Immunoglobulin for necrotising soft tissue infections (INSTINCT): protocol for a randomised trial

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ABSTRACT

INTRODUCTION: Necrotising soft tissue infections (NSTI) are aggressive infections that can result in severe disability or death. Intravenous polyspecific immunoglobulin G (IVIG) is used as supplementary treatment for patients with NSTIs. The level of evidence is very low, but suggests that IVIG may have beneficial effects. However, IVIG may also have adverse effects. With this trial we will estimate the effects of IVIG on a patient-reported outcome and other patient-centred outcomes in patients with NSTI.

METHODS: INSTINCT is a randomised, double-blinded, parallel-group, placebo-controlled trial with concealed allocation of patients with NSTI 1:1 to IVIG or an equal volume of 0.9% saline. Patients are recruited at Rigshospitalet, Denmark. The primary outcome is the physical component summary score of the Medical Outcomes Study 36-Item Short-Form Health Survey as assessed six months after randomisation. Secondary outcomes are: mortality; time to resolution of shock; bleeding; sequential organ failure assessment scores on days 1-7; use of renal-replacement therapy, mechanical ventilation and vasopressors; days alive and out of hospital; amputation; and severe adverse reactions.

CONCLUSION: This study will be the only completed trial testing IVIG for NSTI, thereby providing important data on a severely sick patient group.

FUNDING: The trial is supported by CSL Behring in the form of trial medication and a € 92,182 grant for trial conduct, research, nurse salary and statistical analyses.

TRIAL REGISTRATION: The trial is registered with clinicaltrials.gov (NCT02111161).

Necrotising soft tissue infection (NSTI), formerly known as necrotising fasciitis, is an aggressive progressing bacterial infection that often results in severe morbidity (e.g. organ failure and amputation) and is associated with high mortality rates (20-40%) [1]. Some of the patients have comorbidities predisposing for NSTI [2]. The most common microbiological finding is mixed flora or group-A streptococcus (GAS) infection [3].

The cornerstones in the treatment of NSTI are extensive surgery (see **Figure 1**), general resuscitation and broad-spectrum antibiotics [4]. Furthermore, hyperbaric oxygenation therapy (HBO) and/or intravenous polyspecific immunoglobulin G (IVIG) are used at many centres specialised in treating NSTI [5], and are part of the treatment protocol at Rigshospitalet (RH), Denmark, to which most severe NSTIs in Denmark are referred.

In patients with streptococcal toxic shock syndrome (STSS), low levels of neutralising superantigen antibodies have been shown to be present in plasma. Because a vast number of different streptococcal superantigens may trigger STSS, broad antibody specificity may be required to dampen the infection. IVIG has been shown to inhibit GAS in vitro [6]; and in an in vivo study of 15 patients with GAS, Norrby-Teglund et al showed that IVIG inhibited the activity of streptococcal antigens to elicit cytokine production in the plasma that was treated with IVIG [7]. This has led to the theory that IVIG therapy could be beneficial for patients with GAS infections.

Only one trial has compared the effect of IVIG with placebo in patients with NSTI. Darenberg et al [8] aimed to evaluate the effect and safety of IVIG as an adjunctive therapy in patients with STSS in a randomised, double-blinded, placebo-controlled trial. Patients were randomly assigned to either IVIG or to an equal volume of 1% albumin. After enrolment of 21 patients, ten receiving IVIG and 11 receiving placebo, the trial was prematurely terminated due to slow enrolment. The primary endpoint was mortality over 28 days, which was 3.6-fold higher in the placebo group than in the IVIG group, but the difference was not statistically significant.

IVIG is generally considered safe, but adverse reactions do occur, the most common being fever [9]. More severe adverse reactions (SAR) such as thrombotic events [9-11], acute kidney injury (AKI) [9] and haemolysis [12] have been described. The US Food and Drug Administration has recently updated the safety information regarding IVIG and the risk of haemolysis due to cases of severe haemolysis-related kidney injury and disseminated intravascular coagulation [13]. Recent data have strengthened the association between the use of human immune globulin products and thrombosis.

There is increased focus on the use of patient-reported outcomes [14], including health-related quality of life (HRQOL), because it is based on the patient's own perceptions.

Therefore, we designed the INSTINCT trial to assess the effect of IVIG versus placebo on HRQOL, mortality, use of life support and SARs in patients with NSTI to test

PROTOCOL ARTICLE

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FIGURE 1

Patient with necrotising soft tissue infection after massive debridement.
Photo: INFECT, www.fp7infect.eu/.

