Pulse Glucocorticoid Therapy in Patients With ST-Segment Elevation Myocardial Infarction
PULSE-MI

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PULSE GLUCOCORTICOID THERAPY IN PATIENTS WITH ST-SEGMENT ELEVATION MYOCARDIAL INFARCTION

A prospective randomized, double-blinded, placebo-controlled clinical trial to investigate whether single-dose pulse therapy with glucocorticoid treatment prior to primary PCI can reduce final infarct size in patients with STEMI.

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1. Abbreviations

AEs	Adverse Events
AMI	Acute Myocardial Infarction
CAG	Coronary Angiography
CABG	Coronary Artery Bypass Graft
CEC	Clinical Event Committee
CFR	Coronary Flow Reserve
CMR	Cardiac Magnetic Resonance
CRP	C-Reactive protein
ECG	Electrocardiogram
GCP	Good Clinical Practice
HF	Heart Failure
IRA	Infarct-Related Artery
ICF	Informed Consent Form
IMR	Index of Myocardial Resistance
LAD	Left Anterior Descending Artery
LGE	Late Gadolinium Enhancement
LVEF	Left Ventricular Ejection Fraction
MVO	Microvascular Obstruction
MSI	Myocardial Salvage Index
PCI	Percutaneous Coronary Intervention
PI	Principal Investigator
RD	Referring Cardiology Fellow Doctor
SAEs	Serious Adverse Events
SCAD	Spontaneous Coronary Artery Dissection
SD	Standard Deviation
STEMI	ST-segment elevation myocardial infarction
SUSARs	Suspected Unexpected Serious Adverse Reactions
TIA	Transient Ischemic Attack
TIMI	Thrombolysis In Myocardial Infarction

2. Background

2.1 Purpose of the Study

The overall primary objective of the Pulse Glucocorticoid Therapy in Patients with ST-Segment Elevation Myocardial Infarction (STEMI) trial (PULSE-MI) is to test the hypothesis that administration of single-dose glucocorticoid pulse therapy in the *pre-hospital* setting reduces final infarct size in patients with STEMI treated with primary percutaneous coronary intervention (PCI).

2.2 Clinical Relevance

STEMI remains one of the leading causes of mortality globally despite significant advances in reperfusion therapies with timely primary PCI[1]. One-year mortality following STEMI has decreased to around 10%, yet a substantial number of patients with STEMI develop heart failure (HF) (22%) which impairs prognosis significantly[2-6]. A main driver for mortality and heart failure following STEMI is infarct size that is the cause of ischemia and reperfusion injury[7]. Of note, inflammation is induced immediately after the onset of acute myocardial ischemia and is subsequently exacerbated following reperfusion[8]. Hence, inflammation in the setting of STEMI is an important factor in both acute myocardial ischemia and reperfusion injury, which is why inflammation *per se* is a feasible and desirable target for improving prognosis in these patients.

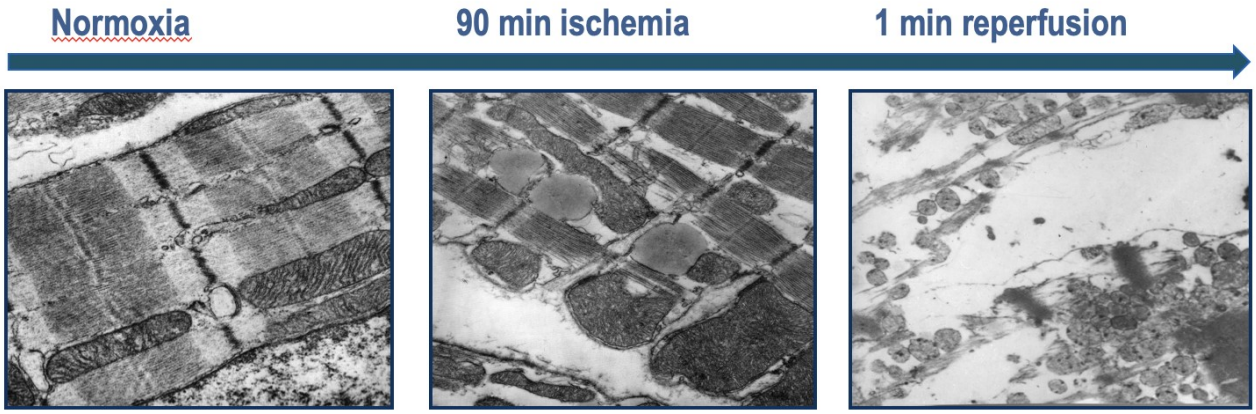
2.3 Reperfusion Injury and Inflammation in STEMI

Up to 60% of patients with STEMI have reperfusion in the pre-hospital setting due to effective antithrombotic treatment with heparin and aspirin[9]. Yet, primary PCI including stent implantation is the preferred revascularization strategy in patients with STEMI to reduce the occurrence of re-infarction and improve the overall prognosis in these patients[10, 11]. However, an additional myocardial injury may occur when coronary blood flow is restored, a phenomenon called "*reperfusion injury*"[7]. The injury occurs immediately after the restoration of blood flow requiring early treatment and may account for up to 50% of the damaged myocardium (Figure 1)[4, 6]. Thus, the extent of myocardial damage and resulting final infarct size are major components in determining the clinical course following STEMI[7, 12]. However, the phenomenon is not completely understood, and no preventive treatment exists. Multiple pathophysiological factors may contribute to reperfusion injury such as microvascular obstruction (MVO), oxidative stress, and inflammation.

Following reperfusion, platelets are the first cells to migrate to the damage myocardium inducing accumulation of leukocytes in the infarct-related-artery (IRA) prompting local inflammation

and release of pro-inflammatory cytokines inducing systemic inflammation[13, 14]. Hence, inflammation *per se* may drive excessive cardiomyocyte death resulting in decreased contractility and increased infarct size post-acute myocardial infarction (AMI)[15]. Moreover, in the course following AMI and subsequently reperfusion, the myocardium starts healing and scarring resulting in remodeling of the ventricle potentially causing either compensatory hypertrophy or thinning of the myocardium, which may lead to reduced left ventricular ejection fraction (LVEF) and HF[16]. Of note, inflammation plays a critical role in ventricular remodeling post-AMI, thus inflammation in relation to reperfusion injury may extend of myocardial damage following STEMI[17].

Figure 1. Microscopy of a rabbit cardiac myocyte.



