Study Protocol

Prospective, randomised, multicenter trial about the effect of an ajunctive intravenous therapy with sodium selenite (selenase®, double-lblind) and a procalcitonin guided causal therapy (unblinded) on the survival of patients with severe sepsis and septic shock

SISPCT-trial

A clinical trial of the:

Competence Network Sepsis (SEPNET)

Funded by the Federeal Ministry of Education and Research

Grant number: 01 KI 01 06

<table>
<thead>
<tr>
<th>Principal investigator</th>
<th>Biometry / Coordination / Data Management</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prof. Dr. med. Konrad Reinhart</td>
<td>Prof. Dr. med. Markus Löffler</td>
</tr>
<tr>
<td>Dept. of Anesth.y and Intensive Care Med.</td>
<td>Clinical Trial Centre Leipzig</td>
</tr>
<tr>
<td>Jena University Hospital</td>
<td>University Leipzig</td>
</tr>
<tr>
<td>Erlanger Allee 101</td>
<td>Härterstraße 16-18,</td>
</tr>
<tr>
<td>07747 Jena, GERMANY</td>
<td>04107 Leipzig, GERMANY</td>
</tr>
</tbody>
</table>

Sponsor
Friedrich-Schiller-University Jena

EudraCT-No.: 2007-004333-42
INHALTSVERZEICHNIS

1 VISIT SCHEDULE .......................................................................................................................... 3

2 GOALS OF THE TRIAL ................................................................................................................. 4
   2.1 PRIMARY GOAL .......................................................................................................................... 4
   2.2 SECONDARY GOALS ............................................................................................................... 4

3 DESCRIPTION OF THE TRIAL ..................................................................................................... 5
   3.1 STUDY DESIGN ......................................................................................................................... 5

4 STUDY POPULATION .................................................................................................................... 6
   4.1 INCLUSION CRITERIA ............................................................................................................... 6
   4.2 EXCLUSION CRITERIA ............................................................................................................. 8

5 STUDY INTERVENTIONS ............................................................................................................... 9
   5.1 STUDY MEDICATION ............................................................................................................... 9
   5.2 MANUFACTURING AND SHIPPING OF THE STUDY MEDICATION ...................................... 9
   5.3 APPLICATION OF THE STUDY MEDICATION ...................................................................... 9
   5.4 PCT GUIDED CAUSAL THERAPY ......................................................................................... 10

6 INDIVIDUAL COURSE OF THE CLINICAL TRIAL ................................................................... 13
   6.1 CHECKING INCLUSION AND EXCLUSION CRITERIA ......................................................... 13
   6.2 INFORMED CONSENT ........................................................................................................... 13
   6.3 ENROLMENT INTO THE STUDY ............................................................................................ 15
   6.4 STUDY PROCEDURES .......................................................................................................... 15
   6.5 FOLLOW-UP ........................................................................................................................ 15

7 DIAGNOSTIC PROCEDURES AND DATA CAPTURE ................................................................ 16
   7.1 PROCALCITONIN MEASUREMENT ....................................................................................... 16
   7.2 RESPONSE TO THERAPY VISIT .......................................................................................... 16
   7.3 TEST OF CLINICAL CURE ................................................................................................... 17
   7.4 TEST OF MICROBIOLOGICAL CURE ................................................................................... 17

8 ADVERSE EVENTS (AE/SAE) ....................................................................................................... 18
   8.1 ADVERSE EVENTS (AE) ...................................................................................................... 18
   8.2 SEVERE ADVERSE EVENTS (SAE) ...................................................................................... 20

9 BIOMETRY .................................................................................................................................... 20
   9.1 RANDOMISATION .................................................................................................................. 20
   9.2 ENDPOINTS .......................................................................................................................... 20
   9.3 STATISTICAL MODEL OF THE STUDY QUESTION ............................................................... 21
   9.4 DATA ANALYSIS .................................................................................................................. 22
   9.5 INTERIM ANALYSIS ............................................................................................................. 24
   9.6 FINAL ANALYSIS ................................................................................................................ 24
   9.7 ESTIMATE OF THE EFFECT SIZES .................................................................................... 24
   9.8 DROP-OUTS ........................................................................................................................ 24
   9.9 SAMPLE SIZE CALCULATION ............................................................................................. 24

10 SOURCE DATA MONITORING .................................................................................................... 25

11 ADMINISTRATIVE REGULATIONS ............................................................................................ 26
   11.1 STUDY COMMISSION ......................................................................................................... 26
   11.2 INDEPENDENT DATA MONITORING COMMITTEE ............................................................ 26
# VISIT SCHEDULE

<table>
<thead>
<tr>
<th>Study intervention</th>
<th>Days after Randomisation</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>S</td>
</tr>
<tr>
<td>Inclusion and exclusion criteria</td>
<td>X</td>
</tr>
<tr>
<td>Randomization</td>
<td>X</td>
</tr>
<tr>
<td>Blood culture</td>
<td>X</td>
</tr>
<tr>
<td>Demographic data</td>
<td>X</td>
</tr>
<tr>
<td>Diagnoses, coexisting diseases</td>
<td>X</td>
</tr>
<tr>
<td>Surgery</td>
<td>X</td>
</tr>
<tr>
<td>APACHE II-Score / SAPS-II</td>
<td>X</td>
</tr>
<tr>
<td>SOFA-Score</td>
<td>X</td>
</tr>
<tr>
<td>Sepsis criteria, infections</td>
<td>X</td>
</tr>
<tr>
<td>Comedication</td>
<td>X</td>
</tr>
<tr>
<td>Response to Therapy</td>
<td>X</td>
</tr>
<tr>
<td>Test of Clinical Cure</td>
<td>X</td>
</tr>
<tr>
<td>Test of Microbiological Cure</td>
<td>X</td>
</tr>
<tr>
<td>Survival</td>
<td></td>
</tr>
</tbody>
</table>

1 Administration of study medication during the whole ICU stay but max. until day 28

2 PCT group only;

3 if discharged from ICU before day 14.

