRT is stopped.

7.2 Participant Withdrawal from the Study

A participant may withdraw or be withdrawn for reasons including:

- The participant or family requests withdrawal without being required to provide a reason.
- The investigator determines that continued participation is not in the participant’s best interest, including substantial clinical deterioration.
- The participant is transferred or discharged and declines further follow-up.
- A clinically required treatment conflicts with the study intervention.
- The participant is lost to follow-up according to the prespecified definition.

Data collected before withdrawal will be retained for safety analysis as permitted by the informed consent and applicable requirements. The study team will seek permission to continue noninterventional follow-up, including collection of Day 28 and Day 90 survival status.

7.3 Loss to Follow-up

A participant will be considered lost to follow-up if any of the following apply:

- Three consecutive attempts to contact the participant and family according to the follow-up plan are unsuccessful.
- Survival status cannot be obtained within 15 days after a scheduled follow-up time point.
- The participant is transferred and clinical information cannot be obtained.

Measures to reduce loss to follow-up include collection of at least three contacts, education before discharge, scheduled reminders by telephone, text message, or WeChat, and use of a centralized follow-up team.

Missing data will be handled as described in Section 5.5.3, and sensitivity analyses will be performed for the primary endpoint.

8. Adverse Events and Unanticipated Events

8.1 Adverse Events

8.1.1 Definition

An adverse event is any unfavorable medical occurrence during the study, regardless of whether it is considered causally related to the study intervention. Events that must be recorded include, but are not limited to:

- New or worsening clinical symptoms, including fever, chills, rash, pruritus, dyspnea, chest discomfort, altered consciousness, fatigue, nausea, vomiting, diarrhea, or bleeding tendency.
- Clinically significant changes in vital signs, including heart rate below 50 or above 130 beats/minute, systolic blood pressure below 90 or above 180 mmHg, respiratory rate below 8 or above 35 breaths/minute, or temperature above 38.5°C or below 35°C.
- New or worsening laboratory abnormalities, including a platelet reduction of at least 30% or an absolute reduction of at least 50 × 10^9/L; clinically significant WBC changes; international normalized ratio of at least 1.5; activated partial thromboplastin time prolonged by at least 10 seconds; fibrinogen below 1.5 g/L; alanine or aspartate aminotransferase above 3 times the upper limit of normal; increased bilirubin; lactate above 2 mmol/L; creatinine above 1.5 times baseline; clinically important abnormalities of potassium, sodium, calcium, magnesium, or phosphate; metabolic acidosis; or a clinically significant reduction in arterial oxygen tension.
- Device- or treatment-related abnormalities, including filter malfunction, blood-line problems, risk of air embolism, pressure alarms, clotting, or vascular-access infection.
- Other medically important events, including clinical deterioration, secondary infection, new organ dysfunction, or the need for additional organ support.

For each event, the investigator will assess the event type, severity, relationship to the intervention, actions taken, and outcome, and will report the event as required.

8.1.2 Event Categories

- Filter-related events: unplanned filter clotting, defined as filter life below 12 hours with CRRT interruption and filter replacement because of clotting; and other unplanned filter replacement because of clotting, increased pressure, hemolysis, or suspected adsorption-related dysfunction.
- Bleeding events: classified using relevant International Society on Thrombosis and Haemostasis definitions as major bleeding, clinically relevant nonmajor bleeding, or minor bleeding.
- Other events, including:
  - HIT, defined by a platelet decrease of at least 50% or a 4Ts score of at least 4 with a positive HIT antibody test.
  - Electrolyte or metabolic disturbances.
  - Medication-adsorption events, such as inadequate exposure to an essential anti-infective agent requiring supplemental dosing.
  - Device- or procedure-related events, such as air in the infusion line or abnormal circuit pressure.
  - Serious adverse events, including death, a life-threatening event, hospitalization or prolonged hospitalization, persistent or significant disability, or congenital anomaly.

8.1.3 Evaluation of Adverse Events

Each AE will be evaluated in three domains:

- Severity, using the following categories based on the Common Terminology Criteria for Adverse Events framework: mild, moderate, severe, life-threatening, or death.
- Relationship to the intervention, classified by the investigator as definitely related, possibly related, possibly unrelated, or definitely unrelated.

- Expectedness, based on the oXiris instructions for use, CRRT guidance, and published literature. Known events may include filter clotting, thrombocytopenia, electrolyte disturbance, hemodynamic instability, and reduced antibiotic exposure due to adsorption.

8.1.4 Reporting and Follow-up

- All AEs will be documented in the case report form, including onset, duration, clinical features, severity, treatment, and outcome. Events will be reviewed at each follow-up assessment.
- SAEs will be reported to the principal investigator and ethics committee within 24 hours and documented using the required SAE reporting form.
- Unexpected serious events will be reported on an expedited basis according to applicable requirements.
- All AEs and SAEs will be followed until a clear clinical outcome is established, including recovery, improvement, persistence, or death.

8.2 Events of Special Interest Affecting Vulnerable Populations

Events or circumstances that may disproportionately affect vulnerable populations, including children, pregnant individuals, or persons who are economically or socially disadvantaged, require particular attention to ensure safety and equitable conduct. Children and pregnant or breastfeeding individuals are not eligible for this study.