Engstrøm et al., Unpublished data. *During 90 minutes of ischemia, the structure integrity of the cardiomyocyte is preserved. Following 1-minute of reperfusion, the integrity and structure of the cardiomyocyte is lost.*

2.4 Glucocorticoids and STEMI

Glucocorticoids are crucial in regulation of the systemic inflammatory response and may therefore be beneficial in limiting myocardial injury following STEMI[18, 19]. Glucocorticoids mediate two different mechanisms: the genomic effect mediated by glucocorticoid receptor occupation, gene transcription, and translation within the cells which is induced within hours and the non-genomic effect, which is induced rapidly (<15 minutes) after administration via plasma membrane bound receptors and independent of cytosolic receptor occupation and genomic regulation[20, 21]. Some of the proposed nongenomic effects of glucocorticoids on the cardiovascular system include decreased vascular inflammation and reduced infarct size by activating the endothelial nitric oxide synthase (eNOS), cardioprotection through membrane stabilization through nitric oxide (NO) production, and increasing the contractility of the vascular smooth muscle cells[20]. Taken altogether, the non-

genomic actions secure that the anti-inflammatory effect is initiated quickly to protect the myocardium immediately whereas, in addition, the genomic actions protects the tissue in the subsequent period.The use of glucocorticoid in relation to ischemia has therefore been investigated in experimental, in-vivo, and clinical studies in the 1970's and 1980's. In experimental animal studies, glucocorticoids showed beneficial effects on hemodynamics and infarct size following ischemia, however most studies were performed in non-reperfused animals[16, 19, 22]. Albeit this, clinical studies in patients with AMI have shown conflicting results[16, 19, 22]. Further, definitive conclusions on glucocorticoids' efficacy in patients with STEMI are difficult to draw due to different study designs, investigational agents, and medicine dosage[16]. However, most importantly, all studies performed in patients with AMI were before the era of primary PCI[16], and since the implementation of PCI in treatment of STEMI, the prognosis in these patients has improved significantly[1]. Considering these advances in reperfusion strategies, glucocorticoids may now add an additional beneficial role in limiting infarct size and improving prognosis in patients with STEMI[19]. Although questions have been raised of impaired infarct heeling in relation to glucocorticoid treatment in the setting of STEMI, the concern is not supported by randomized controlled clinical trials[16, 23]. Long-term use of glucocorticoids is, however, associated with adverse side-effects, but short-term use of glucocorticoids has only few side-effects[24]. None of the previous studies have investigated single-dose pulse therapy with glucocorticoids in patients with STEMI who were treated with primary PCI. Based on reperfusion advances, role of inflammation in both STEMI and reperfusion injury, and few side effects of short-term use of glucocorticoid, intravenous short-term treatment with glucocorticoid could add potential benefit and have an important therapeutic role in limiting the degree of myocardial injury and thereby improving prognosis in patients with STEMI.

2.5 Safety and Rationale

In light of the above, we will investigate the well-known, widely used, and highly potent glucocorticoid, methylprednisolone, administrated as single pulse-dose therapy prior to primary PCI in patients with STEMI in a randomized clinical trial.

Pulse glucocorticoid therapy was first used for acute rejection following kidney transplantation[25], and is defined as treatment with ≥250 mg prednisolone or equivalent for one or more days[26]. Pulse glucocorticoid therapy is now used as effective and safe treatment of a wide range of diseases such as rheumatic diseases, dermatologic diseases, optic neuritis, multiple sclerosis, acute disseminated encephalomyelitis, glomerulonephritis, and systemic vasculitis[27]. Doses up to 2500

mg methylprednisolone, depending on disease severity, are recommended and used in treating these inflammatory conditions[27]. The beneficial acute effects of pulse glucocorticoid therapy in these conditions are thought to be mediated by the nongenomic effects of glucocorticoids via plasma membrane bound receptors, and the estimated complete glucocorticoid receptor occupation is reached at approximately 100 mg methylprednisolone, reaching maximum activation around 250 mg[27, 28]. Genomic mechanisms cannot explain the beneficial effects of glucocorticoid doses >100 mg, and the extent of receptor activation is directly dependent on methylprednisolone dosage[29]. Despite concern of serious adverse events (SAEs) following high-dose methylprednisolone, intravenous pulse therapy is not associated with increased risk of mortality, adverse events (AEs), or AEs leading to discontinuation of pulse methylprednisolone[30, 31]. The Norwegian "Summary of Product Characteristics" (SPC) for Solu-Medrol (methylprednisolone) recommends doses up to 30 mg/kg. The Danish SPC for Solu-Medrol (methylprednisolone) recommends infusion of 250 mg methylprednisolone over a period of ≥5 minutes[32]. This is due to the potential side-effects of a rapid infusion of methylprednisolone. These potential side-effects are, however, not well supported by the literature. In fact, pulse glucocorticoid therapy administrated intravenously over a short period of time has few side effects compared with long-term oral treatment[24, 30, 33]. Considering this knowledge, safety, and potential beneficial dual effects in patients with STEMI undergoing primary PCI, administration of 250 mg methylprednisolone administrated in the *pre*-hospital setting may well offer substantial advantages in these patients. The expected effect will then translate into reduced infarct size, better clinical outcomes including fewer patients with HF and subsequently reduced mortality following STEMI.

3. Hypothesis

In patients with STEMI undergoing primary PCI, 250 mg methylprednisolone administrated in the *pre-hospital* setting limits reperfusion injury and reduce final infarct size measured by late gadolinium enhancement (LGE) on cardiac magnetic resonance (CMR) 3 months after STEMI.

4. Objectives of the Study

4.1 Overall Primary Objective

To determine whether 250 mg methylprednisolone administrated in the *pre-hospital* setting reduces final infarct size on CMR in patients with STEMI undergoing primary PCI.

4.2 Secondary Objectives

- To determine whether the response of methylprednisolone differs in pre-specified sub-groups: age, gender, pre-PCI Thrombolysis in Myocardial Infarction (TIMI) flow, culprit in left anterior descending artery (LAD), time from first medical contact to primary PCI, and duration of symptoms prior to primary PCI.
- To determine whether methylprednisolone improves LVEF within 48 hours and 3 months after primary PCI.
- To determine whether methylprednisolone decreases acute infarct size and edema measured on CMR within 48 hours of primary PCI.
- To determine whether methylprednisolone decreases degree of microvascular obstruction (MVO) and hemorrhage measured on CMR within 48 hours of primary PCI.
- To determine whether methylprednisolone inhibits the inflammatory response at admission,24 hours after admission and after 3 months. Markers of inflammation: High sensitivity C-reactive protein (hsCRP), leucocyte- and differential count, plasma cytokine levels (IL-1b, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-10, IL-12, IL-13, IL-17A, G-CSF, GM-CSF, MCP-1, MIP-1beta, INF-g and tumor-necrosis factor α (TNF-α)), and procalcitonin
- To determine whether methylprednisolone increases myocardial salvage index (MSI) on CMR
- To determine whether methylprednisolone increases coronary flow reserve (CFR) and decreases index of microcirculatory resistance (IMR).
- To determine whether methylprednisolone improves clinical outcomes (all-cause mortality and hospitalization for HF) at one year after primary PCI.
- To determine whether methylprednisolone decreases peak Troponin-T and creatine kinase myocardial band (CKMB) during admission.

All these outcomes are relevant to patients with STEMI and have the potential to change the clinical course following STEMI and standard clinical practice.

5. Endpoints and Follow-Up

Primary and secondary endpoints are listed in Table 1.

Table 1. PULSE-MI endpoints

	Intervention with glucocorticoid vs. no intervention	*Timeframe*
Primary endpoint	Final infarct size (% of ventricular mass) measured by LGE on CMR	3 months after primary PCI
Secondary endpoints	The extent of MVO on CMR	Within 48 hours after primary PCI
	The extent of hemorrhage on CMR	Within 48 hours after primary PCI
	Other CMR efficacy parameters as specified in the appendix A	Within 48 hours and 3 months after primary PCI
	All-cause mortality and hospitalization for HF	3 months after primary PCI
	Peak Troponin-T and CKMB	During admission
	Safety: Incidence of adverse events the first seven days of index event	Within 7 days of STEMI

Exploratory endpoints:

1. Pre-PCI and post-PCI TIMI flow
2. C-reactive protein (CRP), leucocyte- and differential count, thrombocytes, HbA1c, p-Glucose, pro-brain natriuretic peptide (BNP), creatine kinase, creatinine, potassium, and sodium at admission, during admission and after 3 months
3. Arrhythmia (ventricular tachycardia/ventricular fibrillation) during transportation in the ambulance and peri-procedural during PCI
4. Bolus CFR, IMR, absolute CFR, and microvascular resistance reserve (MRR) following primary PCI
5. LVEF on transthoracic echocardiography during admission and one to three months following primary PCI
6. Killip Class at admission

7. Biomarkers of inflammation at admission, 24 hours after admission, and after 3 months
8. All-cause mortality and hospitalization for HF after 1 and 10 years

5.1 Endpoint Definition

All clinical endpoints (all-cause mortality and hospitalization for HF) are defined in Appendix B. The CMR efficacy parameters are described in Appendix A. Secondary endpoints will be identified using hospital files in which all deaths and hospital referrals are reported.

