

Supplemental file 1 - Eligibility criteria

Trial Population

The target patient population consist of adult patients in the ICU or intermediate care unit with sepsis and new, recent onset AKI. Consecutive adult patients with sepsis requiring vasopressor therapy will be systematically screened for AKI as soon as possible following start of vasopressor treatment. In order to enrol a typical, random sample, reflecting the entry criteria, informed consent will be sought in all patients with AKI and no exclusion criteria.

Only patients with a signed and dated informed consent form (ICF) in compliance with local regulations will be enrolled and randomly assigned to trial drug providing all inclusion criteria and none of the exclusion criteria are met. Deviations from the inclusion and exclusion criteria could potentially jeopardize the scientific integrity of the trial, regulatory acceptability, and, most importantly, patient safety. Therefore, strict adherence to the eligibility criteria as specified in the protocol is essential. Prospective approval of protocol deviations to recruitment and enrolment criteria, also known as protocol waivers or exemptions, are not permitted.

Inclusion Criteria

To be eligible for this trial, a patient must meet all of the following inclusion criteria:

1. 18 years or older.
2. In the ICU or intermediate care unit for clinical reasons.
3. Have sepsis requiring vasopressor (norepinephrine, epinephrine, dopamine, phenylephrine, vasopressin, or angiotensin II) therapy, i.e.:
 - a) suspected or proven bacterial or viral infection.

and

 - b) on vasopressor therapy ($\geq 0.1 \mu\text{g}/\text{kg}/\text{min}$ norepinephrine or equivalent) for sepsis-induced hypotension for at least one hour despite adequate fluid resuscitation according to clinical judgement. Following the initial one hour on at least $0.1 \mu\text{g}/\text{kg}/\text{min}$ norepinephrine or equivalent, any dose of vasopressor counts as vasopressor therapy.

The combination of a) and b) automatically ensures that patients fulfil the Sepsis-3 criteria as $0.1 \mu\text{g}/\text{kg}/\text{min}$ norepinephrine corresponds to a score of +4 on the Cardiovascular sub-score of the SOFA score.

4. Have AKI according to at least one of the below KDIGO criteria, a to d:
 - a) An absolute increase in serum or plasma creatinine (CR) by $\geq 0.3 \text{ mg}/\text{dL}$ ($\geq 26.5 \mu\text{mol}/\text{L}$) within 48 hours.

or

 - b) A relative increase in CR to ≥ 1.5 times the pre-AKI reference CR value, which is known or presumed to have occurred within prior 7 days.

or

 - c) A decrease in urinary output to $< 0.5 \text{ mL}/\text{kg}/\text{hour}$ for a minimum of 6 hours following adequate fluid resuscitation.

or

 - d) If the patient does not have a known history of CKD and there is no pre-AKI reference CR value available from the past 12 months: a CR value greater or equal to the levels presented in the Table, with the increase in CR presumed to have occurred within prior 7 days.

Table: Gender and Race Corrected Cut-off Values for Serum or Plasma CR Based on 1.5 Times Estimated Normal Values for Age Group² Age (years)	Black males mg/dL ($\mu\text{mol}/\text{L}$)	Other males mg/dL ($\mu\text{mol}/\text{L}$)	Black females mg/dL ($\mu\text{mol}/\text{L}$)	Other females mg/dL ($\mu\text{mol}/\text{L}$)
20-24	2.3 (200)	2.0 (173)	1.8 (159)	1.5 (132)
25-29	2.3 (200)	1.8 (159)	1.7 (146)	1.5 (132)
30-39	2.1 (186)	1.8 (159)	1.7 (146)	1.4 (120)
40-54	2.0 (173)	1.7 (146)	1.5 (132)	1.4 (120)
55-65	2.0 (173)	1.7 (146)	1.5 (132)	1.2 (107)
>65	1.8 (159)	1.5 (132)	1.4 (120)	1.2 (107)

5. Provision of signed and dated ICF in accordance with local regulations.

Exclusion Criteria

A patient who meets any of the following criteria is excluded from participation in this trial:

1. a) At sites where enrolment of 'moderate to severe' (eGFR 25-45 mL/min/1.73 m²) CKD patients is allowed, patients with a pre-AKI reference eGFR <25 mL/min/1.73 m² are excluded.

- For patients with known CKD, the most recent eGFR prior to index hospitalization needs to be documented as ≥25 mL/min/1.73 m².