8.3 Unanticipated Events

An unanticipated event is an event not reasonably expected from the protocol, device information, or informed consent materials. Examples include procedural errors, device failures, or data-management problems. All such events will be documented, evaluated, and reported as required.

9. Data Collection and Management

9.1 Case Report Forms and Electronic Data Capture

- Case report forms. Study data will be collected using standardized case report forms to support consistency and reliability. The forms will include baseline information, primary and secondary endpoint data, clinical assessments, and AE documentation.
- Electronic data capture. An electronic data-capture system or electronic case report form system will be used for data entry, processing, and management. Secure login and role-based access will protect the system and allow timely monitoring and review.

9.2 Data Management

- Data sources. Study data will be obtained directly from participants or legally authorized representatives when applicable and abstracted from medical records, then entered into the electronic data-capture system.
- Data storage. Electronic data will be stored on secure servers. Paper records may be retained as backup in locked cabinets accessible only to authorized study personnel.
- Data validation. Built-in logic checks will identify potential entry errors or inconsistencies. The study team will conduct periodic data review to support accuracy and completeness.
- Database design. The study database will be configured to capture all required data elements and support data-quality procedures.
- Data security. The data-management platform will use encryption, user-access controls, backup, and recovery procedures to protect data integrity and confidentiality.
- Data-quality review. The data-management team will periodically assess completeness, consistency, and accuracy. Identified issues will be documented and resolved promptly.
- Database lock. The database will be locked after data collection and cleaning are complete, all data queries are resolved, and the statistical analysis plan is finalized. Any post-lock change will require a formal change-control procedure.

10. Ethical Considerations

The study will comply with applicable Good Clinical Practice requirements, the Chinese Measures for the Administration of Investigator-Initiated Clinical Research in Medical and Health Institutions (Trial), and the Declaration of Helsinki. The study may begin only after approval by the institutional ethics committee. Amendments requiring ethics review will be submitted and may not be implemented until approval has been obtained, except where an immediate change is necessary to eliminate an apparent hazard to a participant.

Written informed consent will be obtained from each participant or legally authorized representative. A copy of the informed consent form and contact information for the study team and ethics committee will be provided. The study will collect clinical and personal information for scientific research. Study personnel and data analysts will be subject to confidentiality requirements. Participant identifiers and health information will not be disclosed to persons or organizations not involved in the study except as required by law, ethics oversight, or authorized monitoring. Data will be centrally managed using measures intended to prevent unauthorized disclosure.

References

[1] Rudd KE, Johnson SC, Agesa KM, et al. Global, regional, and national sepsis incidence and mortality, 1990-2017: analysis for the Global Burden of Disease Study. Lancet. 2020;395(10219):200-211.

[2] Levi M, van der Poll T. Coagulation and sepsis. Thromb Res. 2017;149:38-44.

[3] Kellum JA, Ronco C. The role of endotoxin in septic shock. Crit Care. 2023;27(1):400.

[4] Ronco C, Chawla L, Husain-Syed F, et al. Rationale for sequential extracorporeal therapy (SET) in sepsis. Crit Care. 2023;27(1):50.

[5] Berlot G, Tomasini A, Zanchi S, et al. The techniques of blood purification in the treatment of sepsis and other hyperinflammatory conditions. J Clin Med. 2023;12(5).

[6] Prescott HC, Antonelli M, Alhazzani W, et al. Surviving Sepsis Campaign: international guidelines for management of sepsis and septic shock 2026. Intensive Care Med.

[7] Siew LY, Lee ZY, Yunos NM, et al. Outcomes of extracorporeal blood purification with oXiris membrane in critically ill patients: a systematic review and meta-analysis. J Crit Care. 2024;83:154844.

[8] Wang G, He Y, Guo Q, et al. Continuous renal replacement therapy with the adsorptive oXiris filter may be associated with lower 28-day mortality in sepsis: a systematic review and meta-analysis. Crit Care. 2023;27(1):275.

[9] Zhang L, Srisawat N, Lee CC, et al. Extracorporeal blood purification with the oXiris filter for patients with sepsis and hyperinflammatory conditions: the Asia-Pacific oXiris Expert Meeting 2024 consensus statements. Blood Purif. 2025;54(11):621-638.

[10] Emergency Medicine Branch of the Chinese Medical Association; Emergency Medicine Branch of the Beijing Medical Association. Chinese expert consensus on emergency hemoadsorption (2025). Chinese Journal of Emergency Medicine. 2025;34(10):1343-1351. [Article in Chinese].

[11] Broman ME, Hansson F, Vincent JL, et al. Endotoxin- and cytokine-reducing properties of the oXiris membrane in patients with septic shock: a randomized crossover double-blind study. PLoS One. 2019;14(8):e0220444.

[12] Turani F, Barchetta R, Falco M, et al. Continuous renal replacement therapy with the adsorbing filter oXiris in septic patients: a case series. Blood Purif. 2019;47(Suppl 3):1-5.