Efficacy and safety of iloprost in patients with septic shock-induced endotheliopathy—Protocol for the multicenter randomized, placebo-controlled, blinded, investigator-initiated trial

Morten H. Bestle1,2 | Niels E. Clausen3 | Peter Søe-Jensen4 | Klaus T. Kristiansen5 | Theis Lange6 | Pär I. Johansson7 | Jakob Stensballe7,8 | Anders Perner9

Background: In Europe 700,000 new cases of sepsis occur annually and more than 100,000 of these patients die due to multiorgan failure (MOF). We have identified shock-induced endotheliopathy (SHINE) to be associated with development of MOF and mortality. Furthermore, in patients with septic shock those with circulating levels of thrombomodulin (TM) above 10 ng/mL have twice the mortality (56% vs 28%) than those with levels below this level. Pilot studies indicate that infusion of iloprost (1 ng/kg/min) is associated with improved endothelial function in patients with septic shock.

Material and Methods: This is a multicenter, randomized, blinded, investigator-initiated, adaptive phase 2B trial in up to 384 patients with septic shock-induced endotheliopathy defined by TM > 10 ng/mL who are allocated 1:1 to 72 hours continuous infusion of iloprost 1 ng/kg/min or placebo (equal volume of saline). The primary outcome is the mean daily modified Sequential Organ Failure Assessment (SOFA) score in the ICU up to day 90. Secondary outcomes include 28- and 90-day all-cause mortality, days alive without vasopressor in the ICU within 90 days, days alive without mechanical ventilation in the ICU within 90 days, days alive without renal replacement therapy in the ICU within 90 days, numbers of serious adverse reactions, and the number of serious adverse events within the first 7 days.

Discussion: This trial tests the safety and efficacy of iloprost vs placebo for 72 hours in patients with septic shock and SHINE. The outcome measures focus on the potential effect of the intervention to mitigate organ failure.

Trial registration: COMBAT-SHINE trial—EudraCT no. 2019-001131-31—Clinicaltrials.gov: NCT04123444—Ethics Committee no. H-19018258.
1 \ INTRODUCTION

More than 700,000 new cases of sepsis occur annually in Europe and more than 100,000 of these patients do not survive, and in Denmark approximately 1,500–2,000 patients with sepsis die annually. Sepsis is the leading cause of death in general intensive care units (ICU) and is by far the most expensive condition treated in European hospitals, including those in Denmark. In August 2017, The World Health Organization adopted a resolution recognizing Sepsis as a Global Health Priority. Patients with the most severe type of sepsis that is with septic shock have a mortality rate between 30% and 45% and these patients succumb due to multiple organ failure (MOF). Interventions targeting various pathways of the coagulo-, inflammatory, complement and cytokine systems to combat MOF have been investigated for the past 30 years in more than 140 clinical trials including >30,000 patients. Unfortunately, all these trials have failed and no specific therapy to combat MOF in septic shock has been introduced.

We have studied >1,500 patients with various severities of sepsis and identified that the degree of endothelial damage, measured by soluble thrombomodulin (TM), was independently associated with development of MOF and death. Importantly, when stratifying the sepsis patients into quartiles, using TM concentration at admission, significant differences in organ failure and mortality were found between the quartiles and can thereby be used prognostically. Furthermore, we also found that sympathico-adrenal overactivation (shock) resulting in, for the endothelium, toxic levels of circulating catecholamines was a driver of this condition, which we entitled shock-induced endotheliopathy (SHINE). Prostacyclin (PGI₂) is an endogenous prostanoid formed and released by endothelial cells with paracrine function including vasodilation and platelet inhibition. Because of these properties it was introduced in 1979 as a pharmacological therapy in high doses for patients with primary pulmonary hypertension and critical limb ischemia. We studied critically ill patients needing renal replacement and found that those who used PGI₂ as anticoagulant in the dialysis filter had substantially lower 30-day mortality than patients receiving heparin (21% vs 39%), despite being more critically ill. We speculated that this may be due to an endothelial cytoprotective effect from the spill over of PGI₂ to the systemic circulation as reported in the literature.