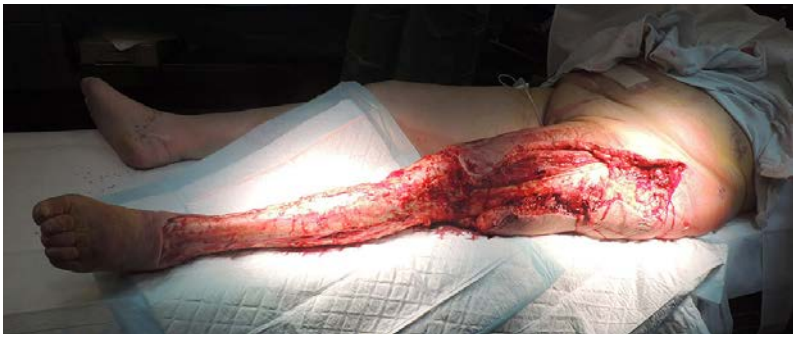

TABLE 1

Inclusion and exclusion criteria.

Inclusion criteria
NSTI infection based on surgical findings
Aged ≥ 18 yrs
Admitted to or planned to be admitted to the ICU at RH
Exclusion criteria
> 48 h from the primary diagnosis to arrival at RH
> 1 dose of IVIG given within current admission
Known hypersensitivity to IVIG
Hyperprolinaemia
Pregnancy or breast feeding

ICU = intensive care unit; IVIG = intravenous polyspecific immunoglobulin G; NSTI = necrotising soft tissue infection; RH = Rigshospitalet.

the hypothesis that IVIG improves the physical component of the HRQOL.

METHODS

Trial design

INSTINCT is a randomised, double-blinded, parallel-group, placebo-controlled trial with concealed allocation of patients with NSTI 1:1 to IVIG or an equal volume of 0.9% saline.

Selection and withdrawal of patients

Trial inclusion and exclusion criteria are listed in **Table 1**.

Patients have the right to withdraw their consent for participation in the trial at any time and still receive the considered best treatment throughout their hospital stay. Patients may be withdrawn from the trial intervention at any time if consent is withdrawn by the person(s) who gave proxy-consent or by the patient. The person(s) demanding withdrawal from trial intervention will be asked for permission to continue data registration. In case the patient does not prohibit collection of the outcome measures, these will be obtained centrally. Thus, the following types of withdrawal may occur: 1. from intervention only (allowing for all data registration and follow-up); 2. from intervention and further registration (but maintaining already registered data and centralised outcome assessment); and 3. from intervention, further registration, follow-up, and previously registered data.

Only the patient can demand deletion of already registered data and only if the patient has not consented previously. If so, data will be deleted and a new patient will be randomised to ensure that the study reaches the full sample size. Patients who are withdrawn from the trial protocol will undergo follow-up and analysis like the remaining patients. Patients who are transferred to another intensive care unit (ICU) will be followed up for the primary outcome measure.

Randomisation

Randomisation will be stratified according to primary presentation of NSTI (either on the extremities/head/neck or thorax/abdomen/back) as streptococci mainly affect the extremities/head/neck. Two lists for randomisation with variable block sizes have been generated using SAS version 9.4 (SAS institute Inc., Cary, NC, USA). Two separate boxes contain sequentially numbered, opaque and sealed envelopes. Eligible patients are allocated to either the IVIG or the placebo group by dedicated personnel who draw an envelope from either of the boxes dependent on the anatomical site of the infection (extremities/head/neck versus other sites).

Intervention

Trial medicine is given when the patient arrives in the ICU and the following two consecutive days. If appropriate, the first dose of trial medicine may be given in the operating theatre. The trial medicine is either IVIG 25 g/day (250 ml) (Privigen, CSL Behring) or 250 ml of 0.9% saline. The dosage of IVIG is 25g/day for three consecutive days for all patients, which is in accordance with the clinical protocol used at RH. All other interventions are given at the discretion of the treating clinicians in accordance with the clinical protocols in place at RH, including those for the treatment of NTSI (repeated surgical revisions, antibiotics (meropenem, clindamycin and ciprofloxacin) and three sessions of HBO), sepsis and supportive intensive care. If consent is denied, the patient will receive IVIG according to the clinical protocol in place at RH.

Blinding

The trial medicine is prepared in the ICU at the time of patient arrival and the following two consecutive days by an ICU nurse not otherwise involved in the care of the trial patient. The nurse is supervised by another staff member who is also not involved in the care of the pa-

tient. Privigen has a slight yellowish appearance, whereas saline is clear. Furthermore, Privigen is produced in bottles of 50 and 100 ml, respectively. A bottle of Privigen is packed with a bag of saline, containing the same volume, in a black plastic bag that is sealed with a plastic strip identical to the bags used in the Scandinavian Starch for Severe Sepsis/Septic Shock (6S) trial [15]. For each daily dose of trial medicine, a set of three bags (2 × 100 ml and 50 ml) is allocated to the patient. An orange, transparent infusion set (B. Braun, ref. 8700127SP) is inserted into either the Privigen bottle or the bag containing saline, dependent on whether the patient has been allocated to IVIG or placebo. The orange colour of the tubing masks the fluid colour while allowing air bubbles to be seen.