Events will subsequently be adjudicated by members of our clinical events committee (CEC), who will review all relevant medical records.

5.2 Event Recording

Violations of the protocol will be monitored continuously. The CEC is responsible for adjudicating the clinical secondary endpoints. No members of the CEC participate in recruitment or data collection or have access to any information regarding treatment allocation. A list of all CEC members is given in Appendix C.

5.3 Monitoring

Monitoring of the study will be done by a local Good Clinical Practice (GCP) unit. The trial will be conducted in compliance with the published trial protocol, the Helsinki Declaration, the GCP guidelines (ICH-GCP), and national laws. The Sponsor of the trial allows investigators and institutions involved in the trial to perform trial-related monitoring, audit, and inspection from the authorities including direct access to the data and documents related to the trial.

The first monitoring meeting will be held after inclusion of 2 patients. The following meetings will be held as needed. A full monitoring plan will be elaborated and composed before initiation of the study by the local GCP unit.

6. Study Setup

6.1 Study Design

This study is an investigator-initiated, 1:1 randomized, double-blinded, placebo-controlled prospective clinical proof-of-concept trial to investigate whether pulse therapy with 250 mg methylprednisolone in the *pre-hospital* setting limit reperfusion injury and reduce infarct size, and subsequently improve clinical outcomes in patients with STEMI undergoing primary PCI. The trial will be

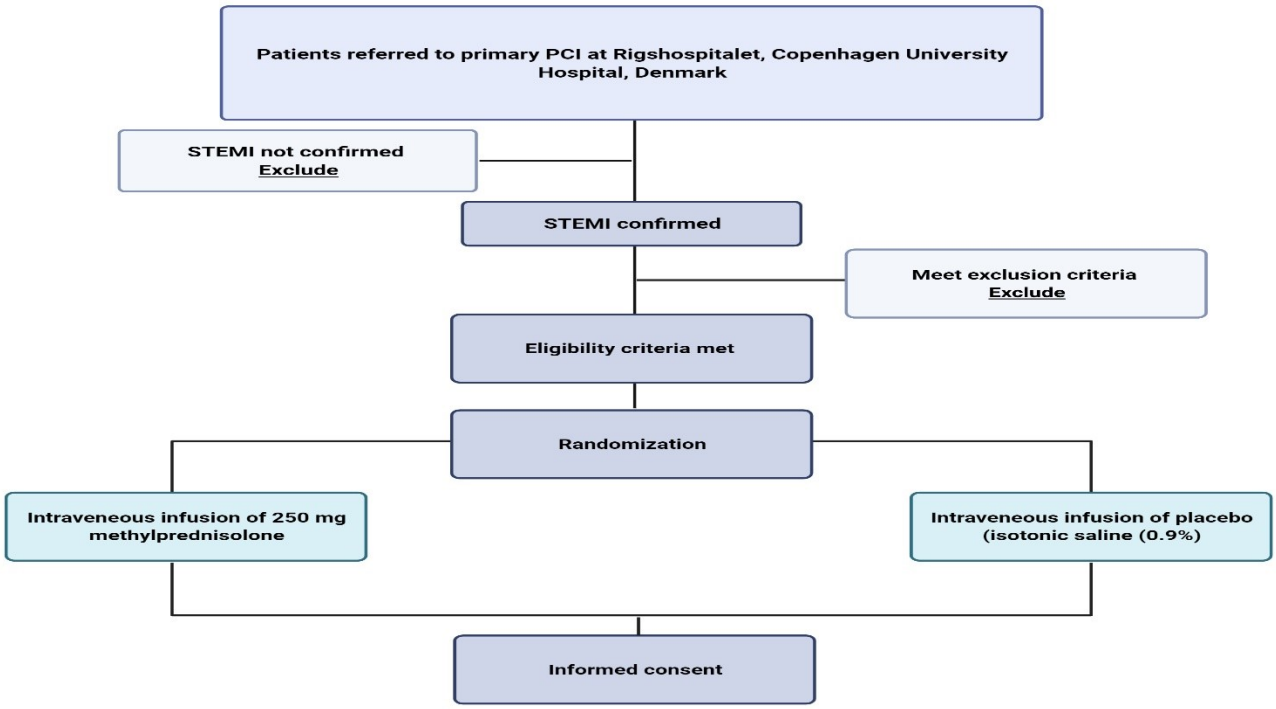
chaired at the Department of Cardiology, Rigshospitalet, Copenhagen University Hospital, Denmark.

6.2 Target Patient Population and Number of Patients

The target patient population is male and female patients aged 18 years and over with documented evidence of STEMI, eligible planned for primary PCI who had symptom onset within 12 hours before randomization who were initially managed by an ambulance paramedic and included by the referring and including doctor (on-call cardiology fellow doctor (RD) at Rigshospitalet) in the *pre-hospital* setting.

A total of 378 patients with STEMI (189 patients treated with intravenous infusion of 250 mg methylprednisolone and 189 patients with intravenous placebo infusion) are included for the primary endpoint of final infarct size (% LGE of the LV) on CMR at 3 months. The inclusion will continue until this number has been reached and with an expected drop-out rate of 40%, 530 patients need to be included and randomized. All patients will be included in the *pre-hospital* setting from throughout the Capital Region and Region Zealand of Denmark (Figure 2).

Figure 2. Flowchart of the study



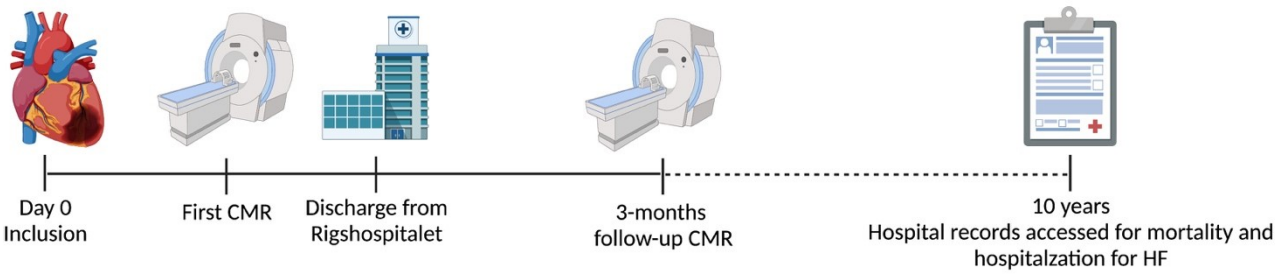
6.3 Study Setting

In all, patients with STEMI will be included in the *pre-hospital* setting throughout the Capital Region and Region Zealand of Denmark prior to primary PCI at Rigshospitalet, Copenhagen University Hospital, Denmark if eligible to participate in the study. Eligibility will be carefully screened by the RD at Rigshospitalet. Data collection and analysis will be undertaken through the Department of Cardiology, Rigshospitalet, Copenhagen University Hospital, Copenhagen, Denmark.