S: Screening; B: Baseline; D: Discharge from ICU (terminates all study interventions if before day 28)

1× at any time when informed consent is available
2 GOALS OF THE TRIAL

2.1 Primary goal

The primary goal of the trial is to investigate whether an adjunctive therapy with sodium selenite (loading dose 1000 µg of selenium as sodium selenite as 20 min infusion immediately followed by max. 21 continuous intravenous administrations of 1000µg/24h of selenium as sodium selenite) and the guidance of causal sepsis therapy (source control, antimicrobial therapy) by measurement of serum procalcitonin concentrations affects 28 day all cause mortality in patients with severe sepsis or septic shock.

2.2 Secondary goals

The secondary goals aim to investigate

1. whether and to what extend administration of sodium selenite or proclacitonin guided causal therapy has a benefit for single organ systems (lung, circulation, liver, kidney, central nerval system, and coagulation).
2. whether and to what extend the interventions affect hospital and ICU length of stay as well as 90 days all cause mortality.
3. whether and to what extend the interventions affect frequency and duration of mechanical ventilation.
4. whether and to what extend the interventions affect frequency and duration of renal replacement therapy.
5. whether and to what extend the interventions affect frequency and duration of vasopressor therapy.
6. whether the interventions are safe.
7. whether and to what extend the primary outcome and the above mentioned secondary goals 1 – 6 differ in a subgroup analysis including only patients with a survival of at least 48 hours.
8. whether and to what extend procalcitonin guided therapy causes a faster eradication of underlying infection.
9. whether and to what extent procalcitonin guided therapy reduces duration of antimicrobial therapy, costs of antimicrobial therapy, and antibiotic resistancies (VRE, MRSA; ESBL).

10. Continue sampling for the SepNet biomaterial bank.

3 DESCRIPTION OF THE TRIAL

3.1 Study design

This clinical trial is a multicenter prospective randomised bifactorial (2x2 groups) study. Sodium selenite is given in a double-blinded fashion. The procalcitonin guided causal therapy is investigated in an unblinded design.

This clinical trial is a treatment optimising study and is assigned as phase III study.

There is no reason to assume an interaction between the two study interventions and both interventions can be investigated simultaneously in one clinical trial with a bi-factorial study design without increasing the sample size.

3.1.1 Group assignment in the 2x2 factorial design

By randomisation, each patient is assigned to one of four groups

- **Group SelPCT** (Treatment with sodium selenite + guidance of causal therapy with procalcitonin): After study inclusion, 20 ml selenase® T ad 50 ml of 0.9% sodium chloride or – in case of hypernatremia – 5% aqueous glucose solution (corresponds to 1000 µg sodium selenite) is administered intravenously as loading dose over 20 min. This is followed by intravenous continuous administration with 20 ml selenase® T ad 50 ml of 0.9% sodium chloride or – in case of hypernatremia – 5% aqueous glucose solution (corresponds to 1000 µg sodium selenite) until discharge from the intensive care unit but maximally 21 administrations over 24 hours. A procalcitonin-based algorithm for the management of causal sepsis therapy is AVAILABLE for the treating physician.

- **Group SelCon** (Treatment with sodium selenite + no procalcitonin measurement [Control]): After study inclusion, 20 ml selenase® T ad 50 ml of 0.9% sodium chloride or – in case of hypernatremia – 5% aqueous glucose solution (corresponds to 1000 µg sodium selenite) is administered intravenously as loading dose over 20 min. This is followed by intravenous continuous administration with 20 ml selenase® T ad 50 ml of 0.9% sodium chloride or – in case of hypernatremia – 5% aqueous glucose solution...
(corresponds to 1000 µg sodium selenite) until discharge from the intensive care unit but maximally 21 administrations over 24 hours. A procalcitonin-based algorithm for the management of causal sepsis therapy is NOT AVAILABLE for the treating physician.

- **Group PlacPCT** (Treatment with placebo + guidance of causal therapy with procalcitonin): After study inclusion, 20 ml placebo ad 50 ml of 0.9% sodium chloride or – in case of hypernatremia – 5% aqueous glucose solution (corresponds to 1000 µg sodium selenite) is administered intravenously as loading dose over 20 min. This is followed by intravenous continuous administration with 20 ml placebo ad 50 ml of 0.9% sodium chloride or – in case of hypernatremia – 5% aqueous glucose solution (corresponds to 1000 µg sodium selenite) until discharge from the intensive care unit but maximally 21 administrations over 24 hours. A procalcitonin-based algorithm for the management of causal sepsis therapy is AVAILABLE for the treating physician.

