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Randomized Trial of Early mTOR Inhibition in Patients with Acute ST-Segment Elevation Myocardial Infarction

Brief title: mTOR Inhibition in acute STEMI

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ABSTRACT

Background: Early inflammation following acute ST-segment elevation myocardial infarction (STEMI) treated by primary percutaneous coronary intervention (PCI) affects myocardial infarct (MI) size and left ventricular remodeling. The mammalian target of rapamycin (mTOR) is involved in the enhanced inflammatory response and its inhibition has exerted beneficial effects on MI size in preclinical models of acute MI.

Objectives: The Controlled Level EVERolimus in Acute Coronary Syndromes trial (CLEVER-ACS) evaluated the effects of targeting inflammation by mTOR inhibition in patients with STEMI undergoing PCI.

Methods: CLEVER-ACS was a randomized, multicenter, international, double-blind, placebo-controlled trial. A total of 150 patients with STEMI undergoing PCI were randomly assigned to oral everolimus (days 1-3: 7.5 mg qd, days 4-5: 5.0 mg qd) or placebo for 5 days. The primary endpoint was the change in myocardial infarct size, the secondary endpoint the change in microvascular obstruction (MVO) from baseline (12 hours – 5 days after PCI) to 30 days as assessed by cardiac magnetic resonance imaging (CMR).

Results: The changes in MI size from baseline to 30 days, the primary endpoint, were -14.2 (95% CI -17.4 to -11.1) g and -12.3 (95% CI -16.0 to -8.7) g in the everolimus and placebo groups (p=0.99). Corresponding changes in MVO were -4.8 (-6.7 to -2.9) g and -6.3 (-8.7 to -4.0) g in the everolimus and placebo groups (p=0.14). Adverse events did not differ between the study groups.

Conclusions: Among STEMI patients undergoing PCI, early mTOR inhibition with everolimus did not reduce MI size or MVO at 30 days.

CONDENSED ABSTRACT

Early inflammation following acute ST-segment elevation myocardial infarction (STEMI) treated by primary percutaneous coronary intervention (PCI) affects myocardial infarct (MI)

size. CLEVER-ACS evaluated the effects of early mTOR inhibition by everolimus on MI size in a total of 150 STEMI patients. The changes in MI size from baseline to 30 days as assessed by cardiac magnetic resonance imaging, the primary endpoint, were -14.2 (95% CI -17.4 to -11.1) g and -12.3 (95% CI -16.0 to -8.7) g in the everolimus and placebo groups (p=0.99). Hence, among STEMI patients undergoing PCI, everolimus did not reduce MI size at 30 days.

KEY WORDS: Acute myocardial infarction; percutaneous coronary intervention; inflammation; everolimus.

ABBREVIATIONS

CANTOS	Canakinumab Antiinflammatory Thrombosis Outcomes Study
CEC	Clinical Event Committee
CLEVER-ACS	Controlled Level EVERolimus in Acute Coronary Syndromes
CMR	Cardiac magnetic resonance
COLCOT	Colchicine Cardiovascular Outcomes Trial
CTU	Clinical Trials Unit
CX	Left circumflex coronary artery
DSMB	Data Safety Monitoring Board
ECG	Electrocardiogram
eGFR	Estimated glomerular filtration rate
HIV	Human immunodeficiency virus
LAD	Left anterior descending coronary artery
LGE	Late gadolinium enhancement
LVEDV	Left ventricular end-diastolic volume
LVEF	Left ventricular ejection fraction
LVESV	Left ventricular end-systolic volume

MVO	Microvascular obstruction
mTOR	Mammalian target of rapamycin
NSTEMI	Non-ST-segment elevation myocardial infarction
PCI	Percutaneous coronary intervention
RCA	Right coronary artery
SD	Standard deviation
STEMI	ST-segment elevation myocardial infarction
TIMI	Thrombolysis in Myocardial Infarction

CLEVER-ACS, ClinicalTrials.gov number, NCT01529554.

INTRODUCTION

Inflammation plays an important role in the pathogenesis of acute myocardial infarction, involving multiple cell types such as neutrophils, monocytes-macrophages, lymphocytes, and as well as numerous proinflammatory cytokines.(1, 2) Inflammation is considered one of the key triggers for coronary plaque rupture or erosion, eventually causing thrombus formation and vessel occlusion.(3) Following revascularization, inflammatory responses and oxidative stress also critically affect ischaemia/reperfusion injury and microvascular obstruction (MVO), and may subsequently impact myocardial infarct size, adverse left ventricular remodelling, and heart failure.(4-7) There is accumulating evidence supporting a beneficial role of targeted anti-inflammatory therapies for preventing adverse events in patients with coronary artery disease.(8, 9) Among patients with a recent myocardial infarction, inhibition of interleukin-1β by the monoclonal antibody canakinumab lowered cardiovascular events in the Canakinumab Antiinflammatory Thrombosis Outcomes Study (CANTOS), and colchicine reduced cardiovascular events in the Colchicine Cardiovascular Outcomes Trial (COLCOT) and Low-Dose Colchicine (LoDoCo) 2 trial.(8-10)

The mammalian target of rapamycin (mTOR) is a protein kinase involved in the regulation of inflammatory cascades and the orchestration of immune cells.(11) Its expression has further been implicated in ischaemia/reperfusion injury, the regulation of cardiomyocyte apoptosis and necrosis, and the development of myocardial hypertrophy.(7, 11-13) Inhibitors of mTOR such as everolimus exert a broad spectrum of anti-inflammatory effects, (14-16) and rapamycin has been shown to reduce myocardial infarct size and adverse left ventricular remodelling in an animal model of myocardial infarction.(17) These data support the concept that targeting the early and high surge of inflammation by mTOR inhibition in patients with acute ST-segment elevation myocardial infarction (STEMI) may favourably affect myocardial infarct size. Thus, the Controlled Level EVERolimus in Acute Coronary Syndromes

(CLEVER-ACS) trial was designed to assess the effects of everolimus on myocardial infarct size as assessed by cardiac magnetic resonance (CMR) imaging in patients with STEMI.