6.4 Estimated Study Duration

We expect the study to begin once the study has been approved by the government. Rigshospitalet performs approximately 1000 primary PCI procedures per year[34]. An inclusion is planned to be completed in 36 months, and the 3-months follow-up period will be completed after 39 months. However, the study will be stopped before when 378 patients have completed CMR at 3 months. Following completion of the trial, the hospital records will be accessed at 1 and 10 years to obtain information on mortality and hospitalization for HF (Figure 3).

Figure 3. Timeline of the study



6.5 Inclusion and Exclusion Criteria

Inclusion criteria for recruitment are listed below and must be fulfilled for the patient to be randomized:

1. Age ≥18 years including fertile women*.
2. Acute onset of chest pain with < 12 hours duration.
3. STEMI as characterized on electrocardiogram (ECG) by one of the following:

   1) at least two contiguous leads with ST-segment elevation ≥2.5 mm in men < 40 years, ≥2 mm in men ≥40 years, or ≥1.5 mm in women in leads V_2-V_3 and/or ≥1 mm in the other leads,

   2) presumed new left bundle branch block with ≥1 mm concordant ST-segment elevation in leads with a positive QRS complex, or concordant ST-segment depression ≥1 mm in V_1-V_3, or discordant ST-segment elevation ≥5 mm in leads with a negative QRS complex,

   3) Isolated ST depression ≥0.5 mm in leads V_1-V_3 and ST-segment elevation (≥0.5 mm) in posterior chest wall leads V_7-V_9 indicating posterior acute myocardial infarction (AMI),

   4) ST-segment depression ≥1 mm in eight or more surface leads, coupled with ST-segment elevation in aVR and/or V_1 suggesting left main-, or left main equivalent-coronary obstruction.

Exclusion criteria for recruitment are listed below and none of the criteria must be fulfilled for a patient to be eligible:

1. Presentation with cardiac arrest (out of hospital cardiac arrest (OHCA)).
2. Time from symptoms onset to primary PCI > 12 hours.
3. Known allergy to glucocorticoid or known mental illness with maniac or psychotic episodes.

* It is not possible to perform a pregnancy test (HCG urine test) in the pre-hospital setting. However, methylprednisolone is not contraindicated in pregnant women.

4. Patients with previous AMI in the assumed culprit artery.
5. Previous coronary artery bypass graft (CABG).
6. Unable to read and understand Danish.

7. Ethical Assessment

The patient will be informed and treated according to the international guidelines (ICH-GCP) and protected under the General Data Protection Regulation (GDPR) including the processing of personal data (data protection law) and the Danish Health Act. As to the need for fast transportation and treatment (primary PCI), the main ethical issue that could arise from this trial is that informed consent cannot be obtained prior to intervention and treatment with primary PCI due to the acute setting of the study. Intervention before informed consent is done to secure initiation of the study treatment as soon as possible due to the need of acute intervention and treatment in STEMI and vulnerable patients. Moreover, the ability of the patient to provide informed consent in the *pre-hospital* setting is questionable due to vulnerability, chest pain and acute setting. This implies the following:

- Recruitment and randomization in the *pre-hospital* setting will be performed by the paramedic in the ambulance.
- The including doctor is the RD at Rigshospitalet. The including RD is continuously in touch with the paramedic in the ambulance by telephone from the time of screening until randomization has been done.
- The patient will provide informed consent as soon as possible following the acute management of the patient (primary PCI), once the patient is stable.
- The process of informed consent is described thorough in section 9.2. The process follows article 35 of Clinical Trial Regulation EU No 536/2014 and Law of Clinical Trials, §3.
- The justification for providing informed consent following intervention due to the emergency situation is described thorough in the section 9.2.1.

8. Study Progress and Conduct

8.1 Screening and Inclusion

Screening, inclusion and registration will be done in the ambulance in the *pre-hospital* setting. The including RD at Rigshospitalet will carefully check the eligibility of the patient to participate in the study. The including RD is continuously in touch with the paramedic in the ambulance by telephone

from the time of screening until randomization has been done. The including RD has all the necessary information about the patient including previous medical and psychotic history, actual medications, and clinical status. Clinical status includes patients' general condition, blood pressure, heart rate, and ECG changes. As of the acute nature of STEMI and study design, the qualifying ECG will be performed in the *pre-hospital* settings as normal procedure. These data will be used for eligibility evaluation by the including doctor (RD) at Rigshospitalet. The RD that is responsible for inclusion of the patient via telephone must ensure that there are no contraindications for administration of glucocorticoids (including history of known mental illness with maniac or psychotic episodes) as part of the eligibility check. This information is available via the patient charts that can be accessed by the RD. If the including RD find the patient eligible to participate in the study, the including RD will ask the paramedic to include the patient in the study in the *pre-hospital* setting.
In the *pre-hospital* setting, the paramedic in the ambulance will do the following:

1. Include the patient in the study following eligibility check by the RD at Rigshospitalet.
2. Randomize the patient by picking a random box from a storage in the ambulance containing the study medicine (methylprednisolone or placebo). All boxes are identical, and the paramedic is therefore blinded when randomizing.
3. Register the patient in an electronic database, REDCap.

All patients, or the legally designated representative must personally sign and date the *informed consent form* (ICF) as soon as possible following the primary PCI procedure (described in detail in section 9.2). The independent legally designated representative of patients who are enrolled and unable to provide informed consent will be Dr. Reza Jabbari. If patients remain unable to provide informed consent 3 days following admission, the nearest relative to the patient will also sign the ICF on behalf of the patient as soon as possible. Informed consent from the patient will subsequently be obtained as soon as possible. The procedure of informed consent is described thorough in section 9.2.

8.2 Registration

All paramedics have access to the registration form in REDCap. The registration form can be accessed through a QR-code on the study medicine box or through an internet browser. When a patient is registered, the patient will be allocated a patient identification number by a computer-generated code. The identification number will be used to identify the patient throughout the whole study. The identification number consists of three digits assigned in sequential order as patients are

screened (001, 002, 003, etc.). All patients who are screened for eligibility will be registered in an electronic database, REDCap. In the registration form, the paramedic will annotate the following information about the patient: 1) civil registration number, 2) the number of the study medicine box, 3) time of intervention, and 4) if the study medicine was given according to the protocol (yes/no), and if not, why the study medicine was not given. The paramedic in the ambulance will sign the registration form following inclusion.

8.3 Randomization

Randomization of the patient will take place in the *pre-hospital* setting. Patients who satisfy all the eligibility requirements will be randomized. Randomization is done by the paramedic in the ambulance. Patients will be randomized in a 1:1 ratio to either infusion of 250 mg methylprednisolone or placebo that are placed in identical boxes. The paramedic randomizes the patient by picking a random box stored in the ambulance delivered by the pharmacy, including methylprednisolone or placebo, and administrates (over a period of 5 minutes) the content by a bolus infusion. Following infusion of the randomized treatment, the ambulance will bring the box to the hospital alongside with ECGs obtained in the ambulance in case of emergency unblinding. All boxes are identical and has a unique box serial number. Once a patient has been randomized, the patient is considered enrolled and included in the study. The box serial number is not necessarily the same as the patient identification number. In case of any technical issues, the responsible Ph.D. student or other investigators from the steering committee will be available.