- **Group PlacCon** (Treatment with sodium selenite + no procalcitonin measurement [Control]): After study inclusion, 20 ml placebo ad 50 ml of 0.9% sodium chloride or – in case of hypernatremia – 5% aqueous glucose solution (corresponds to 1000 µg sodium selenite) is administered intravenously as loading dose over 20 min. This is followed by intravenous continuous administration with 20 ml placebo ad 50 ml of 0.9% sodium chloride or – in case of hypernatremia – 5% aqueous glucose solution (corresponds to 1000 µg sodium selenite) until discharge from the intensive care unit but maximally 21 administrations over 24 hours. A procalcitonin-based algorithm for the management of causal sepsis therapy is NOT AVAILABLE for the treating physician.

### 4 STUDY POPULATION

#### 4.1 Inclusion criteria

All inclusion criteria have to be fulfilled for inclusion into the study.

1. Presence of severe sepsis or septic shock
2. Onset of severe sepsis or septic shock not longer than 24 hours ago.
3. Age at least 18 years
4. Written informed consent of the patient or the legal representative (see below for further explanations).
4.1.1 Definitions of severe sepsis and septic shock

Severe sepsis

Severe sepsis is defined as the presence of the following 3 criteria

- Diagnosis of infectious origin of infection by one of the following criteria
  - microbiological proof of infection
  - Clinical proof of infection
  - suspected infection

- Diagnosis of Systemic Inflammatory Response Syndrome (SIRS) by at least two of the following criteria
  - Hypo- (36°C) or hyperthermia (38°C)
  - Tachycardia (90/min)
  - Tachypnea (20/min) and/or arterial pCO₂ 4.3 kPa (33 mmHg) and/or mechanical ventilation
  - Leukocytosis 12,000/µl or leukopenia 4,000/µl and/or immature cells 10%

- Organ dysfunction within the last 24 hours (at least one of the following)
  - Encephalopathy
  - Thrombozytopenia (thrombocyte count 100,000/µl or decrease of thrombocytes >30% in 24 hours without evidence of bleeding)
  - Arterial hypoxemia (paO₂ < 10 kPa (75 mmHg) when breathing room air, paO₂/FiO₂ 33kPa (250 mmHg) not caused by pulmonary or cardial disorder)
  - Arterial hypotension (systolic blood pressure ≤ 90 mmHg or mean blood pressure ≤ 70 mmHg) for at least 1 hour despite adequate fluid resuscitation; other types of shock
  - Renal dysfunction (urinary output 0.5 ml/kg/h for at least 1 hour despite sufficient fluid resuscitation and/or increase of serum creatinine 2 above the reference range)
  - Metabolic acidosis (base deficit 5.0 mEq/l or serum lactate concentration 1.5
above the reference range)

**Septic shock**

- Diagnosis of infectious origin of infection as defined above
- Diagnosis of SIRS as defined above
- Arterial hypotension (systolic blood pressure $\leq 90$ mmHg or mean blood pressure $\leq 70$ mmHg) for at least 2 hours despite adequate fluid resuscitation which requires the administration of vasopressors (dopamine 5 µg kg$^{-1}$ min$^{-1}$; norpeinpehrine or epinephrine 0.05 µg kg$^{-1}$ min$^{-1}$; phenylephrine or vasopressin in any dosage) to maintain systolic blood pressure 90 mmHg or mean systolic pressure 70 mmHg.

### 4.2 Exclusion criteria

1. Pregnant or lactating women
2. Fertile female patients (<2 years after last menstruation) without appropriate contraception during study participation.
3. Participation in a clinical study within the last 30 days.
4. Current participation in another study or clinical trial.
5. Previous participation in this clinical trial.
6. Selenium intoxication
7. No commitment to full patient support (i.e. DNR-order)
8. Patient’s death is considered imminent due to coexisting disease
9. Relationship to the study team (i.e., colleague, relative, employee)
10. Infection where guidelines recommend a longer duration of antimicrobial therapy: (1) Infections with *Listeria* spp, *Legionella pneumophila*, *Pneumocystis jiroveci* or *Mycobacterium tuberculosis*; (2) viral or parasitic infections (hemorrhagic fever, malaria); (3) bacterial endocarditis, brain abscess, bone or deep soft tissue infections, (4) chronic local infections (osteomyelitis).
11. Severely immunocompromised patients (i.e. HIV-infection with CD4 count <200 cells/mm$^3$, neutropenic patients (<500 neutrophils/mm$^3$), or patients with immunosuppressive therapy after solid organ transplantation)
5 STUDY INTERVENTIONS

5.1 Study medication

A bottle with 20 ml solution contains either:

1.000 µg selenium as sodium selenite-pentahydrate in 0.9% aqueous sodium chloride
(selenase® T pro injectione)

or

0.9% aqueous sodium chloride (placebo)

Other components:

sodium, water, hydrochloric acid (not in placebo).

5.2 Manufacturing and shipping of the study medication

The study medication is manufactured, packaged, and labeled by biosyn Arzneimittel GmbH,
Schorndorfer Straße 32, 70734 Fellbach, Germany. The study medication is manufactured
according to the German legislation and the Good Manufacturing Practice (GMP) according
to the latest edition. Drug accountability is documented in the eCRF. It is necessary to
reproduce conformity of shipped, used, and returned study medication at the end of the trial.