METHODS

Study design

This prospective, randomized, international, multi-centre, double-blind, placebo-controlled trial (NCT01529554), coordinated by the University Hospital Zurich, Switzerland, evaluated the efficacy and safety of everolimus (RAD, [40-O-[2-hydroxyethyl]-rapamycin], Votubia®, SDZ RAD, RAD001, Novartis) for the reduction of myocardial infarction size in STEMI patients.(18) Patients with STEMI due to proximal occlusion of a coronary artery undergoing reperfusion therapy by means of primary PCI were screened between 0 and 5 days following PCI and randomized in a 1:1 ratio to receive oral everolimus (days 1-3: 7.5 mg qd, and days 4-5: 5.0 mg qd) or matching placebo for 5 days. Randomization was stratified by center and performed based on computer-generated randomly varying blocks of 2, 4, and 6 numbers using the statistical software Stata 13.1 (StataCorp LLC 2021. Stata Statistical Software. Texas. USA). The access to the codes was controlled and documented locally and centrally by the Clinical Trials Unit (CTU) Bern, University of Bern, and the Institute for Hospital Pharmacy, University Hospital Bern, both Bern, Switzerland. All patients were treated with drug-eluting stents and received concomitant evidence-based therapies, including high-dose statin treatment, as recommended by the prevailing European Society of Cardiology guidelines.(19) Patients were assessed at baseline, at discharge (or within 5 days), and at 30 days (±3 days) after randomization for efficacy and safety. Study visits included the assessment of adverse events, a physical examination, laboratory analyses, and a 12-lead electrocardiogram (ECG). Routine blood samples were collected at baseline, at discharge (or within 5 days), and at 30 days, and laboratory parameters were measured using standard protocols. In patients participating in an optional biomarker substudy, Core Lab blood samples (30-40 ml) were drawn at each time point and urine samples (8 ml) at 12-24 hours.

Cardiac magnetic resonance imaging was performed at baseline (12 hours to 5 days after PCI) and at 30 days follow-up.

An independent Clinical Events Committee (CEC) assessed all adverse events and an independent Data Safety Monitoring Board (DSMB) ascertained and adjudicated all serious adverse events. The study was approved by the local ethics committees at each participating center, and all patients provided written informed consent prior to inclusion in the study. The study was conducted following the principles of the Declaration of Helsinki, and in accordance with local law and regulations.

Study population

Between November 2014 and October 2021, 150 patients (75 per treatment arm) with STEMI undergoing primary PCI were enrolled at 8 centers in Switzerland and Germany. Patients were eligible if they were between 18 and 90 years of age, experienced a first myocardial infarction, had chest pain duration of >10 minutes, ST-segment elevations >1 mm in at least 2 contiguous leads, new left bundle branch block, or posterior myocardial infarction with ST-segment depression >1 mm in at least 2 contiguous leads, angiographic evidence of occlusion of the proximal third of either the left anterior descending coronary artery (LAD), the circumflex coronary artery (CX), or the right coronary artery (RCA), or occlusion of the mid segment of the LAD or RCA when the latter reached the left ventricular apex, primary PCI performed within 24 hours of chest pain onset with implantation of a drug-eluting stent in the culprit lesion, and were able and willing to give written informed consent and adhere to the study protocol. Exclusion criteria comprised contraindications (i. e. known drug hypersensitivity or allergy) to mTOR inhibitors or placebo, contraindications to CMR or CMR contrast agent, concomitant use of immunosuppressants (e. g. steroids, methotrexate, cyclosporine, tumor necrosis factor [TNF]-α antagonists, or rituximab) up to 4 weeks prior to the index STEMI event, mechanical complications of acute myocardial infarction, multivessel coronary artery disease requiring revascularization of a non-culprit artery within the trial

period, scheduled PCI within 30 days, major elective surgery during the duration of the trial, estimated glomerular filtration rate (eGFR) <30 ml/min, malignancy (unless cured or in remission for >5 years), known chronic infection (human immunodeficiency virus [HIV], tuberculosis, empyema), known or suspected non-compliance, drug or alcohol abuse, pregnancy or lactation, women and men of childbearing potential not willing to use reliable contraception for the duration of the trial, participation in another trial with an investigational product within 30 days or 5 terminal half-lives, and positive PCR for SARS-CoV-2 or at least one positive answer to symptoms/contact SARS-CoV-2 questionnaire (since September 2020).

The primary endpoint was evaluated in patients with available efficacy data.

Study endpoints

The primary endpoint was the change in myocardial infarct size measured in grams (g) from baseline (12 hours – 5 days after PCI) to 30 days (±3 days) as assessed by CMR. The secondary endpoint was the change in MVO from baseline (12 hours – 5 days after PCI) to 30 days (±3 days) as assessed by CMR. Tertiary endpoints comprised corresponding changes in left ventricular volumes. Safety endpoints comprised any clinical events within 30 days.

Cardiac magnetic resonance imaging

Cardiac magnetic resonance imaging protocols included functional and morphological sequences for volumetric measurements, T1- and T2-weighted sequences for tissue characterization, and late gadolinium enhancement (LGE) for imaging of scar (gram or percentage of left ventricular mass) and MVO (gram or percentage of left ventricular mass).(20)

Functional imaging of the left ventricle was performed using standard ECG-triggered, steady state-free precession acquisitions (in-plane resolution: typically 1.5 mm x 1.5 mm; slice thickness: 8 mm) during repetitive breath holds in contiguous short-axis orientation, covering the entire left ventricle and 3 long-axis orientations (4-, 3-, and 2-chamber

orientation). A bolus of conventional extracellular gadolinium-chelates contrast medium at a dose of 0.20 mmol/kg of body weight was administered to assess scar tissue, using an inversion recovery fast-gradient echo imaging sequence (in-plane resolution: typically 1.5 mm x 1.5 mm; slice thickness: 8 mm). Scar imaging was performed 15 minutes after administration of contrast medium in the same slice position as the functional data.