All including RD's at Rigshospitalet, legally designated representative, and paramedics have relevant knowledge of the study from the handing out of the study protocol, protocol summaries, and pocket cards as well as via e-mail and on-going project recaps. None of the including RDs, legally designated representative, or paramedics are involved with the trial except for screening, enrollment, and inclusion of the patients.

8.4 Study Medicine and Placebo

Patients will be allocated to either infusion of 250 mg methylprednisolone or placebo that are placed in identical boxes. Each box will either contain Solu-Medrol or placebo and will be numbered randomly by the pharmacy. Administration of the study medicine will not prolong the trans-

portation or any aspect of the standard treatment of the patient. The patients will be treated according to standard procedures in the ambulance. Primary PCI and antithrombotic regimens will be performed according to standard procedures at Rigshospitalet, Denmark.

All study medicine and placebo boxes are identical and numbered in a random manner at the pharmacy. The numbers on the identical boxes are unique, and the pharmacy has the allocation list. The study medicine is 2 x 125 mg/2 mL Solu-Medrol, a total of 250 mg/4 mL, which comes as a sterile powder with preservative free isotonic NaCl as diluent. The medicine takes 30 to 60 seconds to mix and needs to be used within 48 hours when opened. The placebo will be 0.9% NaCl in 4 mL ampoules. The infusion of both the study medicine and placebo will be done over a period of 5 minutes during transportation.

8.5 Blinding

The pharmacy of the Capital Region of Denmark will pack inbox and label the study medicine and placebo ampoules. The randomization will be allocated by the pharmacy and their independent statistician using a random number generator. The pharmacy will keep the allocation list and will not be involved otherwise in the trial. Sponsor and Investigators of the trial keep an allocation list locked at in a safety cabinet in case of emergency unblinding. Sponsor and Investigators are responsible for any emergency unblinding. Moreover, Investigators can unblind the study medication without prior contact to the sponsor. According to the ICH-GCP guideline 4.3.1, the Investigators are responsible for all trial-related medical decisions and are able to unblind immediately without restrictions. The pharmacy is not able to make matching placebo ampoules as Solu-Medrol does not come in a premixed ampoule and needs to be mixed before use in the ambulance. The treating paramedic is therefore unblinded after inclusion, however, the paramedic will not take any further part in the trial nor the treatment of the patient once arrived at Rigshospitalet, Denmark.

Following preparation, the boxes will be shipped to all the critical care unit stations in the Capital Region and Region Zealand of Denmark. All units are manned 24 hours 7 days a week. The investigators will make sure that all ambulances at the different stations always have boxes in storage and ready for use. Before each shift, the paramedic will make sure that a minimum of five boxes are stored in each ambulance.

8.6 Dosage Adjustments

The dose of the study medicine will be fixed. In case of any clinical signs of an allergic reaction or severe side effects are suspected, the infusion will be terminated immediately. No dosage adjustments are allowed.

8.7 Discontinuation of Study Participation

A patient can withdraw from the study at any time, or when medically necessary, as judged by the Investigator. The information about withdraw at any time from the trial is emphasized to the patient. Emergency unblinding may occur judged by the Investigators. Sponsor and Investigators are responsible for the unblinding procedure. Investigators can unblind the study medication without prior contact to the sponsor. According to the ICH-GCP guideline 4.3.1, the Investigators are responsible for all trial-related medical decisions and are able to unblind immediately without restrictions. Other reasons for ST-segment elevation (e.g. myocarditis, pericarditis, takotsubo) and patients who does not fulfill inclusion- and exclusion criteria (post-randomization exclusions) will be excluded from the study but followed for 3 months for safety monitoring.

8.8 Research Biobank

During the study, around 50 mL of blood in total will be drawn for a research biobank. Blood samples will be drawn at admission in the catherization laboratory, after 24 hours in the ward, and at follow-up after 3 months. The research biobank will be constructed in accordance with national legislation. Biological material in the research biobank will be used for analyses of biomarkers for inflammation as well as other markers of interest as described in the section “4.2 Secondary objectives and 5. Endpoints”. The analysis of biomaterial in the research biobank will occur following completion of the trial, i.e. during 2026, and all samples and data will be stored pseudo-anonymously. Following analyses, the research biobank will be terminated in 2026, before the 31th of December. If any biological material remains following analyses in 2026, the material will be frozen in a biobank for future research and destroyed after ten years. Any further future analyses of unused research biobank material associated to this project will require ethics committee approval.

8.9 Concomitant Procedures

8.9.1 Primary PCI Procedure

The patient is admitted to the hospital because of STEMI with symptom onset less than 12 hours before contact, defined as a primary PCI according to guidelines[10]. The primary PCI will be performed as usual clinical practice. The patient is pretreated with aspirin (300 mg) and 10.000 units of heparin in the *pre-hospital* setting by the paramedics as soon as the including RD at Rigshospitalet has set the diagnosis of STEMI and referred the patient to primary PCI at Rigshospitalet, Denmark. These procedures are of standard clinical practice and in accordance with guidelines. Primary PCI is prepared and performed as usual in the catheterization laboratory. Immediately subsequent to primary PCI, a loading dose of either 60 mg prasugrel or 180 mg ticagrelor is given per os. Clopidogrel 600 mg can be chosen alternatively at the discretion of the treating physician (e.g. high bleeding risk, ongoing anticoagulation therapy).

8.9.2 Follow-Up Medical Treatment

The follow-up medical treatment will be completely in accordance with international guidelines and local standard operating procedures. Dual antiplatelet therapy with aspirin 75 mg once daily and prasugrel 10 mg or 5 mg b.i.d., or ticagrelor 90 mg will be given for 12 months. Deviation from this is allowed at the discretion of the PCI operator.

8.9.3 Follow-Up Investigations

ECG's and cardiac enzymes will be obtained as of standard practice and routine. Echocardiography will be performed both bedside before the primary PCI, to ensure no mechanical complications are present at admission, and within the first days of index event in a stable phase. Additionally, a standard echocardiography will be performed at 3 months following index event as part of standard care.

CMR, as part of the study, will be performed on all study participants without contraindications towards CMR (decreased renal function (estimated glomerular filtration rate (eGFR <30 ml/min), severe claustrophobia, ferrous metallic foreign body, pacemaker/implementable cardioverter-defibrillator (ICD), or allergy to contrast (Gadolinium)) within the first 48 hours of index event and 3 months after the index event at Rigshospitalet, Copenhagen University Hospital, Denmark. The CMR will be performed during admission at Rigshospitalet within 48 hours to measure

area at risk, MVO, edema, hemorrhage, and acute infarct size. The CMR at 3 months will be performed to measure final infarct size which is the primary endpoint of the trial. Patients will be offered transportation renumeration to undertake the CMR scan at 3 months. CMR is not associated with any complications. All scans are analyzed for in Circle Cardiovascular Imaging (CVI42). The CMR protocol is described in detail in Appendix A.

8.9.4 10 Year Follow-Up

The study is completed following total inclusion of 378 patients for the primary endpoint. Records of the patients are assessed after 10 years to access mortality and hospitalization for HF. No safety event recording and annual safety reports will be done in the time between completion of the trial and the 10-year follow-up assessment.

9. Ethical Considerations

9.1 Ethical Conduct of the Study

The ethics committee will approve the study protocol and ICF before the initiation of the trial. The study protocol will be registered at the Danish Data Protection Agency. Good Clinical Practice will be observed throughout the whole trial and appropriate international, European, and national legislations and guidelines will be respected. The Data Safety Monitoring Board (DSMB) will monitor the study by assessing the safety and efficacy of interventions during the trial and the overall conduct of the clinical trial. The responsibilities of the DSMB is described thorough in the DSMB charter.. The trial will be conducted in compliance with the published trial protocol, the Helsinki Declaration, the GCP guidelines (ICH-GCP), and national laws.