5.3 Application of the study medication

5.3.1 Dosage

The study medication has to be administered within 2 hours after inclusion into the study.

The study medication is administered as an intravenous bolus injection followed by a
continuous infusion. At the beginning of the study, 1000 µg selenium (selenase® T) or
placebo equivalent to one bottle of the study medication are dissolved in 0.9% sodium
chloride (or 5% glucose) to a total of 50 ml and given intravenously over 20 min. Then,
1000 µg selenium (selenase® T) or placebo equivalent to one bottle of the study medication
are administered over 24 hours. Likewise, one bottle of the study medication is dissolved in
0.9% sodium chloride (or 5% glucose) to a total of 50 ml. The drug is given with a syringe
pump over 24 hours per dose (2 ml/h).
The study medication is given for the whole ICU stay but maximally until day 21. The application of the study medication can stopped earlier if a central venous catheter is no longer indicated.

### 5.3.2 Prohibited concomitant medication

Following medication is not permitted during the application of sodium selenite:

- Additional selenium administration > 100 µg per day
- Administration of the following antioxidants:
  - Vitamine C: > 500 mg per day
  - Vitamien E: > 400 i.E. per day

A substitution with vitamin combinations during parenteral nutrition is permitted as long as it complies with the guidelines of the German Association for Nutritional Medicine. Only the recommended daily dose of such a vitamin combination may be administered once a day.

### 5.4 PCT guided causal therapy

#### 5.4.1 Control arm

Patients with procalcitonin measurement before the screening process can be included into the study.

In the control group (SelCon + PlacCon), blood samples are obtained for later measurement of serum procalcitonin measurements. The measurements occur after finalisation of the data capture for this trial. Thus, procalcitonin values are not available for the treating physicians. Clinical data are recorded in the CRF as required by the study protocol. Changes in causal therapies are at the discretion of the treating physicians. The treating physician is asked about continuation of antimicrobial therapy at the clinical cure visits; the physician has to give reasons for the decision. A clinical cure visit occurs at days 4, 7, 10, and 14 if the patient is still on the ICU. A microbiological cure visit occurs at the same days.

#### 5.4.2 Procalcitonin group

In this group, results of the PCT-measurements are available for the treating physicians within 4 hours. Depending on the individual PCT concentration or changes in the individual PCT concentration from baseline to day 4, from day 4 to day 7, from day 7 to day 10, and from
day 10 to day 14, the protocol recommends a change in the causal therapy (Table 1). The
treating physician does not have to follow the recommendation but has to justify deviations
from the recommendation. A clinical cure visit occurs at days 4, 7, 10, and 14 if the patient
is still on the ICU. A microbiological cure visit occurs at the same days. The highest PCT
concentration from the measurements at randomization and day 1 is defined as the baseline
PCT concentration.
Table 1: Algorithm to guide causal therapy with procalcitonin

<table>
<thead>
<tr>
<th>Days after randomization</th>
<th>Observation</th>
<th>Recommended intervention</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Day 4</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>PCT-decrease ≥50 % from baseline(^1)</td>
<td>No change</td>
</tr>
<tr>
<td></td>
<td>PCT-decrease &lt;50 % from baseline(^1)</td>
<td>Change or optimization (i.e. dosage, kind of application, interval) of antimicrobial therapy recommended</td>
</tr>
<tr>
<td></td>
<td></td>
<td>New intervention for source control recommended (i.e. surgical or interventional source control, diagnostics, change or removal of intravascular catheters)</td>
</tr>
<tr>
<td><strong>Day 7</strong></td>
<td>PCT ≤1,0 ng/ml</td>
<td>Discontinuation of antimicrobial therapy recommended(^2)</td>
</tr>
<tr>
<td></td>
<td>PCT &gt;1,0 ng/ml and PCT decrease ≥50 % from day 4</td>
<td>Discontinuation of antimicrobial therapy recommended(^2)</td>
</tr>
<tr>
<td></td>
<td>PCT &gt;1,0 ng/ml and PCT decrease &lt;50 % from day 4</td>
<td>Change or optimization (i.e. dosage, kind of application, interval) of antimicrobial therapy recommended</td>
</tr>
<tr>
<td></td>
<td></td>
<td>New intervention for source control recommended (i.e. surgical or interventional source control, diagnostics, change or removal of intravascular catheters)</td>
</tr>
<tr>
<td><strong>Day 10</strong></td>
<td>PCT ≤1,0 ng/ml</td>
<td>Discontinuation of antimicrobial therapy recommended(^2)</td>
</tr>
<tr>
<td></td>
<td>PCT &gt;1,0 ng/ml and PCT decrease ≥50 % von Tag 7</td>
<td>Discontinuation of antimicrobial therapy recommended(^2)</td>
</tr>
<tr>
<td></td>
<td>PCT &gt;1,0 ng/ml and PCT decrease &lt;50 % from day 7</td>
<td>Change or optimization (i.e. dosage, kind of application, interval) of antimicrobial therapy recommended</td>
</tr>
<tr>
<td></td>
<td></td>
<td>New intervention for source control recommended (i.e. surgical or interventional source control, diagnostics, change or removal of intravascular catheters)</td>
</tr>
</tbody>
</table>