We conducted a clinical trial in healthy volunteers showing that low dose (1 ng/kg/min) of the PGI₂ analogue iloprost (Ilomedin, Bayer) did not increase hypotensive episodes or bleeding complications but instead appeared to improve endothelial functionality as evaluated by circulating TM. The effect of iloprost infusion (1 ng/kg/min) was, thereafter, investigated in randomized, double-blind pilot studies in coronary stent (n = 18), major surgery (n = 56) and septic shock patients (n = 18). These trials showed no increases in the incidences of hypotensive episodes or bleeding or in the transfusion requirements of iloprost infusion, but instead significantly improved endothelial function and integrity, measured by endothelial-derived biomarkers including TM. In septic shock patients, we also found that sequential organ failure assessment (SOFA) score was significantly reduced, as was the time of ventilation, in patients receiving iloprost infusion as compared with placebo. In addition, 30-day mortality in iloprost-treated patients was 8% as compared to 34% in those receiving placebo.

1.1 \ Trial hypotheses

We hypothesize that continuous infusion of low-dose iloprost, ie 1 ng/kg/min, for 72 hours after randomization as compared with placebo in patients with septic shock-induced endotheliopathy, defined by a circulating level of TM > 10 ng/mL, will reduce the mean daily SOFA score in the intensive care unit (ICU) up to 90 days post-randomization.

2 \ MATERIAL AND METHODS

2.1 \ Trial design

The COMBAT-SHINE trial is an investigator-initiated, pragmatic, randomized, blinded, parallel-group, adaptive phase 2b/3 trial allocating adult patients with septic shock-induced endotheliopathy, defined by circulating TM > 10 ng/mL at the time of inclusion, investigating the efficacy and safety of continuous intravenous administration of iloprost (1 ng/kg/min) vs placebo for 72 hours, in a total of up to 380 patients. After inclusion and 90-day follow-up of 200 patients, an interim analysis will explore if adequate effect on the mean daily SOFA score has been observed. Only if adequate effect is observed at that time the trial will continue and enrol the additional 180 patients.

2.2 \ Registration

The trial was registered at the European Union Clinical Trial Register (EudraCT no. 2019-001131-31 approved June 21, 2019) and at ClinicalTrials.gov (Identifier: NCT04123444) before inclusion of the first patient, which occurred on the 3rd of November 2019.

2.3 \ Setting

ICUs in the Capital Region of Denmark admitting adult patients. A complete list of participating trial sites is available at ClinicalTrials.gov (Identifier: NCT04123444).

2.4 \ Study population

2.4.1 \ Inclusion criteria

Adult patients with septic shock defined as (a) suspected or documented infection, and (b) persisting hypotension requiring
vasopressors to maintain MAP ≥ 65 mm Hg and a lactate level >2 mmol/L despite fluid therapy and circulating TM > 10 ng/mL.

2.4.2 | Exclusion criteria

Patients are not eligible for inclusion in this trial if they fulfil one or more of the following criteria:

1. Withdrawal from active therapy
2. Pregnancy (non-pregnancy confirmed by a negative urine or plasma hCG in age below 60 years)
3. Known hypersensitivity to iloprost or to any of the other ingredients.
4. Life-threatening bleeding as defined by the treating physician
5. Known severe heart failure (NYHA class IV)
6. Suspected acute coronary syndrome
7. Previously included in this trial
8. Screened later than 12 hours after diagnosis of septic shock
9. Informed consent cannot be obtained
10. Included in other clinical trials with prostacyclin within 90 days

Patients enrolled in other interventional trials will not be excluded unless the protocols of the two trials collide. A co-enrolment agreement will be established between the sponsors.

2.5 | Screening

All patients admitted to a participating clinical trial site is considered for participation. Experienced ICU nurses screen patients for septic shock at least two times a day. When an adult patient at the ICU is diagnosed with septic shock, the patient is screened for eligibility of enrolment by local investigators using paper CRF. The distribution of screened patients will be displayed in a Consolidated Standards of Reporting Trials (CONSORT) diagram.