- For patients with known CKD but no known eGFR prior to hospitalization, presentation eGFR between 25-60 mL/min/1.73 m² can also be used to rule out 'severe' CKD.

b) At sites where enrolment of 'moderate to severe' CKD patients is NOT allowed, patients with 'moderate to severe' CKD defined as a pre-AKI reference eGFR <45 mL/min/1.73 m² are excluded.

- For patients with known CKD, the most recent eGFR prior to index hospitalization needs to be documented as ≥45 mL/min/1.73 m².

- For patients with known CKD but no known eGFR prior to hospitalization, presentation eGFR between 45-60 mL/min/1.73 m² can also be used to rule out 'moderate to severe' CKD.

Due to limited renal reserve, even mild renal insults may trigger the diagnosis of AKI in patients with severe CKD. These "acute on chronic" incidences are often transient with a good prognosis and lower mortality. Therefore, patients with 'moderate to severe' CKD are excluded from the main trial population. However, as CKD patients are more prone to SA-AKI, a limited number of 'moderate to severe' CKD patients with eGFR ≥25-45 mL/min/1.73 m² will be enrolled in order to assess the effect of recAP in these patients. NOTE: a recent eGFR value below the thresholds (i.e., below <45 or <25 ml/min/1.73 m², respectively) at time of screening does not exclude the patient if the patient does not have a pre-AKI reference eGFR below the required threshold.

2. Advanced chronic liver disease, defined as a Child-Pugh score of 10 to 15 (Class C).

Patients with advanced chronic liver disease have a very high mortality rate due to their underlying disease, which is unlikely to be influenced by treatment with trial drug.

3. Acute pancreatitis without proven infection.

Acute pancreatitis may mimic sepsis. Therefore, a proven infection is needed in these patients. Without an established infection, these patients are excluded.

4. Urosepsis related to suspected or proven urinary tract obstruction.

AKI in urosepsis patients due to obstruction often resolves quickly with no sequela following elimination of the obstruction. As the mechanism of AKI may be different than in classical SA-AKI, these patients are excluded.

5. Main cause of AKI not sepsis.

If AKI is believed to be due to other causes than sepsis, e.g. nephrotoxic drugs, renal perfusion-related (e.g., acute abdominal aortic aneurysm, dissection, renal artery stenosis) or rhabdomyolysis the patient is excluded as these other causes of AKI have a different pathophysiology that is less likely to be influenced by treatment with recAP.

6. Proven or suspected SARS-CoV-2 infection. NOTE: This exclusion criterion does not apply to patients in the COVID-19 population, in which COVID-19 should be the main cause of SA-AKI.

At the time of completion of this protocol, there is limited knowledge about the pathophysiology of AKI in COVID-19 patients and associated outcomes. Also, there is currently no experimental data available showing an effect of recAP in these patients. Therefore, patients with proven or suspected SARS-CoV-2 infection are excluded from the main trial population. However, a small separate cohort of COVID-19 patients will be enrolled to provide exploratory data of the effect of recAP in COVID-19 patients.

7. Severe burns requiring ICU treatment.

Clinical symptoms and signs following severe burns may resemble sepsis but have special characteristics such as the lack of barrier function, leading to prolonged infection risk, excessive fluid loss, and prolonged recovery. Therefore, patients with severe burns requiring ICU treatment are excluded from the trial.

8. Severely immunosuppressed, e.g. due to:

- hematopoietic cell transplantation within past 6 months prior to Screening or acute or chronic graft-versus-host disease
- solid organ transplantation
- leukopenia not related to sepsis, i.e., preceding sepsis

- Human Immunodeficiency Virus (HIV)/Acquired Immune Deficiency Syndrome (AIDS)
- receiving chemotherapy within 30 days prior to Screening.

Patients who are severely immunosuppressed have a significantly worse prognosis and a very high mortality rate that may not be related to AKI. Therefore, such patients are excluded from the trial.

9. At high risk of being lost to follow up, e.g., due to known current or recent (within the last 6 months) IV drug abuse or known to be homeless.

It is very important to obtain data to the end of the 180-day follow-up period in accordance with the protocol. Therefore, patients at high risk of not showing up to scheduled trial visits and being LTFU are excluded from the trial.

10. Limitations to use of mechanical ventilation (MV), RRT or vasopressors and inotropes (NOTE: limitation of cardiopulmonary resuscitation (CPR) only is not an exclusion criterion).