\(^1\) Highest PCT-concentration from baseline or day 1

\(^2\) BeiFor the following infections which necessitates antimicrobial therapy for more than 7 days, antimicrobial therapy should be continued according to clinical decision: 1) infections with Listeria spp, Legionella pneumophila, Pneumocystis jiroveci or Mycobacterium tuberculosis; 2) viral or parasitic diseases (hemorrhagic fever, malaria); 3) bacterial endocarditis, brain abscess, deep bone or soft tissue abscesses; 4) chronic localized infections (osteomyelitis); 5) severe immunocompromised patients (i.e. HIV-infection with CD4 count < 200 cells/mm\(^3\); neutropenic patients (<500 neutrophils/mm\(^3\)); or patients with immunosuppressive therapy after solid organ transplantation.
Day 14

<table>
<thead>
<tr>
<th>Condition</th>
<th>Action</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCT (\leq 1.0) ng/ml</td>
<td>Discontinuation of antimicrobial therapy recommended(^2)</td>
</tr>
<tr>
<td>PCT &gt;(1.0) ng/ml and PCT decrease (\geq 50%) from day 10</td>
<td>Discontinuation of antimicrobial therapy recommended(^2)</td>
</tr>
</tbody>
</table>
| PCT >\(1.0\) ng/ml and PCT decrease <50 % from day 10 | - Change or optimization (i.e. dosage, kind of application, interval) of antimicrobial therapy recommended  
- New intervention for source control recommended (i.e. surgical or interventional source control, diagnostics, change or removal of intravascular catheters) |

6 INDIVIDUAL COURSE OF THE CLINICAL TRIAL

6.1 Checking inclusion and exclusion criteria

The check of the inclusion and exclusion criteria of potential study patients (screening) is carried out with a check list. All patients fulfilling all inclusion criteria are documented in the screening log. If a patient fulfills all inclusion but no exclusion criteria (eligible patient), it has been documented in the screening log, whether the patient was included into this study (enrolled). If the patient was not included into the study, the reason has to be documented in the screening log.

6.2 Informed consent

6.2.1 Patients capable of giving informed consent

Before randomization, each patient receives written informations about this trial regarding importance, goals, and possible risks of the study. A study physician also discusses the study with the patient. The informed consent is signed in duplicates if the patient decides to participate in the trial. The patient receives one copy and the other copy is stored in the investigator site file.

6.2.2 Patients not capable of giving informed consent

It has to be expected that most eligible patients will not be capable of giving informed consent because of the severity of the disease. However, early intervention is necessary to demonstrate a beneficial effect for these patients.
An already existing legal representative can sign the informed consent in consideration of the
presumed patient’s will. Otherwise, a legal representative has to be identified and appointed.
However, it is expected that an informed consent from a legal representative frequently
cannot be obtained in due time. Since therapeutic interventions in patients with severe sepsis
or septic shock have to be initiated as soon as possible, it is justified according to the
German Drug Act that as a start the patient can be enrolled into the study without informed
consent. In this case, the circumstances of such an enrolment have to be confirmed by an
independent physician. The independent physician must not be involved in this study or
employed in the department responsible for this trial. A legal representative has be appointed
and informed consent of this legal representative has to be obtained as soon as possible.
Other procedure might be defined by the local ethics committees.

The study centers shall comply to the local procedure defined by their local ethics
boards regarding the informed consent process of patients inacapable of giving
informed consent. If a legal representative needs to be appointed, this should occur as
soon as possible. It is the resposiblility of the local investigator to identify the
appropriate person and to induce the application at court. If the legal representative
disagrees with the study participation, the trial and the data capture has to be stopped
immediately in this patient. All blood samples have to be destroyed. All data recorded
until then can still be used to

a) assess the effect of the study drug

b) assure that the safety of the patient is preserved

c) to allow the submission of complete files for drug licensing

It is aimed for that patients not capable of giving informed consent are informed later if they
become contractually capable during their hospital stay. If the patient then decides not to
participate in the study, any samples from the study are destroyed. Data collected until then
can be used as mentioned above if applicable.

6.2.3 Withdrawl of consent

Patients or their legal representatives can always withdraw their consent and abort their
participation in the study without justification. The patient is informed that the saved data can
still be used to
• assess the effect of the study drug
• assure that the safety of the patient is preserved
• to allow the submission of complete files for drug licensing

6.3 Enrolment into the study

6.3.1 Randomization
Randomization occurs by an internet-based service.

6.3.2 Violation of inclusion or exclusion criteria after enrolment
Violation of inclusion or exclusion criteria is in general no reason for discontinuing the study intervention in this patient. If it is observed that a violation of inclusion or exclusion criteria was present during recruitment of the patient, the medical study coordinator has to be informed as soon as possible. The medical study coordinator decides whether study interventions should be stopped. Documentation has to be continued in any case.

6.4 Study procedures
The daily visits until day 21 according to the visit schedule have to be undertaken only as long as the patient is treated on the intensive care unit. The occurrence of AEs, SAEs and SUSARs have to be documented outside of the visit schedule, as well.

6.5 Follow-up
Whereabouts of the patient and survival status are recorded on day 28 and day 90. In addition, ICU and hospital length of stay, frequency and duration of vasopressor therapy, mechanical ventilation, and renal replacement therapy are documented.
7 DIAGNOSTIC PROCEDURES AND DATA CAPTURE

7.1 Procalcitonin measurement

PCT is measured locally at the regional study centers. In general, 5 ml of EDTA blood or heparinized plasma is necessary according to the requirements of the local laboratory. The study centers have to provide certificates of interlaboratory tests for the used devices.