Due to the acute nature of STEMI, vulnerability, and inability of the patient to provide informed consent in the *pre-hospital* setting, intervention will occur in the *pre-hospital* setting prior to obtaining informed consent. Patients referred to primary PCI at Rigshospitalet are in an acute, vulnerable, and painful situation during transportation to the hospital, therefore, we find it acceptable to include and initiate the intervention in the *pre-hospital* setting before provision of informed consent. The study medicine (methylprednisolone) is a known and well-described drug used in several acute settings and is therefore considered safe without any harm/risks to the patient. Additionally, time from medical contact to intervention and treatment is pivotal to secure the best possible prognosis of patients with STEMI. Thus, intervention in the *pre-hospital* setting without informed consent ensure no delay of the urgent treatment of the patient's medical condition.

9.2 Informed Consent

Up to 92.5% of patients with STEMI will be stable, without chest pain, and able to provide informed consent as soon as possible following the acute treatment with primary PCI. Following the acute phase, a study nurse or one of the investigators will provide detailed and thorough information about the study and obtain informed consent from the patient (signature of the ICF). Signature of ICF is designed and encrypted in REDCap and will be signed in REDCap following informed consent. The patient will provide informed consent as soon as possible following the acute initial treatment (primary PCI), once the patient is stable. The right to withdraw informed consent at any time will be emphasized to the patient. If the patient does not wish to participate in the study, the data collection of the patient will be stopped immediately.

In the remaining 7.5% patients with STEMI unable to provide informed consent following primary PCI, consent will be obtained by the legally designated representative who will sign the ICF. The legally designated representative consent will be followed by informed consent and signature of the ICF by the patient, once the patient is stable and able to provide consent. If the patient remains unable to provide informed consent 3 days following admission, the closest relative to the patient and legally designated representative will provide consent on behalf of the patient and sign the ICF as soon as possible. The patient will subsequently provide informed consent as soon as possible. In cases of death prior to informed consent, the legally designated representative will provide informed consent as soon as possible. The closest relative will be informed about participation in the study including the use of the patient's data in relation to the study. Further, the closest relative will be informed of the patient's right to discontinue from the study at any time. In case of any objection or unwillingness towards participation in the study, the patient will then be excluded from the study. However, medical records of the patient following death informed consent will be checked for causes of death to ensure no relation to the study medicine.

The informing doctor/study nurse will ensure:

1. Each patient is given full and adequate oral and written information about the nature, purpose, possible risk, and benefit of the study,
2. The patient is aware of the right to discontinue from the study at any time,
3. The patient is given the opportunity to ask questions about the study.

9.2.1 In the Emergency Situation

The inherent acute nature of the study necessitates circumvention of the usual requirements of a 24-hour time for consideration, as it has previously been done in comparable acute studies e.g. ATLANTIC[35], EUROMAX[36], DANAMI-3[37], CONDI-2/EPIC-PPCI[38], and DANEGAPTIDE[39]. This approach is reasonable because the intervention is without any known risk and because the intervention has potential benefit for the individual patient.

Up to 60% of patients with STEMI have reperfusion in the *pre-hospital* setting due to effective antithrombotic treatment with heparin and aspirin. To investigate the effect on glucocorticoid on reperfusion induced inflammation and injury, randomization and intervention is done in the *pre-hospital* setting. Hence, intervention in the ambulance before reperfusion is needed to initiate the potentially beneficial and immediate nongenomic effects and subsequent protective genomic actions of pulse glucocorticoid therapy as soon as possible. Moreover, as demonstrated in previous trials, the potential effect of a treatment seems to be more pronounced if the treatment is initiated early after the onset of STEMI. In addition to reperfusion induced inflammation, ischemia itself, immediately after occlusion of the artery, induces inflammation. Hence, initiation of the intervention is needed as close to the debut of symptoms as possible to inhibit the inflammation adequately and effectively in relation to STEMI. Thus, by performing intervention in the *pre-hospital* setting, we expect that participation in the trial will have the potential to produce a direct clinically relevant benefit for the patient resulting in a measurable health-related improvement alleviating the suffering and potentially improving the health of the patient and the prognosis of the medical condition. Moreover, the trial relates directly to the patient's medical condition and it is not possible to obtain prior informed consent from the patient and to supply prior proper information about the trial due to the acute nature of STEMI. Therefore, the trial is of such nature that it may be conducted solely in emergency situations. This encounters that the usual requirements about a 24-hour time for consideration and discussion with a lay representative cannot be met. We are aware of this relationship, which we found reasonable to deviate from.

Patients with STEMI are unable to provide prior informed consent and receive prior information on the clinical trial, as STEMI is a sudden, life-threatening, and serious medical condition requiring urgent treatment. Most of patients with STEMI treated with primary PCI at Rigshospitalet, Denmark have less than 30 minutes of transportation to the hospital. Therefore, it is not possible within the therapeutic window to supply all prior information about the trial and obtain prior informed consent. The including RD is continuously in touch with the paramedic in the ambulance by

telephone from the time of screening until randomization has been done. The including RD has all necessary information about the patient through medical records to ensure that the patient does not have any objections towards participating in the trial. In addition, the trial and intervention pose minimal risk and burden on the patient in comparison with standard treatment of the patient's condition, as Solu-Medrol (methylprednisolone) is a well-known and thorough investigated drug of minimal risk if administrated as a single dose administration.

Most of patients with STEMI will be able to provide informed consent and receive adequate information about the trial as soon as possible following the acute treatment. Continuation of participation in the trial will therefore be ensured with adequate information and informed consent as soon as possible following the acute treatment.

The Principal Investigator(s) will ensure:

1. Each patient will give informed consent based on the ICF as soon as possible following the acute treatment (primary PCI) including signature of the ICF.
2. In patients who are unable to provide informed consent as soon as possible following the primary PCI procedure, informed consent will be obtained by the legally designated representative including signature of the ICF followed by informed consent from the patient as soon as possible.
3. In patients who remain unable to provide informed consent 3 days following admission, the legally designated representative and nearest relative will provide informed consent and signature of the ICF on behalf of the patient. Informed consent from the patient will subsequently be obtained as soon as possible.
4. In patients who do not wish to participate in the study, the data collection of the patient will be stopped immediately.
5. In cases of death prior to informed consent, the legally designated representative will provide informed consent as soon as possible. The closest relative will be informed about participation in the study including the use of the patient's data in relation to the study. In case of any objection or unwillingness towards participation in the study, the patient will then be excluded from the study.
6. All signed ICFs are encrypted and kept in REDCap.
7. A copy of the signed ICF is offered to the patient.

9.3 Benefits and Risks

The expectation of the trial is, that administration of 250 mg methylprednisolone in the *pre-hospital* setting will reduce the infarct size and thereby better prognosis in patients with STEMI undergoing primary PCI. The patients included in the study will have a closer medical control due to participation and detailed follow-up, however, there is no guarantee that the individual participants achieve any benefits from participating in the trial. The study will not delay or interfere with standard therapeutic or diagnostic procedures. Only patients who consent are included in the trial.