Patients who do not wish to receive standard of intensive care with MV, RRT or vasopressors/inotropes are excluded from the trial as these patients are likely to die due to refusal of required organ support. Patients who at time of informed consent allow for active care except CPR can be included in the trial.

11. Previous administration of recAP.

12. Use of a non-marketed drug within the last month or concurrent or planned participation in a clinical trial for a non-marketed drug or device. (NOTE: Co-enrolment or concurrent participation in observational, non-interventional trials using no protocolized treatments or procedures may be allowed. Co-enrolment or concurrent participation in trials using protocolized treatments or procedures, e.g. blood draws, requires pre-approval by the TSC).

To assess efficacy and safety of recAP without confounding factors, e.g. use of other investigational drugs or devices, co-enrolment in trials involving non-marketed products is prohibited. A non-marketed product is defined as a drug or device that currently do not hold a marketing authorization in any indication. Participation or co-enrolment in purely observational, non-interventional trials using no protocolized procedures may be allowed. Participation or co-enrolment in trials using protocolized treatments or procedures, e.g. blood draws, requires pre-approval by the TSC and will only be allowed if judged by the TSC to not have an impact on the assessment of efficacy or safety of recAP.

13. Current or planned extracorporeal membrane oxygenation (ECMO) or other devices that support hemodynamics.

These patients also have a higher mortality rate that may not be related to AKI. In addition, they are often transferred to special centers interfering with trial procedures and follow-up in accordance with the protocol.

14. On RRT >24 hours before start of trial drug.

Only new onset AKI is accepted (see above). Anticipated RRT need following enrolment is NOT an exclusion criterion.

15. No longer on vasopressor therapy at time of randomization.

The requirement for ongoing vasopressor need despite adequate fluid resuscitation is included to ensure a certain severity of the condition and to align with the patient population of STOP-AKI in which 90% of patients received vasopressor therapy.

16. On continuous vasopressor therapy for >72 hours before start of trial drug.

Prolonged vasopressor therapy is associated with a risk of organ damage and a poor prognosis. Also, in patients on vasopressors for >72 hours, the link between sepsis as a cause of AKI, i.e., SA-AKI is weaker. Therefore, patients having received vasopressor therapy for >72 hours are not eligible. Start of vasopressor therapy is defined as the start time of any dose of vasopressor in the first vasopressor treatment period that includes a continuous infusion of ≥ 0.1 $\mu\text{g}/\text{kg}/\text{min}$ norepinephrine (or equivalent) for at least 1 hour for sepsis-induced hypotension in patients who have received adequate fluid resuscitation in accordance with clinical judgement and the recommendations of the Surviving Sepsis Campaign guidelines. A minimum of 12h without any vasopressor is needed to consider start of vasopressor therapy as a new episode. Short-lived vasopressor needs, e.g., during procedures/sedation, does not constitute vasopressor-dependent sepsis.

17. Estimated glomerular filtration rate (eGFR) >60 mL/min/1.73 m² based on the most recent available CR sample at time of screening (NOTE: will often be the sample used to diagnose AKI). eGFR should be calculated using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) formula. In Japan, the CKD-EPI formula with Japanese coefficient should be used. If local regulations prohibit correcting for race in the calculation of eGFR, it is acceptable to use the formula without correcting for race.

The mortality rate in patients with an eGFR >60 mL/min/1.73 m² at time of screening is expected to be low and hence these patients are not suitable for a trial with mortality as the primary endpoint.

18. Not feasible to start trial drug within:

a) 48 hours from AKI diagnosis, when AKI diagnosis precedes start of vasopressor therapy.

or

b) 24 hours from AKI diagnosis, when AKI is diagnosed after start of vasopressor therapy.

The intention is to start treatment with trial drug as early as feasible to avoid that irreversible organ damage following prolonged AKI prevents the ability to document an effect of recAP. Time of AKI diagnosis is the timepoint of the serum or plasma CR sample or the end of the urine collection period used to establish the AKI diagnosis. Due to the associated risk of ischemia, the risk of permanent organ damage is higher in patients with sepsis-induced hypotension requiring vasopressor therapy, therefore the time window for enrolment is shorter in patients already on vasopressor therapy at time of AKI diagnosis. Patients with AKI without the need for vasopressor therapy are less severely ill and may not yet be in the ICU or intermediate care unit, therefore, the time window for enrolment of these patients is extended to 48 hours from start of AKI.

19. Pregnant or nursing women.

Reproductive toxicology studies to exclude an effect of trial drug on fetal and postnatal development have not been conducted.