2.6 | Randomization

The randomization will be done in permuted blocks of variable sizes stratified for trial site using centralized, concealed allocation. The randomization sequence will be 1:1 (active:placebo) using the online randomization software 'Sealed Envelope' (https://www.sealedenvelope.com/). Once generated the randomization sequence will be formatted and uploaded into REDCap to facilitate centralized, web-based allocation. The patient randomization at each site will be performed in the electronic system REDCap, where each patient will be given a unique trial ID number. The randomization sequence will be concealed from all clinicians, patients, investigators and statisticians and will first be opened after completion of all trial-related procedures and statistical analyses are finalized.

2.7 | Trial intervention

Enrolled patients are allocated to receive either continuous infusion of intravenous iloprost (Ilomedin®, Bayer AG) at a dose of 1 ng/kg/min or placebo (sodium chloride® 9 mg/mL) equal volume. The intervention period will be for 72 hours from randomization. If a trial participant is discharged from the ICU before the 72-hour infusion has been completed, the trial medication is discontinued.

2.8 | Outcome measures

2.8.1 | Primary outcome measure

The primary outcome is the mean daily modified Sequential Organ Failure Assessment (SOFA) score (Appendix S1), involving respiratory, coagulation, liver, cardiovascular and renal function in the intensive care unit up to day 90 (scores for each of five systems range from 0 to 4, with higher scores indicating more severe dysfunction; the maximum score is 20).

2.8.2 | Secondary outcome measures

- 28 and 90-day mortality (note that 90-day mortality is also included in the hierarchical testing procedure and the type-I error rate is therefore protected at 5% for both the primary outcome and 90-day mortality).
- Days alive without vasopressor therapy in the ICU within 90 days
- Days alive without ventilator therapy in the ICU within 90 days
- Days alive without renal replacement therapy in the ICU within 90 days
- Total number and numbers of patient with one or more serious adverse reactions within the first 7 days
- Total numbers and numbers of patients with one or more serious adverse events within the first 7 days (SAE is defined as ischaemic events and bleeding events (defined as requiring >2 RBCs within 24 hours or on-going bleeding)).

2.9 | Blinding

Iloprost is a colourless solution contained in an ampoule which will be diluted according to weight into isotonic saline 0.9%. The solution will be prepared as described in the Danish SmPC. Saline 0.9% (sodium chloride, which is a marked product) is used as placebo in this trial. The two solutions (iloprost in saline and saline) appear identical. The preparation will be done by an unblinded research assistance, who will be responsible for preparing the investigational drug in standard infusion set so that it can be administered blinded via a pump. The preparation will be verified by two persons not involved in the care of the patients.
2.10 | Data registration and monitoring

Data will be entered into a central web-based electronic case report form (eCRF) using the data management system REDCap® software (REDCap 8.10.18 - © 2019 Vanderbilt University). The eCRF is password protected, audit-trailed, encrypted and allows for detailed centralized and de-centralised surveillance of data completeness overall and at each site. Each participating trial site will only have access to their own data. Details and definitions of collected data are presented in Appendix S2.

2.11 | Serious adverse reactions and serious adverse events

Known adverse reactions are specified in Section 4.8 of the summary of product characteristics of iloprost and listed in Appendix S2. We consider the following conditions possibly related to the intervention as they are mentioned in the SmPC; the final adjudication as SARs will be done by the investigators:

1. Bleeding events (intracerebral haemorrhage (verified by CT scan)) and lower gastrointestinal bleeding (defined as bloody diarrhoea and rectal bleeding)
2. Severe cardiac failure (defined as severe cardiogenic shock and ejection fraction <20% cardiac ultrasound)
3. Pulmonary embolism (symptomatic and verified by CT scan)
4. Deep vein thrombosis (symptomatic and verified by ultrasound scan)

SARs will be evaluated and recorded daily in the eCRF until day 7. The distribution of SARs will be compared by the DMSC at interim and final analyses. Suspected Unexpected Serious Adverse Reactions (SUSARs) are defined as SARs not described in the summary of product characteristics of iloprost. Trial investigators will report SUSARs to the sponsor within 24 hours, and further reporting to national health authorities is done by the sponsor within 7 days. On a yearly basis, the sponsor will submit a safety report of all reported SARs and SUSARs to the Danish Medicines Agency and Ethics Committee.