Outcome measures

The primary outcome is the physical component summary (PCS) score of the Medical Outcomes Study 36-Item Short-Form Health Survey (SF-36) as assessed six months after randomisation. SF-36 is a generic, self-administrated general health survey with 36 questions distributed on eight health domain scales: General Health (GH), Physical Functioning (PF), Role Physical (RF), Role Emotional (RE), Social Functioning (SF), Bodily Pain (BP), Vitality (VT) and Mental Health (MH) [16]. The PCS score reflects physical health. It is aggregated from all eight scales; the scales GH, PF, RP and BP contribute the most. A higher score indicates better health. The survey is translated into Danish [17], and its reliability and validity have been assessed in an ICU setting and found satisfactory [18], and SF-36 has previously been recommended as a generic measure in the assessment of critical care patients [19].

All outcome measures are listed in **Table 2**.

Data collection

Data are entered into the web-based electronic case report form (eCRF) from patient records by trial personnel. The eCRF was designed specifically for this purpose by Evolve-It.

To ensure as high a response rate as possible, the SF-36 questionnaire is administrated by telephone interview six months after randomisation. The questionnaire is sent in paper-form before the telephone interview to ease the respondents' understanding of the questions. If the patient does not answer the telephone, repeated contact is tried once weekly the following four weeks.

Safety

Patients are withdrawn from the trial protocol if a SAR or a suspected unexpected serious adverse reaction (SUSAR) occurs. Patients withdrawn from the trial receive the standard protocol treatment for NSTI with the exception of IVIG. A list of SARs is presented in **Table 3**.

TABLE 2

Outcome measures.

Primary outcome measure
PCS score of SF-36 6 months after randomisation
Secondary outcome measures
Mortality at 28, 90 and 180 days
Time to resolution of shock[a]
Severe bleeding as clinical bleeding and use of 3 units of RBC within 24 h at any time in the ICU
Any bleeding in the ICU
Use of blood products: total volumes during the ICU admission
SOFA scores days 1-7, excluding the GCS score
Use of RRT, ventilation and vasopressor in the ICU
Days alive off life support in the 90 days after randomisation
Days alive and out of hospital in the 180-day follow-up period
Amputation, any location, within 180 days
SARs in the ICU

CORTICUS = The Corticosteroid Therapy of Septic Shock; GCS = Glasgow coma scale; ICU = intensive care unit; PCS = physical component summary score; RBC = red blood cells; RRT = renal replacement therapy; SAR = serious adverse reaction; SF-36 = Medical Outcomes Study 36-Item Short-Form Health Survey; SOFA = sequential organ failure assessment.
a) As defined in the CORTICUS study: maintenance of a systolic blood pressure of ≥ 90 mmHg without vasopressor support for 24 h.

TABLE 3

Serious adverse reactions[a] described with the use of trial medicine.

Medicine	Serious adverse reactions
Privigen	Allergic reactions
	Haemolytic anaemia
	Aseptic meningitis syndrome
	Thrombus
	Transmittable agents
	Acute kidney injury
0.9% saline	None expected for the volume given

a) Obtained from the respective summary of product characteristics.

The trial definition of SARs is seen in **Table 4**. Adverse events (AEs) and serious adverse events (SAEs) are not recorded as entities because the majority of ICU patients experience several SAEs during their critical illness. Nevertheless, most SAEs will be captured in the secondary outcome measures (sequential organ failure assessment scores and bleeding).

The trial will be terminated prematurely if new, definite information arises regarding the use of IVIG in patients with NSTI.

Statistics

Sample size estimation

A total of 50 subjects in each group are needed to detect a difference of seven points in PCS score of SF-36 based

TABLE 4

Definition of serious adverse reactions described with the use of trial medication.