Cardiac magnetic resonance imaging data analysis was performed by experienced physicians blinded to treatment groups in the CMR Core Laboratory at the University Hospital Zurich, Switzerland, using dedicated cardiac analysis software (GTVolume, Gyrotools Ltd, Zurich, Switzerland). Left ventricular end-diastolic (LVEDV) and end-systolic (LVESV) volumes, left ventricular ejection fraction (LVEF), and left ventricular mass were assessed. The extent of MVO, delineated as dark areas in the core of the necrotic zone in the LGE images, was quantified by manually contouring the dark core areas.

Statistical analysis

Sample size calculation was based on the assumptions for the primary endpoint. Myocardial infarction scar mass was expected to be 23 g with a standard deviation (SD) of 14 g in the control group.(21, 22) An intra-patient correlation between baseline and 30-day follow-up myocardial infarction scar mass of 0.55 was assumed, representing a safe assumption and considering the baseline variation in myocardial infarction scar mass among patients as well as the high CMR reproducibility.(21) Based on an animal model of myocardial infarction, the treatment was expected to reduce the myocardial infarction scar mass by 34.7%.(17) Using a two-sided, two-sample repeated measures test on the change in myocardial infarct scar mass with a SD of 14 g in both arms, an intra-patient correlation of 0.55, and an alpha level of 5%, 75 patients were needed in each group for the study to have a power of at least 90% to detect a difference in the change in myocardial infarct scar mass of 7 gr.

All efficacy analyses were performed in the intention-to-treat population, defined as all randomized patients with CMR performed both at baseline and at 30 days. In addition, analyses were performed in the per-protocol population, defined as all randomized patients with CMR performed both at baseline and at 30 days, who took all 13 tablets according to their randomization. The consistency of the treatment effect on the primary endpoint was assessed among two prespecified subgroups, defined by the size of the scar mass at baseline and the time delay between PCI and first study drug administration (with time of PCI being defined as time of first balloon inflation).

Baseline, efficacy, and safety data are reported using descriptive statistics. Continuous variables are presented as mean ± standard deviation, or median and interquartile range. Categorical variables are given as numbers and percentages. The primary and secondary endpoints are presented as difference with 95% confidence interval (CI). Comparisons between groups were performed with the two-sided unpaired t-test or the Mann-Whitney U test for continuous variables and the Chi-square or Fisher's exact test for categorical variables. Within-group comparisons (i.e. baseline versus 30 days) were performed using the paired t-test or the Wilcoxon signed-rank test. Mixed-effects models were used to compare primary and secondary outcome measures as well as lab values between treatment groups, adjusting for baseline measurements in each model and considering the center trials as the random effect. The normality assumption of the outcome was tested by using the Kolmogorov-Smirnov (K-S) test.

All testing was two-sided and a p-value of <0.05 was considered statistically significant. All statistical analyses were performed using Stata 17.0 (StataCorp LLC 2021. Texas. USA).

RESULTS

Patient characteristics

A total of 150 patients with STEMI were enrolled. One patient withdrew consent. Four patients were lost to follow-up and 2 refused follow-up. Baseline CMR was available in 142

patients and CMR at 30 days in 137 patients, respectively. A total of 135 patients had data available for the analysis of the primary endpoint (Figure 1).

Baseline characteristics are summarized in Table 1. Mean age of the patients was 61.9±11.2 years and 15% were women. Hypertension was present in 41% of patients, dyslipidemia in 46%, and type 2 diabetes in 9%. The proportion of patients on aspirin, statins, angiotensin converting enzyme inhibitors, and beta blockers was high.

Procedural characteristics are summarized in Table 2. The LAD was the culprit artery in 58% of patients, the CX in 10%, and the RCA in 31%, respectively, and one patient presented with left main disease. Total vessel occlusion was observed in 83% of patients. Post-PCI Thrombolysis in Myocardial Infarction (TIMI) 3 flow was achieved in 95% of patients.

Baseline CMR was performed 1.6±1.6 days and 1.3±1.3 days after randomization in the everolimus and placebo groups (p=0.14). Follow-up CMR at 30 days was performed 31±7 days and 33±8 days after randomization in the everolimus and placebo groups (p=0.24).

Laboratory results

Laboratory parameters are given in Table 3. Baseline high-sensitivity cardiac troponin T (hs-cTnT) levels were 4.54 (2.29-9.76) µg/l and 4.79 (1.77-12.10) µg/l in the everolimus and placebo groups. Baseline creatine kinase (CK) levels were 994 (444-2190) U/l and 925 (374-1739) U/l in the everolimus and placebo groups. Both groups showed a decrease in cardiac biomarkers from baseline to 30 days without changes between the groups.

Efficacy results

Efficacy results are summarized in Table 4. In the intention-to-treat population (n=135), the changes in myocardial infarct size from baseline (12 hours – 5 days after PCI) to 30 days were -14.2 (95% CI -17.4 to -11.1) g and -12.3 (-16.0 to -8.7) g in the everolimus and placebo groups, respectively (Figure 2, Central Illustration). There was no difference in the change from baseline to 30 days among groups (p=0.99). Corresponding changes in MVO were -4.8 (-6.7 to -2.9) g and -6.3 (-8.7 to -4.0) g in the everolimus and placebo groups, respectively,

showing no difference between groups (p=0.14). Changes in left ventricular volumes from baseline to 30 days did not differ between the groups.

In the per-protocol population (n=132), the changes in myocardial infarct size from baseline (12 hours – 5 days after PCI) to 30 days were -14.4 (-17.7 to -11.0) g and -12.5 (-16.1 to -8.8) g in the everolimus and placebo groups, respectively. There was no difference in the change from baseline to 30 days between groups (p=0.86). Corresponding changes in MVO were -4.4 (-6.4 to -2.5) and -6.4 (-8.9 to -4.0) g in the everolimus and placebo groups, without difference between groups (p=0.55). Changes in left ventricular volumes did not differ between groups.