2.12 | Approvals

The trial is approved by the Danish Medicines Agency (EudraCT no. 2019-001131-31), the Committees on Health Research Ethics in the Capital Region of Denmark (H-19018258) and the Danish Data Protection Agency (P-2019-298). All patients will be enrolled after consent from a scientific guardian who is independent of the trial.

2.13 | Statistics and interim analysis

This trial involves three hypotheses as part of a hierarchical testing procedure, which control the family-wise error rate (ie combined risk of a Type-I error) at 5%. Significant findings in these three hypotheses can therefore all be viewed as primary findings of the trial. Other secondary outcomes will be tested separately and have the usual interpretation.

The primary outcome will be assessed using a simple ANCOVA adjusted for baseline SOFA score. Significance will be assessed at the 5% level. Effects will be described as adjusted change in means post-baseline daily SOFA scores along with a 95% confidence interval. Results of the SOFA score will be reported in detail in the five organs systems scores in a supplement to the main paper. For the interim analysis, the null hypothesis will be tested: the mean daily SOFA score in the iloprost group is identical to the mean daily SOFA score in the placebo group plus 0.5 units against the alternative that the mean daily SOFA score in the iloprost group is higher to the mean daily SOFA score in the placebo group plus 0.5 units. All other tests will be ordinary two-sided superiority tests.

Twenty-eight- and 90-day survival will be compared using Fisher's exact test and effect sizes expressed as risk ratios with confidence intervals. Other secondary end-points will be compared using Wilcoxon test and differences expressed as changes in medians with non-parametric based bootstrap with a 95% confidence interval.

2.13.1 | Significance

The hierarchical procedure will be conducted at 5% significance level (family-wise for all three hypotheses). All secondary end-points will be assessed at 5% significance level without any adjustment for multiple testing. The wording of the conclusions will reflect this. The manuscript will highlight if one or more of the secondary endpoints are also significant after Bonferroni-Holm correction for multiple testing. Even if the trial is stopped early all secondary endpoints will be assessed following the procedure described.

2.13.2 | Sample size estimation

Patients will be recruited in a 1:1 ratio (iloprost:placebo). The number of patients participating is based on a power calculation using the data on mean daily SOFA score from a recent randomized clinical trial in patients with septic shock: Levosimendan for the prevention of acute organ dysfunction in sepsis (LeoPARD). The mean daily SOFA score in the control group in this trial was 6.68 with a standard deviation (SD) of 3.9. If the true effect of the intervention is a reduction in mean daily SOFA score of 20% (relative) and providing the trial with 90% power to detect this difference at a significance level of 0.05 will require a sample size of 380 patients. Assuming a true reduction in mean daily SOFA score of 20% then there is a 4.3% probability of (falsely) stopping the trial at the interim analysis after 200 analysed patients. For comparison we have a 75.6% probability of stopping the trial early if there was in fact no effect.
2.14 | Populations and subgroups

Definitions of trial populations are as follows:

Intention-to-treat: This will comprise all randomized patients (except those randomized in error and who never received the trial medication). This population will be evaluated for all endpoints.

Per-protocol: This is a subset of the intention-to-treat population encompassing correctly included patients who have received the trial medication according to protocol (ie 72 hours infusion of iloprost or placebo after inclusion or until dead or discharged to ward, whichever comes first). This population will be evaluated for the primary endpoint only.

Safety population: This comprises all randomized patients including those who are withdrawn.

Three subgroup analyses are planned: One evaluating sTM (high vs low sTM, high defined as sTM > 16.5 ng/mL), one evaluating high vs low SMS score (where high SMS score is defined as > 25 which predict a 90-day mortality risk of 50%) and finally one evaluating short vs long-time period from inclusion to start of intervention (where short time is defined less than 6 hours). For all subgroup analyses effect measures on all outcomes will be computed along with P-values and confidence intervals. For each subgroup and patient outcome, a test for no-treatment heterogeneity will also be reported.