The infusion of methylprednisolone needs to be administrated in the acute phase in the *pre-hospital* setting before primary PCI in order to alleviate the inflammatory response following reperfusion. Intervention in the *pre*-hospital setting before informed consent is therefore needed. Methylprednisolone is easy to administrate, has an acute effect, and implementation of methylprednisolone administration in the *pre-hospital* setting is feasible to be a part of the routine clinical care of patients with STEMI undergoing primary PCI. The study medicine is approved and recommended in other medical conditions and is expected to be of minimal risk to patients. If successful, the increased knowledge on the therapeutic beneficial potential of methylprednisolone will 1) increase scientific knowledge, and 2) improve clinical outcomes in patients with STEMI by reducing the number of cardiac deaths and reducing the number of patients with HF following STEMI. The information gathered from this study may therefore help future patients and change future clinical guidelines.

10. Statistics

The data management work up and statistical analyses will be performed at Rigshospitalet, Copenhagen University Hospital, Denmark.

10.1 Power Calculations

The primary endpoint is final infarct size (% of left ventricle mass) measured by LGE on CMR at 3 months. Based on results of the CMR sub-studies of the DANAMI-3 trial[40-45], the mean final infarct size measured by LGE on CMR is 13% with a standard deviation (SD) of 9% in patients with STEMI. To demonstrate a relative reduction in final infarct size of 20% with a two-sided alpha level of 0.05 and a power of 80%, recruitment of 378 patients is needed. A drop-out rate of 40% is expected for the primary endpoint. Therefore, we expect to randomize 530 patients in total. However, patients will be included until 378 patients have completed the CMR at 3 months. The power calculations have been calculated by a biostatistics professor.

10.2 Statistical Analysis

For patients fulfilling all in- and exclusion criteria, the treatment groups will be compared as intention-to-treat principle. Additional as-treated analyses will be performed. Patients included that subsequently are excluded due to meeting exclusion criteria will be considered screen failure. These patients will still be followed for safety reasons. Differences between group means/medians will be assessed with parametric or non-parametric statistics accordingly. Chi-square analysis or Fisher's exact test will be used to test differences between categorical variables. Differences between groups in the time-to event secondary endpoints will be assessed using Kaplan-Meier and Cox regression model. In case of an event or until last patient has been followed, the patients will be censored. Probability of survival will be displayed using Kaplan-Meier methodology. Hazard ratios between groups will be calculated using a Cox proportional hazard model. A two-tailed P-value <0.05 is considered statistically significant.

10.3 Pre-specified Subgroup Analyses

The primary endpoint will be compared between treatment groups in pre-specified subgroup analyses: age, gender, pre-PCI TIMI flow, culprit in LAD, time from first medical contact to primary PCI, and duration from symptom to primary PCI. These subgroups will be analyzed using interaction tests. The pre-specified subgroups are factors that are known to influence the clinical course in patients with STEMI.

11. Sub-study

Prior studies have shown that reperfusion injury following primary PCI in patients with STEMI is equivalent to damage in the microcirculation causing microvascular dysfunction. The microvascular function can be investigated by thermodilution-derived parameters including CFR and IMR as well as absolute flow and absolute resistance. Prior studies in STEMI patients have shown that IMR has a prognostic value and correlates to MVO. Glucocorticoid may limit reperfusion injury by decreasing the inflammatory response in direct proximity to the microcirculation. To investigate this hypothesis, we will measure CFR and IMR on eligible patients included in PULSE-MI directly after initial primary PCI but before the catheter is removed.

CFR and IMR is measured by a pressure gauge wire and a 2-minute infusion of adenosine. This procedure will only prolong the normal PCI by 5 minutes and does not add any additional risk to the procedure and does not increase the risk of complications. The patient may experience a brief

feeling of shortness of breath during the infusion of adenosine but is not associated with other complications or side effects. Adenosine is often used in standard coronary angiography (CAG) and PCI procedure.

12. Data Storing and Handling of Personal Data

All study data will be stored pseudo-anonymously for 25 years. Data of the included patients with STEMI will be stored at an electronic database, RedCap, where data will be blinded, and patients will be identified by a study identification number only. Study participation will be annotated in the medical record of the patient.

13. Assessments of Safety and Harm

Mortality following STEMI has decreased after implementation of primary PCI and drug-eluting stents (DES), yet the 1-year mortality rate has unsatisfactorily plateaued at approximately 10%. Primary PCI is performed daily at Rigshospitalet, Copenhagen University Hospital, and the risks associated with the procedure is well known. STEMI is often complicated by a myocardial dysfunction leading to HF and arrhythmias during the acute phase. However, mechanical complications such as free wall rupture, ventricular septal rupture, and papillary muscle rupture may also occur in the setting of STEMI[10]. These complications are well known, and, as part of standard care, an echocardiography is performed in all patients admitted for primary PCI to assess potential mechanical complications and the function of the left and right ventricle. AEs/SAEs potentially related to the intervention of the study will include: death, infection, gastroduodenal ulcer, hypertension, fluid retention resulting in peripheral edema, anaphylaxis, and skin complications such as acne, atrophy, purpura, and wound healing complications. The half-life of methylprednisolone is 12 hours, and the drug is considered eliminated following 5 half-times. Therefore, AEs will be recorded for the first 3 days following intervention.

13.1 Adverse Events (AEs)

AEs are considered any untoward medical occurrence in the patient to whom intervention (methylprednisolone) has been administrated and which does not necessarily have a causal relationship with this treatment. The AEs that will be recorded is infection and any skin complications.

13.2 Serious Adverse Events (SAEs)

The Principal Investigator and other Investigators will report all SAEs to Sponsor within 24 hours of awareness. All SAEs are any toward medical occurrence that at any dose requires inpatient hospitalization or prolongation of existing hospitalization, results in persistent or significant disability or incapacity, results in a congenital anomaly or birth defect, is life-threatening, or results in death.

All SAEs occurring from the time the study medicine has been administrated until the patient has completed the trial will be reported by filling out and sending the SAE form within 24 hours, after the investigator becomes aware that the event meets the protocol definition of an SAE. Patients will therefore be followed for late-onset SAEs until completion of the trial (1 year).

Hospitalization and prolongation of hospitalization is defined as presentation at hospital for an urgent, unscheduled emergency department visit or hospital admission (>24 hours).

In the electronic database, REDCap, the following events will be reported: Death, hospitalization for heart failure, recurrent AMI, stroke/TIA, bleeding, and cardiac arrhythmia.

13.3 Suspected Unexpected Serious Adverse Reactions (SUSARs)

A SUSAR is defined as a suspected, serious adverse reaction, the nature, severity, or outcome of which is not consistent with the reference safety information. *'Unexpected'* reactions are defined using the summary for product characteristics (SPC) for Solu-Medrol[32]. Sponsor of the trial will be registered in the EudraVigilance system prior to initiation of the trial and is responsible of reporting any SUSARs to the EudraVigilance systemand the ethics committee as soon as possible, at latest 7 days following awareness of event if life-threatening. If the SUSAR is not life-threating the event will be reported at latest 15 days following awareness of the event.

14. Finance and Insurance

This study is investigator-initiated, and the study will be funded by external foundations for medical research. No funding has been obtained at present time. Sponsor is responsible for funding the costs of the trial until funding has been achieved. When funding is achieved, the ethics committee will be noticed. The preliminary budget of the study is approximately 6.2 million DKK.

All patients participating in the trial are insured by the Patient Compensation Association and the Danish patient insurance.

15. Publication

All results (positive, negative, or inconclusive) will be published in international peer-reviewed journals. The data will also be presented at national and international congresses. No participants will be identified in any reports or publications. All analyses will be conducted by the investigators of the trial. The trial will not be unblinded until acceptance from the steering committee. The publication policy of the trial data will be uploaded to the Clinical Trials Information System (CTIS) database within one year after end of trial in accordance with Clinical Trial Regulation 536/2014 Annex IV.