No treatment effect on the primary endpoint was observed in the prespecified subgroups (size of scar mass at baseline and time from PCI to first study drug administration, Supplementary Table 1).

Safety results

Everolimus was generally well tolerated (Table 5). Adverse events at 30 days were reported in 45% and 39% of patients in the everolimus and placebo groups (p=0.44). Most events were mild and deemed not to be associated with the study drug. Serious adverse events were reported in 21% and 15% of patients in the everolimus and placebo groups (p=0.20), and severe or life-threatening serious adverse events in 1.3% and 2.6% (p=1.00), respectively.

DISCUSSION

This study, representing one of the largest contemporary randomized CMR trials in acute myocardial infarction patients, showed that inhibition of mTOR in the early phase after STEMI did not exert favorable effects on myocardial infarct size nor on left ventricular remodeling or MVO at 30 days in patients undergoing primary PCI. Moreover, the drug did not raise significant safety concerns if given as a short-term administration for 5 days.

Targeting mTOR in acute myocardial infarction.

Inflammation is a characteristic feature throughout distinct stages of atherosclerotic lesion formation, at the time of plaque rupture or erosion causing acute myocardial infarction, as a response to the vessel trauma induced by PCI, as part of the ischaemia/reperfusion injury, and during the post-infarction repair phase.(4-6) Components of the innate and adaptive immune system including the pro-inflammatory cytokines interleukin-1 and tumor necrosis factor-α exert prominent functions in MVO and post-infarct remodeling. A persistent and pronounced pro-inflammatory reaction after acute myocardial infarction is considered to adversely affect infarct size, left ventricular remodeling, and the development of heart failure.(4) In support of this concept, the amount of circulating monocytes ($CD14^{+}CD16^{-}$) in patients after an acute myocardial infarction was inversely related to the extent of myocardial salvage as well as to recovery of left ventricular function at 6 months as assessed by CMR,(23) and elevated interleukin-8 levels were related to a larger myocardial infarct size and an increased prevalence of MVO in STEMI patients.(24) In the ASSessing the effect of Anti-IL-6 receptor treatment in Myocardial Infarction (ASSAIL-MI) trial, acute treatment with the interleukin-6 inhibitor tocilizumab increased myocardial salvage and reduced MVO in patients with acute STEMI, without affecting myocardial infarction size.(25) Along these lines, it is not surprising that recent clinical trials including CANTOS and COLCOT have shown favorable clinical outcomes using strategies targeting late inflammation in patients with acute myocardial infarction .(8, 9) These trials included patients with STEMI and non-ST-segment elevation myocardial infarction (NSTEMI) diagnosed according to the universal myocardial infarction criteria, with the majority undergoing coronary revascularization by either PCI or coronary artery bypass grafting and long-term anti-inflammatory therapy initiated within or after 30 days following the index event.(8, 9) Given the pivotal role of the protein kinase mTOR in the regulation of multiple inflammatory pathways and the observed beneficial effects of mTOR inhibition on myocardial infarct size and left ventricular

remodeling in animal models of acute myocardial infarction,(11-13, 17) therapeutic concepts targeting mTOR for the reduction of myocardial infarct size seemed promising.

CLEVER-ACS was a proof-of-concept trial, designed to test whether mTOR inhibition may attenuate the exaggerated inflammatory response that occurs in patients with acute STEMI after PCI and thereby reduce myocardial infarct size as assessed by CMR, considered a valid surrogate endpoint with robust association with mortality and heart failure hospitalization.(26-31) The change in myocardial infarct size from baseline to 30 days was selected as primary endpoint to adjust for the variability in myocardial infarct size in STEMI patients. As everolimus did not affect myocardial infarct size following STEMI, it is possible that the mTOR pathway plays a less important role in the pathogenesis of myocardial scar formation, MVO, and left ventricular remodeling than anticipated. Post-hoc analyses did not suggest that myocardial infarct size or the time interval between PCI and everolimus administration influence the effect of everolimus on myocardial scar formation.

Given that the dosage of everolimus used in CLEVER-ACS was similar to the one used in the successful Oral Sirolimus to Inhibit Recurrent In-stent Stenosis (OSIRIS) trial and that the duration of administration was tailored to the early pro-inflammatory phase after an acute myocardial infarction aiming at avoiding any interference with beneficial repair processes during the healing phase,(32) the characteristics of the treatment regimen appeared adequate. However, we cannot rule out that regimens including peri-procedural drug application with intravenous formulations of mTOR inhibitors or longer treatment durations could have favourably affected the results. In a rat model of myocardial infarction, mTOR inhibition attenuated myocardial remodelling and reduced myocyte size both when initiated on the day after or 3 days after the induction of myocardial infarction, while a beneficial effect on myocardial infarct size was only observed in the early treatment group.(17) To which extent these findings observed in an animal model of myocardial infarction can be translated to STEMI patients undergoing primary PCI remains uncertain. It further needs to be considered

that anti-inflammatory therapies administered during the first days after an acute myocardial infarction may negatively affect the transition of the proinflammatory phase to the proreparative phase of infarct healing. While the comparable improvements of left ventricular systolic function and volumes in the everolimus and placebo groups render a potential harmful effect of mTOR inhibition on infarct healing under the conditions of the present study less likely, the CLEVER-ACS trial was not powered to determine if there is any adverse safety signal from this drug class in patients with acute myocardial infarction. The rather slow patient enrollment in CLEVER-ACS may have introduced a certain bias, particularly given the impressive advancements of treatment strategies for STEMI patients achieved over the last years and the enrollment of early patients which did not receive contemporary evidence-based treatment. Strict inclusion and exclusion criteria, along with logistic challenges related to CMR imaging availabilities, may have prolongated patient enrollment. In addition, enrollment into CLEVER-ACS, testing an immunosupressive agent, was slowed down and temporarily halted during the SARS-CoV-2 pandemic.