2.15 | Trial organization and management

The COMBAT-SHINE trial is performed at the Departments of Intensive Care at Rigshospitalet, Nordsjælland, Herlev, Hvidovre and Bispebjerg Hospitals, Section of Biostatistics and Section of Health Service Research at the University of Copenhagen and the SHINE Group at Rigshospitalet. The Management Committee encompasses the local principle investigators and a clinical data and regulatory manager at SHINE Group and is responsible for the overall management and coordination, which will be supervised by the Steering Committee. Site investigators will manage and coordinate the trial at the sites. The principal investigator is responsible for data collection and maintenance of trial documents. Co-enrolment of participants in other interventional trials has to be approved by the COMBAT-SHINE steering committee, but is generally appreciated.

2.16 | Data sharing

The trial results will be submitted to a peer-reviewed open-source international clinical journal. De-identified data will be made publicly available 12 months after 1-year follow-up of the last randomized patient according to the recent ICMJE recommendations.

2.17 | Finances

The COMBAT-SHINE trial has received financial support from Innovation Fund Denmark (8056-00019A) and the Independent Research Fund Denmark (8020-00155B). The funding sources have no influence on trial design and will have no influences on data collection, analysis or reporting.

3 | DISCUSSION

3.1 | Intervention

Septic shock is a global health priority and remains to be the leading cause of death in general ICUs worldwide. The major cause of death in patients with septic shock is development of MOF and despite extensive efforts in the past decades no specific therapeutic agent has been identified to resolve this condition. Intravenous iloprost has been on the market since the 1990s for the treatment of Pulmonary Arterial Hypertension and critical limb ischemia due to its vasodilating and platelet inhibiting effects. The rationale for this trial is that multiorgan failure in sepsis patients is caused by shock-induced endotheliopathy and that an endothelial cytoprotective effect of iloprost has been identified. The 1 ng/kg/min dose of iloprost necessary for the endothelial cytoprotective effect is well below the dose necessary for its vasodilatory and platelet inhibiting effects contributing to the safety of administering the intervention in septic shock patients. Furthermore, the current trial has an adaptive design where an interim analysis after the first 200 patients will enable a sample size estimation based on the updated 2016 criteria for septic shock. The current sample size estimation is based on the 2012 Surviving Sepsis Campaign guidelines of septic shock and this may not correctly reflect current practice. Consequently, an important task for the DMSB will be to assess the relevant sample size for the current trial.

3.2 | Outcome

In sepsis trials overall 28-day or 90-day mortality has been the gold standard for the primary endpoint when evaluating the effect of the intervention tested. In the current trial, however, we have chosen mean daily SOFA scores up to day 90 after randomization as the primary endpoint similar to the trial reported by Gordon and colleagues evaluating the effect of levosimendan in sepsis patients. The reason for using this surrogate endpoint is firstly that it increases the likelihood of a fair evaluation of the interventional effect by considering that a proportion of the patients die of other complications than organ failure where the intervention is not expected to have any effect. Secondly, this approach enables us to evaluate the novel intervention more rapidly have, as fewer patients need to be included in the study to reach the statistical power needed. Hereby, a potentially beneficial or harmful effect of the intervention can be provided for this category of patients who have a high unmet medical need as reported by the World Health Organization.
measures include 28- and 90-day mortality, number of days alive without vasopressor, ventilator and renal replacement therapy within 90 days together with number of SARs and SAEs within 7 days.

3.3 | Strength

The COMBAT-SHINE trial is an investigator-initiated, randomized placebo-controlled trial of iloprost as compared with placebo in patients with septic shock and SHINE defined by sTM > 10 ng/mL. The trial design is based on a stringent methodology, which includes concealed group allocation, blinding to the patient, clinical staff, the investigators, the outcome assessors and the trial statistician. The trial is GCP-monitored and an independent DMSC will be responsible for the interim analysis. Sample size estimations and trial design are based currently on best evidence and an interim analysis will be performed to correct for potential differences in the diagnosis of septic shock between the 2012 and 2016 version of the Surviving Sepsis Campaign Guidelines making the trial relevant and representative of current practice and survival rates.