In this clinical trial, all measurement techniques are permitted which comply to the following criteria:

- Measurement range of 0.1 – 200 ng/ml
- Functional assay sensitivity of at least 0.06 ng/ml
- Lower detection limit of at least 0.1 ng/ml

Beside the local PCT measurement which are performed in the PCT-guidance group only, PCT is measured after the trial in all patients at baseline, day 1, day 4, day 7, day 10, and day 14 in a central laboratory. For this purpose, 5 ml of EDTA blood are withdrawn and centrifuged, pipetted into 2 ml new Screw microtubes (NeoLab) and frozen at -20 °C.

7.2 Response to Therapy Visit

The Response to Therapy Visit is a secondary endpoint of this study and is documented on day 4, 7 10, and 14 if the patients is still on the intensive care unit. If the patient leaves the ICU before day 14, an additional Response to Therapy Visit is performed at the discharge day.

7.2.1 Criteria

<table>
<thead>
<tr>
<th>Response to Therapy</th>
<th>Recovery of all clinical signs and symptoms of infection and a Clinical Pulmonary Infection Score (CPIS) ≤1 in patients with pneumonia as focus of infection.</th>
</tr>
</thead>
<tbody>
<tr>
<td>“yes“</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Response to Therapy</th>
<th>No recovery of all clinical signs and symptoms of infection or a CPIS &gt;1 in patients with pneumonia as focus of infection.</th>
</tr>
</thead>
<tbody>
<tr>
<td>“no“</td>
<td></td>
</tr>
</tbody>
</table>


7.3 Test of Clinical Cure

The Test of Clinical Cure is documented on day 4, 7, 10, and 14 if the patient is still on the intensive care unit. If the patient leaves the ICU before day 14, an additional Test of Clinical Cure is performed at the discharge day.

7.3.1 Criteria

<table>
<thead>
<tr>
<th></th>
<th>Cure</th>
<th>Improvement</th>
<th>Treatment failure</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1. Recovery from sign and symptoms of infection and</td>
<td>1. Signs and symptoms of infection have improved and</td>
<td>1. Signs and symptoms of infection have improved and</td>
</tr>
<tr>
<td></td>
<td>2. No antibiotic therapy for the infection of interest</td>
<td>2. No antibiotic therapy for the infection of interest</td>
<td>2. No antibiotic therapy for the infection of interest</td>
</tr>
</tbody>
</table>

7.3.2 Pneumonia

Pneumonia is defined as a Clinical Pulmonary Infections Score of at least 6 points in combination with pulmonary infiltration in the chest x-ray. Test of clinical cure for pneumonia is defined as follows:

<table>
<thead>
<tr>
<th></th>
<th>Cure</th>
<th>Improvement</th>
<th>Treatment failure</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1.) No pulmonary infiltration in the chest x-ray and CPIS ≤1;</td>
<td>1.) Regressive pulmonary infiltration in the chest x-ray and a CPIS between 2 and 5;</td>
<td>Persistent pulmonary infiltration in the chest x-ray, or a CPIS ≥6 or a new antimicrobial necessary to treat pneumonia.</td>
</tr>
<tr>
<td></td>
<td>2.) No new antimicrobial necessary to treat pneumonia.</td>
<td>2.) No new antimicrobial necessary to treat pneumonia.</td>
<td></td>
</tr>
</tbody>
</table>

7.4 Test of Microbiological Cure

The Test of Microbiological Cure is documented on day 4, 7, 10, and 14 if the patient is still on the intensive care unit. If the patient leaves the ICU before day 14, an additional Test of...
Microbiological Cure is performed at the discharge day. At these days, microbiological samples are taken from the primary source of infection if possible.

### 7.4.1 Criteria

| **Documented microbiological eradication** | Cultural clearance of the original pathogen from the original site of infection |
| **Presumed microbiological eradication** | Clinical cure, no appropriate culture material available from the original site of infection |
| **Documented microbiological persistence** | Cultural proof of the original pathogen from the original site of infection |
| **Presumed microbiological persistence** | Clinical treatment failure, no appropriate culture material available from the original site of infection |
| **Relapse** | Documented infection by microbiological culture from the original site of infection at day 21 (end of study) in a patient with previously documented or presumed microbiological eradication. |
| **Superinfection** | Each patient classified as treatment failure or clinical improvement where during antibiotic therapy a pathogen is proven which differs from the original pathogen |
| **Colonization** | Microbiological proof of a pathogen different from the original pathogen in clinically cured patient |
| **undetermined** | Any other condition |

### 8 ADVERSE EVENTS (AE/SAE)

#### 8.1 Adverse events (AE)

#### 8.1.1 Definition

Adverse events are all unfavourable medical occurrences in a patient of a clinical trial where drugs or medical products are used; the event does not have to be causally related to the
study treatment (ICH-Richtlinie E2A).