Myocardial infarct size, microvascular obstruction, and left ventricular remodeling in acute myocardial infarction.

Myocardial infarct size and the amount of MVO observed in CLEVER-ACS are in line with previous studies.(25-27) Differences in study designs and patient characteristics may account for some variability among studies, and the inclusion of patients with mostly large STEMI due to proximal vessel occlusion in CLEVER-ACS may explain at least in part the larger than expected myocardial infarct size. In contrast to most CMR studies in acute myocardial infarction patients, CLEVER-ACS comprises both baseline and follow-up CMR analyses, allowing for direct comparisons of CMR parameters over time. In both treatment groups, myocardial infarct size and MVO decreased and LVEF improved from baseline to 30 days. The magnitude of the decrease in myocardial infarct size observed in CLEVER-ACS from baseline to 30 days is in line with previous reports.(25, 33, 34) The increasing left

ventricular volumes may reflect maladaptive remodeling processes following large acute myocardial infarctions. Of note, a considerable number of patients had a left ventricular thrombus detected by CMR in both treatment arms, underscoring the value of CMR in the clinical management of post-STEMI patients.(35)

Safety of everolimus.

Everolimus and other mTOR inhibitors are widely used drugs with acceptable safety profiles.(32, 36) The short-term treatment of everolimus in CLEVER-ACS was generally well tolerated. Everolimus did not result in an increase in infections, as might have been expected given its anti-inflammatory properties. Exanthema, oral ulcers, diarrhea, and renal failure represent well-known, mostly mild side effects of everolimus and were more frequently reported in the everolimus group without reaching significance. Whether the numerically higher occurrence of left ventricular thrombi in the everolimus group is related to mTOR inhibition remains to be determined.(35, 37) Further, given the increased rates of left ventricular thrombi observed with colchicine treatment in the Colchicine for Left Ventricular Infarct Size Treatment in Acute Myocardial Infarction (COVERT-MI) trial,(38) a pro-inflammatory rebound at discontinuation of anti-inflammatory therapies, fostering subsequent thrombus formation in acute STEMI patients, is unlikely, but possible.(39, 40)

Study limitations.

Some limitations need to be considered. First, the particular study design which allowed for patient screening within 5 days following PCI needs to be taken into account when interpreting the results, along with the slow patient enrollment. Second, the relatively low percentage of women enrolled in the study needs to be considered. Third, as follow-up CMR was performed 30 days after the index STEMI event, long-term effects of mTOR-inhibition on myocardial infarct size and LV remodeling beyond 30 days could not be assessed. Fourth, while CLEVER-ACS was adequately powered to assess a potential impact

on mTOR inhibition on infarct size and LV remodeling, the sample size did not allow for the comparison of clinical event rates and safety among the resp. study groups.

CONCLUSION

Early mTOR inhibition using everolimus did not reduce myocardial infarct size or microvascular obstruction at 30 days among patients with STEMI undergoing reperfusion therapy with primary PCI.

PERSPECTIVES

Clinical competencies. Among STEMI patients undergoing PCI, early mTOR inhibition with everolimus did not reduce myocardial infarction size or microvascular obstruction at 30 days.

Translational outlook. Future trials are needed to determine potential effects of mTOR inhibition on clinical outcomes in patients with acute STEMI.

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FIGURE LEGENDS

Figure 1. Study flow chart. Flow chart illustrating randomization and follow-up. CMR = cardiac magnetic resonance imaging.

Figure 2. The primary endpoint in the intention-to-treat population. There were 69 patients randomized in the everolimus group and 66 patients randomized in the placebo group (n = 135). The primary endpoint was the change in myocardial infarct size from baseline to 30 days as assessed by cardiac magnetic resonance imaging.

Central Illustration. Design and Results of the CLEVER-ACS trial. A total of 150 patients with acute ST-segment elevation myocardial infarction undergoing primary percutaneous coronary intervention were enrolled in the study and randomized to either everolimus or placebo. The primary endpoint was the change in myocardial infarct size from baseline (12 hours – 5 days after PCI) to 30 days as assessed by cardiac magnetic resonance imaging. The changes in myocardial infarct size from baseline to 30 days were -14.2 (95% CI -17.4 to -11.1) g and -12.3 (-16.0 to -8.7) g in the everolimus and placebo groups. CMR = cardiac magnetic resonance imaging, PCI = percutaneous coronary intervention, STEMI = ST-segment elevation myocardial infarction.

Table 1. Baseline Characteristics in the Everolimus and Placebo Groups.

Variables	Everolimus (n=75)	Placebo (n=75)
Baseline characteristics		
Age, years	61.6±10.7	62.1±11.7
Women	12 (16)	10 (13)
Family history of coronary artery disease	21 (28)	15 (20)
Diabetes mellitus	8 (11)	6 (8)
Hypertension	32 (43)	29 (39)
Dyslipidemia	39 (52)	30 (40)
Peripheral arterial disease	2 (3)	3 (4)
Previous stroke/TIA	1 (1)	1 (1)
Renal failure requiring dialysis	0 (0)	0 (0)
Malignancy	4 (5)	0 (0)
Chronic obstructive pulmonary disease	2 (3)	2 (3)
History of gastrointestinal bleeding	1 (1)	0 (0)
Systemic inflammatory disease	1 (1)	0 (0)
Liver cirrhosis/chronic hepatitis	2 (3)	0 (0)
Clinically relevant valvular heart disease	0 (0)	1 (1)
Baseline medication		
Aspirin	67 (89)	72 (96)
Clopidogrel	8 (11)	9 (12)

Ticagrelor	40 (53)	37 (49)
Prasugrel	21 (28)	22 (29)
Oral anticoagulation	8 (11)	3 (4)
ACEI	51 (68)	52 (69)
AT II antagonists	7 (9)	7 (9)
Beta blockers	52 (69)	56 (75)
Calcium channel blockers	7 (9)	2 (3)
Diuretics	15 (20)	16 (21)
Statin	63 (84)	67 (89)

Values are given as mean and standard deviation or numbers and percentages. Baseline characteristics are given at the time point of randomization. ACEI = angiotensin converting enzyme inhibitors, AT = angiotensin, TIA = transient ischaemic attack.