3.4 | Limitations

Development of multiorgan failure in sepsis patients is a multicellular event where cells in the circulating blood, the endothelium and the cells of the vital organs are involved. The current trial is limited in its scope to investigate the potential role of the endothelial cell in the process leading to MOF and, hence, the current trial can only assess the potential effect of iloprost for this purpose here. Furthermore, although COMBAT-SHINE is a multicenter trial, it only includes Danish ICU’s and the sample size is limited to show effects on organ failure only.

4 | PERSPECTIVE

The trial is conducted with a stringent methodology, which complies with international guidelines for clinical trials and good clinical practice. Being an adaptive phase 2B/3 trial with an interim analysis and using a surrogate primary endpoint ensures both that the correct number of patients are included and that the results become rapidly available to the scientific and regulatory communities.

5 | TRIAL STATUS

The trial is currently recruiting at five active trial sites. The first patient was enrolled in November 2019. The current protocol is version 1.3 dated 30 September 2019. Inclusion of patients is expected to end in 2021.

CONFLICT OF INTEREST

Dr PJ is co-inventor of a patent covering the use of iloprost for the treatment of SHINE and the use of TM as a diagnostic biomarker here.

ORCID

Pär I. Johansson https://orcid.org/0000-0001-9778-5964
Anders Perner https://orcid.org/0000-0002-4668-0123

REFERENCES

1. Suarez De La Rica A, Gilsanz F, Maseda E. Epidemiologic trends of sepsis in western countries. Ann Transl Med. 2016;4:325.
2. Reinhart K, Daniels R, Kissoon N, et al. Recognizing sepsis as a global health priority. N Engl J Med. 2017;377:414-417.
3. Angus DC, van der Poll T. Severe sepsis and septic shock. N Engl J Med. 2013;369:840-851.
4. Kaukonen K-M, Bailey M, Suzuki S, Pilcher D, Bellomo R. Mortality related to severe sepsis and septic shock among critically ill patients in Australia and New Zealand, 2000–2012. JAMA. 2014;311:1308-1316.
5. Holst LB, Haase N, Weterslev J, et al. Lower versus higher hemoglobin threshold for transfusion in septic shock. N Engl J Med. 2014;371:1381-1391.
6. Opal SM, Dellinger RP, Vincent J-L, Masur H, Angus DC. The next generation of sepsis clinical trial designs: what is next after the demise of recombinant human activated protein C? Crit Care Med. 2014;42:1714-1721.
7. Johansen M, Johansson P, Ostrowski S, et al. Profound endothelial damage predicts impending organ failure and death in sepsis. Semin Thromb Hemost. 2015;41:16-25.
8. Ostrowski SR, Gaini S, Pedersen C, Johansson PI. Sympathoadrenal activation and endothelial damage in patients with varying degrees of acute infectious disease: an observational study. J Crit Care. 2015;30:90-96.
9. Johansson PI, Haase N, Perner A, Ostrowski SR. Association between sympathoadrenal activation, fibrinolysis, and endothelial damage in septic patients: a prospective study. J Crit Care. 2014;29:327-333.
10. Johansson PI, Stensballe J, Ostrowski SR. Shock induced endotheliopathy (SHINE) in acute critical illness – a unifying pathophysiologic mechanism. Crit Care. 2017;21:25.
11. Moncada S, Gryglewski R, Bunting S, et al. An enzyme isolated from arteries transforms prostaglandin endoperoxides to an unstable substance that inhibits platelet aggregation. Nature. 1976;263:663-665.
12. Rubin LJ, Groves BM, Reeves JT, Frosolono M, Handel F, Cato AE. Prostacyclin-induced acute pulmonary vasodilation in primary pulmonary hypertension. Circulation. 1982;66:334-338.
13. Windelev NA, Ostrowski SR, Perner A, Johansson PI. Transfusion requirements and clinical outcome in intensive care patients receiving continuous renal replacement therapy: comparison of prostacyclin vs. heparin prefilter administration. Blood Coagul Fibrinolys. 2010;21:414-419.
14. Bruggeman LA, Pellicer JA, Horigan EA, Klotman PE. Thromboxane and prostacyclin differentially regulate murine extracellular matrix gene expression. Kidney Inter. 1993;43:1219-1225.
15. Sussmann M, Sarbia M, Meyer-Kirchrath J, Nüssing RM, Schrö R, Fischer JW. Induction of hyaluronic acid synthase 2 (HAS2) in human vascular smooth muscle cells by vasodilatory prostaglandins. Circ Res. 2004;94:592-600.
16. Kawabe J, Yuhki K, Okada M, et al. Prostaglandin I2 promotes recruitment of endothelial progenitor cells and limits vascular remodeling. Arterioscler Thromb Vasc Biol. 2010;30:464-470.
17. Birukova AA, Tian Y, Dubrovska O, et al. VE-cadherin trans-interactions modulate Rac activation and enhancement of lung endothelial barrier by iloprost. *J Cell Physiol*. 2012;227:3405-3416.