8.1.2 Documentation

Severe sepsis and septic shock are conditions with high mortality and impairment of several organ systems occurs frequently. For this reason, death of the patient and other sepsis-related occurrences are recorded as clinical result. The documentation of such occurrences is carried out with the daily visit schedules by the assessment of multi organ dysfunction (SOFA-score). These data are included in the safety and efficacy analyses. The report of such an event as AE only occurs if the investigator assumes a relation to the study medication. This rule concerns the following clinical occurrences:

- Death by severe sepsis/septic shock
- Cardiovascular events: administration of vasoactive drugs or hypotension
- Respiratory events: Decrease in PaO₂/FiO₂-ratio, mechanical ventilation, hypoia, ARDS, acute pulmonary dysfunction
- Hepatic events: liver failure or liver dysfunction resulting in an increase of bilirubin levels.
- Renal events: renal failure resulting in an increase of creatinine levels.
- Hematological/coagulation events: coagulopathy, DIC, thrombocytopenia, thrombocytosis
- SIRS criteria: tachypnoea, leukocytosis, leucopenia, hypothermia, hyperthermia, tachycardia

The following side effects have to be documented always as AE independent of a presumed relation to the study medication:

- Occurrence of a new disease after start of study intervention
- Occurrence of symptoms which may be caused by the study medication
- Organ failure which is not related to sepsis (i.e. anaphylaxia, pulmonary embolism)

Recording of AEs begins with start of study medication; documentation ends 3 days after last dosage of the study medication.
8.2 Severe adverse events (SAE)

All adverse events documented as described above which fulfill the following criteria have to be reported to the study office within 24 hours:

- AEs leading to death,
- Lifethreatening AEs,
- AEs necessitating a hospital stay or prolong a hospital stay,
- AEs causing persisting disabilities, or
- causing a congenital abnormality or birth defect.

9 BIOMETRY

9.1 Randomisation

The assignment ratio for the four groups is 1:1:1:1. Randomisation is stratified according to:

- center
- gender
- severity of sepsis (severe sepsis, septic shock)

Randomisation occurs by using the modified minimisation algorithm according to Pocock.

9.2 Endpoints

9.2.1 Primary endpoint

The primary endpoint of the study is the 28-day all cause mortality (number of patients died until day 28 divided by all patients with known survival status at day 28).

9.2.2 Secondary endpoints

1. Mean total SOFA-score and SOFA-subscores during ICU stay but max. until day 21
2. 90-day all cause mortality
3. Frequency and duration of mechanical ventilation until day 90#
4. Frequency and duration of renal replacement therapy until day 90#
5. Frequency and duration of vasopressor therapy until day 90
6. Frequency of AEs and SAEs
7. Clinical Cure and Microbiological Cure on day 4, 7, 10 and 14#
8. Duration of antimicrobial therapy of the infectious epidose present at enrolment; restricted to ICU stay, max. until day 21.##
9. Costs of antimicrobial therapy of the infectious epidose present at enrolment; restricted to ICU stay, max. until day 21.##
10. Duration until adjustment of antimicrobial therapy of the infectious epidose present at enrolment; restricted to ICU stay, max. until day 21.##
11. Antibiotic exposure days; restricted to ICU stay, max. until day 21.##
12. Days alive without antibiotics; restricted to ICU stay, max. until day 21.##
13. Frequencies of antibiotic resistencies (VRE, MRSA, ESBL)##
14. ICU length of stay until day 90#
15. Hospital length of stay until day 90#
16. Frequency of surgical source control##
17. Frequency of diagnostic procedures to identify source of infection##

* procalcitonin intervention only
# ICU/hospital stay primary causing study enrolment only

9.3 Statistical model of the study question

9.3.1 Statistical hypothesis

Hypothesis 1 (H_{Selen}): Administration of sodium selenite (study arms SelPCT and SelKon) versus placebo (study arms PlacPCT and PlacKon) decreases the fraction of patients \( \pi \) who die until day 28.

Hypothesis 2 (H_{PCT}): Procalcitonin guidance (study arms SelPCT and PlacPCT) versus therapy without PCT guidance (study arms SelKon and PlacKon) decreases the fraction of patients \( \pi \) who die until day 28.

For both hypotheses, it is considered that a difference in both directions should be statistically proven (two-sided question). Therefore, the null-hypothesis postulates equality and the alternative hypothesis postulates a difference in both directions:

\[ H_{0, Selen}^{Selenium}: \pi_{PlacPCT+PlacKon} = \pi_{SelPCT+SelKon} \]
\[ H_{A, Selen}^{Selenium}: \pi_{PlacPCT+PlacKon} \neq \pi_{SelPCT+SelKon} \]
\[ H_{0, PCT}^{PCT}: \pi_{PlacPCT+SelPCT} = \pi_{PlacKon+SelKon} \]
\[ H_{A, PCT}^{PCT}: \pi_{PlacPCT+SelPCT} \neq \pi_{PlacKon+SelKon} \]
9.3.2 Significance and power

The level of significance is assigned to $\alpha=5\%$. We aim for a statistical power of $1-\beta=90\%$. No adjustment in the level of significance is planned because of the assumed independence between the two study interventions.

9.4 Data analysis

The detailed procedures of the statistical data analysis will be laid down in a statistical analysis plan.

9.4.1 Planned methods of analysis

The primary hypothesis is assessed by the chi-squared-test. Analysis is undertaken according to the intention-to-treat principle (ITT).