Table 2. Procedural Characteristics in the Everolimus and Placebo Groups.

Variable	Everolimus (n=75)	Placebo (n=75)
Culprit artery		
Left main coronary artery	0 (0)	1 (1)
LAD	48 (64)	39 (52)
CX	9 (12)	6 (8)
RCA	18 (24)	29 (39)
Bypass vessel	0 (0)	0 (0)
Thrombus grade		
Grade 1	2 (4)	5 (9)
Grade 2	5 (9)	9 (16)
Grade 3	48 (87)	42 (75)
Thrombus aspiration	18 (33)	32 (57)
TIMI flow pre-PCI		
0 or 1	73 (97)	70 (93)
2	1 (1)	3 (4)
3	1 (1)	2 (3)
TIMI flow post-PCI		
2	2 (3)	6 (8)
3	73 (97)	69 (92)

Values are given as numbers and percentages. CX = circumflex coronary artery, LAD = left anterior descending coronary artery, PCI = percutaneous coronary intervention, RCA = right coronary artery, TIMI = Thrombolysis in Myocardial Infarction.

Table 3. Change in Biomarker Levels from Baseline to 30 Days in the Everolimus and Placebo Groups.

	Everolimus				Placebo					
Variable	**Baseline**	**Follow-up**	**Change**	**p-value***	**Baseline**	**Follow-up**	**Change**	**p-value***	**Difference****	**p-value*****
CK, U/l	994 (444-2190) [73]	100 (77-137) [64]	-1417 (-1822 to -1013)	<0.001	925 (374-1739) [68]	105 (71-182) [64]	-1037 (-1345 to -728)	<0.001	21 (-102 - 145)	0.74
CK-MB, µg/l	60 (40-140) [35]	11 (4-15) [31]	-106 (-154 to -58)	<0.001	64 (28-196) [33]	9 (3-14) [32]	-78 (-110 to -46)	<0.001	0.2 (-3.7 - 4.2)	0.91
Hs-cTn T, µg/l	4.54 (2.29-9.76) [62]	0.05 (0.01-17.00) [63]	-663 (-1117 to -209)	0.001	4.79 (1.77-12.10) [56]	0.03 (0.01-13.00) [59]	-763 (-1423 to -103)	0.04	1.9 (-8.4 – 12.1)	0.91
Hemoglobin, g/l	139±14 [74]	140±12 [66]	0.21 (-2.68 to 3.11)	0.88	135±16 [71]	140±15 [65]	5.45 (2.62 to 8.29)	<0.001	-3.3 (-6.8 - 0.2)	0.06
Leucocytes, G/l	11.3±3.8 [74]	7.7±2.2 [66]	-3.73 (-4.63 to -2.82)	<0.001	10.2±3.8 [71]	7.6±2.2 [66]	-2.45 (-3.15 to -1.76)	<0.001	-0.3 (-1.0 - 0.4)	0.41
Creatinine, µmol/l	83±20 [73]	89±25 [67]	4.82 (0.73 to 8.91)	0.02	83±22 [69]	88±21 [66]	4.43 (1.59 to 7.27)	0.003	0.5 (-4.4 - 5.3)	0.86

Values are given as mean and standard deviation or median and interquartile range, and difference with 95% CI for the changes between baseline and follow-up and for the difference between the treatment groups. CK = creatine kinase, CK-MB = creatine kinase myocardial band, Hs-Tn = high-sensitivity cardiac troponin, *for comparisons between baseline and follow-up at 30 days, **adjusted mean difference of the changes in the everolimus and placebo groups, ***for between-group comparisons of the change from baseline to follow-up at 30 days.

Table 4. Change in Cardiac Magnetic Resonance (CMR) Endpoints from Baseline to 30 Days in the Everolimus and Placebo Groups.