18. Chen HH, Chen TW, Lin H. Prostacyclin-induced peroxisome proliferator-activated receptor-alpha translocation attenuates NF-kappaB and TNF-alpha activation after renal ischemia-reperfusion injury. *Am J Physiol Renal Physiol*. 2009;297:F1109-F1118.

19. Cheng C-F, Lian W-S, Chen S-H, et al. Protective effects of adiponectin against renal ischemia-reperfusion injury via prostacyclin-PPARα-heme oxygenase-1 signaling pathway. *J Cell Physiol*. 2012;227:239-249.

20. Holmvang L, Ostrowski SR, Dridi NP, Johansson P. A single center, open, randomized study investigating the clinical safety and the endothelial modulating effects of a prostacyclin analog in combination with eptifibatide in patients having undergone primary percutaneous coronary intervention (PCI) for ST-segment elevation myocardial infarction. *Prostaglandins Other Lipid Mediat*. 2012;99:87-95.

21. Johansson PI, Mortensen CR, Nielsen T, et al. The effect of intraoperative and 6-h postoperative intravenous administration of low-dose prostacyclin on the endothelium, hemostasis, and hemodynamics in patients undergoing a pancreaticoduodenectomy: a randomized-controlled pilot study. *Eur J Gastroenterol Hepatol*. 2017;29:400-406.

22. Berthelsen RE, Ostrowski SR, Bestle MH, Johansson PI. Co-administration of iloprost and eptifibatide in septic shock (CO-ILEPSS) a randomised, controlled, double-blind investigator-initiated trial investigating safety and efficacy. *Crit Care*. 2019;23:301.

23. Schulz KF, Altman DG, Moher D. CONSORT 2010 statement: updated guidelines for reporting parallel group randomised trials. *J Pharmacol Pharmacother*. 2010;1:100-107.

24. Gordon AC, Perkins GD, Singer M, et al. Levosimendan for the prevention of acute organ dysfunction in sepsis. *N Engl J Med*. 2016;375:1638-1648.

25. Taichman DB, Sahni P, Pinborg A, et al. Data sharing statements for clinical trials: a requirement of the International Committee of Medical Journal Editors. *Lancet*. 2017;389:e12-e14.

26. Grant SM, Iloprost GKL. A review of its pharmacodynamic and pharmacokinetic properties, and therapeutic potential in peripheral vascular disease, myocardial ischaemia and extracorporeal circulation procedures. *Drugs*. 1992;43:889-924.

27. Singer M, Deutschman CS, Seymour CW, et al. The third international consensus definitions for sepsis and septic shock (sepsis-3). *JAMA*. 2016;315:801-810.

28. Dellinger RP, Levy MM, Rhodes A, et al. Surviving sepsis campaign: international guidelines for management of severe sepsis and septic shock: 2012. *Crit Care Med*. 2013;41:580-637.

**SUPPORTING INFORMATION**

Additional supporting information may be found online in the Supporting Information section.

How to cite this article: Bestle MH, Clausen NE, Søe-Jensen P, et al. Efficacy and safety of iloprost in patients with septic shock-induced endotheliopathy—Protocol for the multicenter randomized, placebo-controlled, blinded, investigator-initiated trial. *Acta Anaesthesiol Scand*. 2020;64:705–711. [https://doi.org/10.1111/aas.13546](https://doi.org/10.1111/aas.13546)