Secondary endpoints are assessed descriptively and by statistical tests. Frequencies are assessed by the chi-squared-test. Continuous data are assessed by variance analysis. Time to event are assessed by Kaplan-Meier-estimates, log-rank-test, and Cox-regression. Estimates are presented with 95% confidence intervals. Multivariate associations are assessed by multivariate regression models (linear, logistic, multinomial Cox).

9.4.2 Analysis population

The confirmatory analysis of the primary endpoint and the secondary endpoints is undertaken according to the ITT-principle on the basis of the Full Analysis Set (FAS). Safety analyses use the Safety Analysis Set (SAS) and sensitivity analyses are based on the Per-Protocol-Set (PPS).

The FAS consists of all patients,

- who were randomized and
- where the consent was not withdrawn.

The safety of the administration of sodium selenite is examined with the Safety Analysis Set SAS$_{SELEN}$ which consists of all patients,

- who were randomized and
- where the treatment with the study medication was started.
The safety of the PCT-guided therapy is examined with the Safety Analysis Set SAS\textsubscript{PCT} which consists of all patients,
- who were randomized
- where causal sepsis therapy was started and
- where at least one PCT concentration was measured (study arms SelPCT and PlacPCT only)

The PPS consists of all FAS patients where no severe protocol deviation occurred. PPS criteria are defined differently for the two interventions.

*Severe protocol deviations for the selenium intervention:*
- Violation of inclusion or exclusion criteria
- Given dosage of the study medication less than 90% of the planned total dosage according to the study protocol.
- Interruption of the administration of the study drug of more than 24 hours at once.

*Severe protocol deviations for the procalcitonin intervention:*
- Violation of inclusion or exclusion criteria
- Lack of both PCT-measurements at baseline and day 1 in the study arms SelPCT and PlacPCT.
- Lack of PCT-measurements at day 4, 7, 10, or 14 in the study arms SelPCT and PlacPCT.
- PCT measurements during the intervention phase in the control group (SelKonv and PlacKonv)

**9.4.3 Pre-planned subgroups**

The primary endpoint 28-day mortality and the following secondary endpoints

- Mean total SOFA-score and SOFA-subscores during ICU stay but max. until day 21
- 90-day all cause mortality
- Frequency and duration of mechanical ventilation until day 90\
- Frequency and duration of renal replacement therapy until day 90\
- Frequency and duration of vasopressor therapy until day 90\
- Frequency of AEs and SAEs
are analyzed in a subgroup of patients which were treated at least 48 hours.

9.5 Interim analysis

In parallel to the second Annual Safety Report (ASR), an interim analysis is performed since the primary endpoint 28-day mortality is an important safety parameter which may be important for a safety analysis. For both interventions, the primary endpoint is analyzed with an early stopping rule defined by using the „alpha spending function“ and calculating the stopping rule according to O’Brien and Fleming. The results of the interim analysis are reported to the Independent Data Monitoring Committee.

9.6 Final analysis

The final analysis is undertaken after finishing recruitment and the 90 days follow-up of 1180 patients. 992 patients including their data (including survival state at day 28) are necessary to assess the study hypotheses.

9.7 Estimate of the effect sizes

The German Prevalence study of the SepNet reported a 28-day mortality of 40 % \(^1\). In a population of patients without the planned study intervention, a 28-day mortality of 40% is assumed \(^2\). A decrease to 30 % is evaluated as clinically relevant and achievable \(^3\).

9.8 Drop-outs

A drop-out rate of 15% is assumed. This will be mainly patients withdrawing their consent.

9.9 Sample size calculation

9.9.1 Sample size calculation

Considering the error size, the randomization ratio, and the expected treatment effects, a sample size of 248 evaluable patients per arm (496 patients per group) is necessary resulting in 992 patients for the entire study (Calculation with PASS 2002 [© 2001 by J.Hintze, NCSS]). Considering the drop-out-rate of 15% as well as a loss of power in the Per-
Protocol-Set, a total sample size of 1180 randomized patients is required (295 patients per arm)

### 9.9.2 Power analysis

The following figure shows the expected statistical power regarding the primary endpoint depending on the sample size. (calculated with PASS 2002 [© 2001 by. J.Hintze, NCSS]):

![Power Analysis Graph](image)

Regarding important secondary outcomes, a statistical power of $\alpha=5\%$, $1-\beta=90\%$ is calculated (n=496 evaluable patients per group, no adjustment for multiple testing):

- Detection of a difference in the mean total SOFA-score by 0.8 points.
- Detection of a difference in length of stay of 3 days.

### 10 SOURCE DATA MONITORING

During the course of the clinical trial, an on-site monitoring is scheduled according to the local recruitment rate, deficiencies in the data quality, and occurrence of severe protocol deviations. The extent of the monitoring visits and source data verification are laid down in the monitoring manual.
11 ADMINISTRATIVE REGULATIONS

11.1 Study commission

The course of this trial is supervised by the SepNet study commission regarding quality of the infrastructure of the research network and regarding the scientific quality of the study. For this purpose, the study commission receives data such as progress of recruitment and completeness of the case report forms. The study commission is an elected institution consisting of the SepNet management board and directors of five study centers.

11.2 Independent Data Monitoring Committee

The Independent Data Monitoring Committee (IDMC) is an independent commission outside of the SepNet. The safety analysis, which is undertaken after recruitment of 590 patients, is presented to the IDMC for assessment. The IDMC may recommend the discontinuation of one study group or the entire study.

References