	Everolimus				Placebo					
Variable	**Baseline**	**Follow-up**	**Change**	**p-value***	**Baseline**	**Follow-up**	**Change**	**p-value***	**Difference****	**p-value*****
MI size, g	42.2±27.8 [72]	27.7±21.5 [69]	-14.2 (-17.4 to 11.0) [69]	<0.001	36.2±26.0 [70]	24.0±19.9 [66]	-12.3 (-16.0 to -8.7) [66]	<0.001	0.02 (-3.47 to 3.51)	0.99
MI size, % of LV mass	31.0±19.7 [71]	22.7±16.2 [69]	-8.7 (-11.4 to 5.9) [69]	<0.001	27.2±16.9 [70]	21.0±16.3 [65]	-6.1 (-9.1 to -3.0) [65]	<0.001	-1.34 (-4.76 to 2.08)	0.44
Myocardial salvage index, % of AAR volume	17.7±21.7 [44]	7.5±15.2 [35]	-11.8 (-19.6 to -3.9) [30]	0.005	18.9±20.8 [46]	9.7±17.2 [41]	-9.5 (-18.6 to -0.4) [38]	0.04	-3.78 (-11.19 to 3.62)	0.32
MVO, g	6.2±7.7 [69]	0.8±2.0 [56]	-4.8 (-6.7 to -2.9) [56]	<0.001	7.0±9.4 [68]	0.5±1.2 [59]	-6.3 (-8.7 to 4-0) [59]	<0.001	0.41 (-0.13 to 0.95)	0.14
MVO, % of LV mass	4.7±6.1 [68]	0.7±1.5 [56]	-3.4 (-4.7 to -2.0) [55]	<0.001	5.0±6.2 [68]	0.5±1.1 [58]	-4.4 (-5.9 to -2.8) [58]	<0.001	0.28 (-0.17 to 0.72)	0.22
AAR, g	40.4±27.7 [50]	17.9±23.3 [43]	-23.4 (-32.9 to -14.0) [41]	<0.001	43.4±32.8 [49]	16.4±21.2 [47]	-27.0 (-35.6 to -18.4) [44]	<0.001	0.71 (-7.85 to 9.26)	0.87
LV hemorrhage, % of LV mass	1.5±2.5 [39]	0.6±1.8 [32]	-1.2 (-2.3 to -0.2) [30]	0.03	2.3±5.0 [40]	0.4±1.2 [39]	-2.1 (-3.7 to -0.5) [36]	0.01	0.01 (-0.55 to 0.56)	0.98
LVEF, %	45.6±9.0 [70]	48.4±10.4 [70]	2.7 (0.7 to 4.7) [68]	0.009	46.3±9.8 [70]	50.7±10.9 [66]	4.3 (2.4 to 6.2) [66]	<0.001	-1.75 (-4.32 to 0.82)	0.18
LVEDV, ml	166±37 [71]	174±47 [70]	6.9 (0.7 to 13.1) [69]	0.03	163±37 [70]	177±47 [66]	11.8 (4.8 to 18.7) [66]	0.001	-5.03 (-13.83 to 3.78)	0.26
LVEDVI, ml/m2	84±15 [70]	88±20 [69]	3.4 (0.2 to 6.5) [68]	0.04	82±18 [69]	89±24 [65]	6.6 (2.7 to 10.5) [65]	0.001	-3.31 (-8.04 to 1.41)	0.17
LVSV, ml	75±18 [71]	83±24 [70]	7.4 (2.7 to 12.0) [69]	0.002	74±17 [70]	86±18 [66]	11.0 (7.2 to 14.9) [66]	<0.001	-3.44 (-8.96 to 2.07)	0.22
LVSVI, ml/m2	37.7±7.7 [70]	41.8±9.4 [69]	4.0 (1.8 to 6.3) [68]	0.001	36.7±8.4 [69]	43.3±8.3 [65]	6.2 (4.0 to 8.3) [65]	<0.001	-1.86 (-4.56 to 0.84)	0.18
LV mass, g	131±30 [71]	118±25 [70]	-12.4 (-16.0 to -8.7)) [69]	<0.001	126±28 [70]	113±23 [66]	-14.5 (-19.1 to -9.9) [66]	<0.001	3.08 (-1.66 to 7.83)	0.20
LVMI, g/m2	66±13 [70]	60±11 [69]	-6.4 (-8.2 to -4.5) [68]	<0.001	64±14 [69]	57±11 [65]	-6.9 (-9.5 to -4.3) [65]	<0.001	1.32 (-1.24 to 3.88)	0.31

Values are given as mean and standard deviation and difference with 95% CI for the changes between baseline and follow-up and for the difference between the treatment groups. AAR = area at risk, LVEDV = left ventricular end-diastolic volume, LVEDVI = left ventricular end-diastolic volume index, LVEF = left ventricular ejection fraction, LVMI = left ventricular mass index, LVSVI = left ventricular stroke volume index, MI = myocardial infarction, MVO = microvascular obstruction. *for comparisons between baseline and follow-up at 30 days, **adjusted mean difference of the changes in the everolimus and placebo groups, ***for between-group comparisons of the change from baseline to follow-up at 30 days.

Table 5. Safety Endpoints at 30 Days in the Everolimus and Placebo Groups.

Variables	Everolimus (n=75)	Placebo (n=75)	p-value
No. of AE	59	47	0.37
Patients with at least 1 AE	34 (45)	29 (39)	0.44
No of SAE	19	12	0.23
Patients with at least 1 SAE	16 (21)	11 (15)	0.20
Patients with SAE leading to study drug interruption or withdrawal	0 (0)	0 (0)	-
All-cause death	0 (0)	0 (0)	-
Non-fatal myocardial infarction	0 (0)	1(1)	1.0
Renal failure	3 (4)	0 (0)	0.25
Bleeding	4 (5)	3 (4)	1.0
LV thrombus	11 (15)	6 (8)	0.30
Infection	4 (5)	4 (5)	1.0
Diarrhea	2 (3)	2 (3)	1.0
Oral ulcer	5 (7)	1 (1)	0.21
Exanthema	4 (5)	0 (0)	0.12
Atrial fibrillation	1 (1)	2 (3)	1.0
Others	27 (36)	26 (35)	1.0

Values are given as numbers and percentages. AE = adverse event, SAE = serious adverse event, LV = left ventricular.

Figure 1.