Disease	Definition
Haemolytic anaemia	Anaemia with the presence of reticulocytosis, low levels of haptoglobulin, high levels of bilirubin and LDH, as defined by a haematologist
Aseptic meningitis syndrome	Clinical suspicion of meningitis due to ≥ 1 of the following symptoms: fever, headache, neck stiffness, nausea and vomiting *and* Negative result of bacteriological and virological test of CSF
Thrombi	
Myocardial ischaemia	If the patient is diagnosed with AMI: ST-elevation myocardial infarction or non-ST elevation myocardial infarction, or unstable angina pectoris according to the criteria in the clinical setting in question, e.g. elevated biomarkers, ischaemic signs on ECG, clinical presentation *and* The patient receives treatment as a consequence of this: reperfusion strategies, PCI/thrombolysis, or initiation/increased antithrombotic drug treatment, on this day
Cerebral ischaemia	Verified by CT- or MR scan
Intestinal ischaemia	Verified by endoscopy or open surgery
Acute limb ischaemia	Clinical signs *and* Need of open/percutaneous vascular intervention, amputation *or* Initiation/increased antithrombotic treatment
Transmittable agents	If the clinician suspects the patient to have a disease caused by a transmittable agent, i.e. HIV, HBV or HCV, a serologic test will be performed If the test is positive *and* a negative test result taken prior exposure to trial medicine is noted in the patient files, the event will be reported as a SAR
Allergic reactions	
Allergic reaction	Urticaria
Severe allergic reaction	Urticaria *and* Worsened circulation: > 20% decrease in blood pressure or > 20% increase in vasopressor dose *or* Increased airway resistance: > 20% increase in the peak pressure on the ventilation *or* Clinical stridor or bronchospasm *or* Subsequent treatment with bronchodilators
AKI	Incident cases in which the patient, who at baseline did not have AKI[a], develops AKI, according to the KDIGO definition AKI will be defined as KDIGO stage 3 Patients who already are in the need of RRT will not be classified as having AKI

AKI = acute kidney injury; AMI = acute myocardial infarction; CSF = cerebrospinal fluid; ECG = electrocardiography; HBV = hepatitis B virus; HCV = hepatitis C virus; HIV = human immunodeficiency virus; ICU = intensive care unit; KDIGO = Kidney Disease: Improving Global Outcomes [20]; LDH = lactate dehydrogenase; PCI = percutaneous coronary intervention; RRT = renal replacement therapy; SAR = serious adverse reaction.
a) AKI is defined as any of the following: increase in plasma creatinine level to ≥ 353.6 µmol/l or increase in plasma creatinine level to ≥ 3.0 times pre-hospital admission creatinine level or initiation of RRT in the ICU or anuria for ≥ 12 h.

on an expected score of 42 (standard deviation = ± 11) in the control group (data from previous non-published follow-up studies), for a power of 80%, a two-tailed significance level of 0.05 and an expected six-month mortality rate of 20% (based on data from a previous cohort, unpublished).

Analyses

The primary analysis will be regression analysis adjusted for the stratification variable "site of infection" (presumed streptococcal infection) in the intention-to-treat population. The detailed statistical analysis plan is published online and publicly available [20].

Trial conduct and monitoring

The trial is conducted in accordance with the International Conference on Harmonisation/Good Clinical Practise (GCP) guidelines and is monitored by an independent inspector from the GCP Unit, RH, following a mutually agreed monitoring plan.

The first patient was enrolled on 7 April 2014, and we expect to enrol the last patient by February 2016 and close the trial data base seven months later.

Finance

The trial is supported by CSL Behring in the form of trial medication (Privigen) and a € 92,182 grant for trial conduct, research nurse salary and statistical analyses. Payments are due in accordance with a Research Agreement between CSL Behring and RH and are paid into the research account of RH. None of the INSTINCT investigators or the authors of this paper have affiliations to or receive honoraria, other funds or gifts from CSL Behring.

Ethics, approval and registration

The trial adheres to the Helsinki Declaration and to Danish law. Patients assessed for enrolment are unable to give informed consent due to severe illness or as a consequence of treatment (sedation). Patients are enrolled after proxy consent given by two physicians (the trial guardian) followed by delayed consent from next of kin and the patient's general practitioner. The protocol is approved by the Regional Ethics Committee (H-3-2013-204), the Danish Health and Medicines Authority (EudraCT-number 2013-003556-20) and the National Data Protection Agency (30-1173).

Trial registration: The trial is registered with clinicaltrials.gov (NCT02111161).

DISCUSSION

The widespread clinical use and the lack of knowledge regarding the benefits and harms of IVIG in patients with NSTI make this study highly relevant. Even though there is a theoretical rationale for using IVIG in the treatment of some NSTIs, every additional therapy holds the risk of additional adverse effects, thereby potentially harming the patient. Patients admitted to the ICU are particularly vulnerable due to the severity of their disease and extensive polypharmacy.

The strengths of this study include thorough methodology (randomised, double-blinded design and preregistration and publications of the protocol and statis-

tical analysis plan) and the use of patient-reported outcome measures in addition to several outcomes related to potential effects and harms. Even though this is a relatively small, single-centre trial, it will be the only completed trial testing IVIG for NSTI thereby providing very important data on a severely sick patient group.

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CONFLICTS OF INTEREST: Disclosure forms provided by the authors are available with the full text of this article at www.danmedj.dk

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