The authorship order: Dr. Jasmine Melissa Madsen will be first author and Dr. Jacob Lønborg and Professor Thomas Engstrøm will be shared last author. The following authorships will be granted in according to ICMJE guidelines. However, all co-investigators will be offered authorship.

16. Annual Safety Report

Throughout the whole study, annual safety reports will be uploaded to the CTIS database every year. Annual safety reports will not be sent in the period between trial completion and the 10 years follow up on mortality and hospitalization of HF.

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Appendix A (Cardiac Magnetic Resonance Protocol and Endpoints)

Purpose:

- To describe the cardiac magnetic resonance (CMR) imaging scan for the PULSE-MI trial.
- To determine final infarct size by late gadolinium enhancement (LGE) on CMR 3 months after ST-Segment Elevation Myocardial Infarction (STEMI).

Setting:

All CMR scans will be performed at Rigshospitalet, Copenhagen University Hospital, Denmark.

Key points

The CMR scanner is of 1.5 Tesla field strength with a standard phased array cardiac surface coil. The CMR scans should be performed twice post-STEMI. The first scan will be performed within 48 hours (Acute scan), and the second scan will be performed 3 months following STEMI (Follow-up scan).

Acute scan: The acute scan will be performed preferably within 48 hours of STEMI due to logistics at Rigshospitalet, Denmark. However, there is no set upper limit to the time from index event to scan, as long as it is performed within 5 days of index event (acute phase).

Follow-up: The follow-up scan should be performed as close to 3 months following the index event as possible.

The primary endpoint of the study is final infarct size (% of the left ventricle (LV) mass) measured by LGE on CMR 10-15 minutes after gadolinium contrast (0.15 mmol/kg) is given.

CMR set-up:

CMR will only be performed in patients who have given written and oral informed consent and if no contraindications exist. The patient will undergo a CMR safety check including renal function prior to scan.

Contraindications to CMR in the study: Ferrous metal foreign body, severe claustrophobia, decreased renal function (eGFR <30 ml/min).

Patients will be profoundly prepared and informed about the CMR scan. Following the scan, patients will be briefed shortly of the findings.

CMR protocol (Acute and Follow-Up Scan):

1	Patient preparation
2	Localizers and planning
3	Cine long axis (2ch, 3ch, 4ch) Short axis (SA) localizer of ventricles and subsets (basal, mid, apical)
4	Cine para-coronal
5	T1-mapping pre-contrast (SA subsets)
6	T2* pre-contrast (full LV-coverage)
7	**Contrast: 0.15 mmol/kg**
8	Dynamic rest
9	Function, SSFP (SAX-stack, full LV-coverage)
10	Late gadolinium enhancement (SAX-stack, 2ch, 3ch, 4ch).
11	Aortic flow rate
12	Patient Briefing

Contrast administration:

The Intravenous contrast agent is gadobutrol (Gadovist®, Bayer; 1.5 mmol/ml solution for injection) which will be administrated in one single-dose. The dose will be given as 0.15 mmol/kg. An automated pump injector will be used for intravenous injection of Gadovist. The amount of isotonic saline (0.9%) given with Gadovist is double the amount of contrast given (0.30 mmol/kg).

Poor breath-holding:

In patients with poor breath-holding, compressed sense sequences will be used to conduct CINE images. These sequences are free breathing.

CMR endpoints

The primary efficacy parameter for the clinical trial is the amount of *infarct size (% of LV mass)* revealed by LGE 10-15 minutes after contrast administration on CMR performed 3 months following STEMI.

Acute CMR

- Microvascular obstruction on LGE (presence/absence)
- Extent of MVO on LGE
- Infarct size on LGE
- Area at risk
- Extent of edema
- Initial myocardial salvage index (acute infarct size/area-at-risk)
- LV ejection fraction (LVEF)
- Hemorrhage

Follow-up CMR

- Final infarct size (Primary endpoint)
- Myocardial salvage index (final infarct size/area at risk)
- Change in infarct size 3 months after procedure
- LVEF
- Change from baseline LVEF to follow-up LVEF

Analyses of CMR

All analyses will be performed in Circle Cardiovascular Imaging (CVI42).

Appendix B (End-point Definitions)

The clinical event committee (CEC) will review and adjudicate all occurrences of the clinical end-points according to the 2014 ACC/AHA Key Data Elements and Definitions for Cardiovascular Endpoint Events in Clinical Trials[46].

1. All-Cause Mortality
2. Recurrent Myocardial Infarction
3. Hospitalization for Heart Failure
4. Stroke/TIA

Mortality will to the extent possible be classified according to underlying disease. The CEC will classify the mortality as cardiovascular or non-cardiovascular death.

Cardiovascular mortality

Mortality considered as cardiovascular unless it is clearly attributable to another cause and thus includes:

- Death due to proximate cardiac cause (e.g. myocardial infarction, cardiac tamponade, worsening heart failure).
- Death caused by non-coronary vascular conditions such as neurological events, pulmonary embolism, ruptured aortic aneurysm, dissecting aneurysm, or other vascular disease.
- All procedure-related deaths, including those related to a complication of the procedure or treatment for a complication of the procedure.
- All valve-related deaths including structural or non-structural valve dysfunction or other valve-related adverse events.
- Sudden or unwitnessed death.
- Death of unknown cause.

Non-cardiovascular mortality

Any death in which the primary cause of death is clearly related to another condition (e.g. infection, cancer, suicide, accident, pulmonary, hepatobiliary, gastrointestinal, renal, overdose, neurological (excluding stroke and or cardiovascular hemorrhage of central nervous system).

Hospitalization for heart failure is prolongation of hospitalization because of worsening of heart failure or presentation of the patient for an urgent, unscheduled clinic/office/emergency department visit or hospital admission (>24 hours), with a primary diagnosis of heart failure (HF), AND
where the patient exhibits new or worsening symptoms of HF on presentation, AND
has objective evidence of new or worsening HF*, AND
receives initiation or intensification of treatment specifically for HF.

*Objective evidence consists of at least 2 physical examination findings OR at least 1 physical examination finding and at least 1 laboratory criterion (chest x-ray or BNP) of new or worsening HF on presentation.

Table 3. Value definition heart failure

Values	Value definition
New or worsening symptoms of HF	Dyspnea Decreased exercise tolerance Fatigue Volume overload
Objective evidence of new or worsening HF	*Either* at least one on physical exanimation: Peripheral edema Increasing abdominal distention or ascites Pulmonary rales/ crackles/crepitations Increased jugular venous pressure and/ or hepatojugular reflux S3 gallop Clinically significant or rapid weight gain *Or* at least on laboratory criterion: Biomarker increases BNP/ NT-pro BNP (> upper reference limit). In patients with chronically elevated natriuretic peptides, a significant increase above baseline is required.

	Radiological evidence of pulmonary congestion: imaging findings consistent with increased intravascular blood volume in the lungs.
Receives initiation or intensification of treatment specifically for HF	Augmentation of oral diuretic therapy Intravenous diuretic, inotrope, or vasodilator therapy Mechanical or surgical intervention including any LV assist device and mechanical fluid removal

Appendix C (the PULSE-MI committee members and investigators)

Steering committee:

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Clinical event committee (CEC):

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Data Safety Monitoring Board (DSMB):

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Nico Pijls (suggested)

Won Yong Kim (suggested)