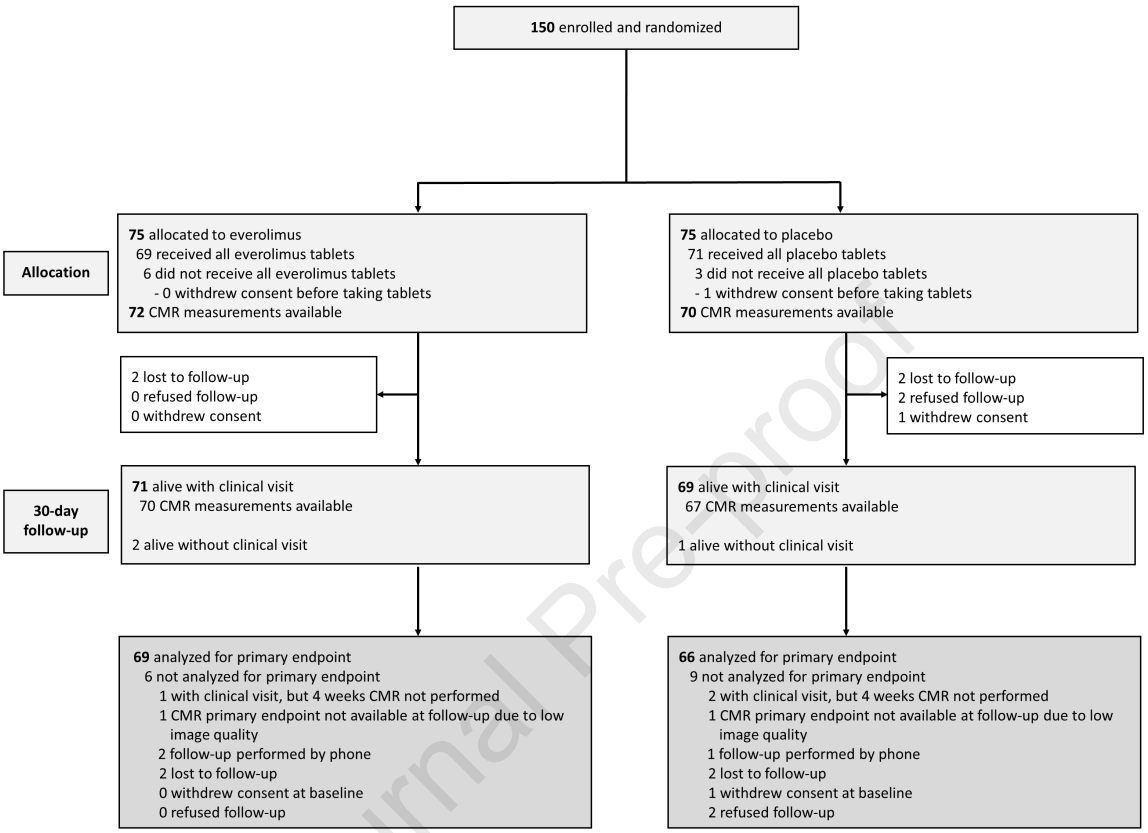
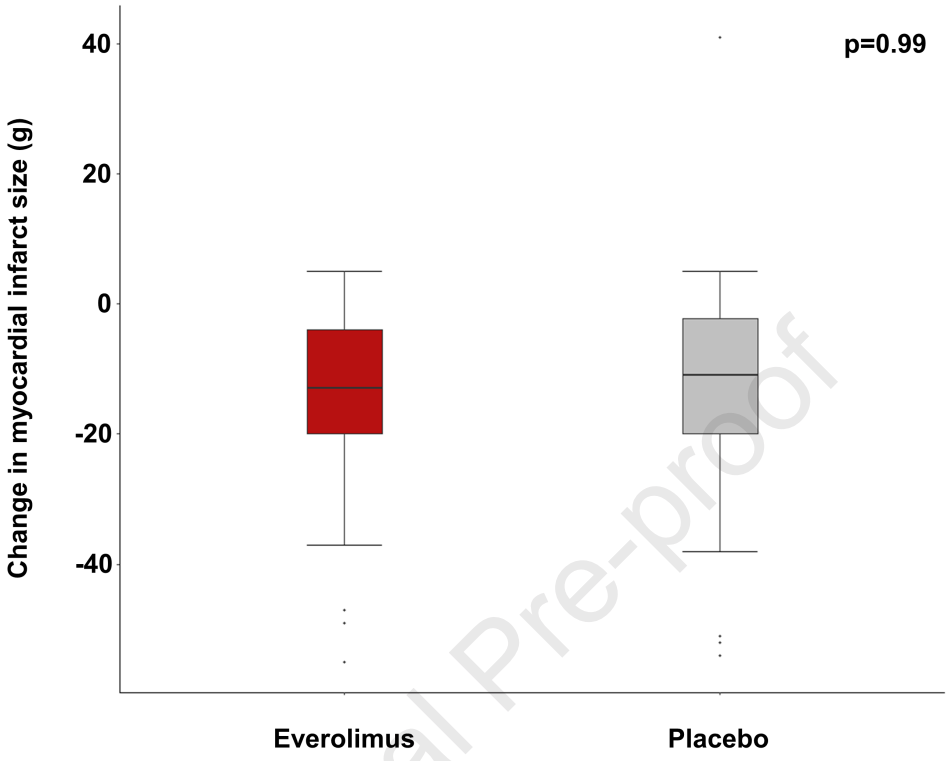


Figure 2.



Supplementary Table 1. Subgroup Analyses in the Everolimus and Placebo Groups.

	Everolimus				Placebo					
Variable	**Baseline**	**Follow-up**	**Change**	**p-value***	**Baseline**	**Follow-up**	**Change**	**p-value***	**Difference****	**p-value*****
Subgroup analysis scar mass										
Baseline scar mass ≤median (38.5 g)	17.3±11.5 [33]	11.4±10.8 [32]	-5.3 (-7.0 to -3.6)	<0.001	17.7±10.1 [38]	12.2±11.1 [36]	-5.1 (-8.5 to -1.8)	0.004	-0.33 (-3.90 - 3.24)	0.86
Baseline scar mass >median (38.5 g)	63.2±18.5 [39]	41.7±18.4 [37]	-22.0 (-26.4 to -17.5)	<0.001	58.3±21.4 [32]	38.1±19.0 [30]	21.0 (-26.4 to -15.5)	<0.001	0.48 (-5.55 - 6.50)	0.88
Subgroup analysis time from PCI to first study drug administration										
≤12 hours	33.8±27.6 [8]	23.0±20.9 [8]	-10.8 (-16.3 to -5.3)	0.006	39.1±26.9 [13]	28.8±23.0 [10]	-13.9 (-24.8 to -3.0)	0.03	-0.19 (-15.02 - 14.64)	0.98
>12 hours	44.9±28.1 [57]	29.3±22.0 [55]	-15.4 (-19.3 to -11.6)	<0.001	35.5±26.4 [47]	30.6±26.6 [46]	-12.5 (-17.0 to -8.0)	<0.001	5.68 (-1.78 - 13.14)	0.14

Values are given as mean and standard deviation and difference with 95% CI for the changes between baseline and follow-up and for the difference between the treatment groups. The time of PCI was defined as time of first balloon inflation. PCI = percutaneous coronary intervention. *for comparisons between baseline and follow-up at 30 days, **adjusted mean difference of the changes in the everolimus and placebo groups, ***for between-group comparisons of the change from baseline to follow-up at 30 days.