The role of early high-dose rosuvastatin loading in patients presenting with acute coronary syndromes without persistent ST-segment elevation

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THE ROLE OF EARLY HIGH-DOSE ROSUVASTATIN LOADING IN PATIENTS PRESENTING WITH ACUTE CORONARY SYNDROMES WITHOUT PERSISTENT ST-SEGMENT ELEVATION

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LIST OF ABBREVIATIONS

ACC - American College of Cardiology

ACS – Acute coronary syndrome

AHA - American Heart Association

AMI – Acute myocardial infarction

ASA – Acetylsalicylic acid

CABG – Coronary artery bypass grafting

CAD – Coronary artery disease

DAPT – Dual antiplatelet treatment

DM – Diabetes mellitus

ESC – European Society of Cardiology

GPIIb/IIIa – Glycoprotein GPIIb/IIIa

LVEF – Left ventricular ejection fraction

MACCE – Major adverse cerebrovascular and cardiovascular events

NSTE-ACS – Non-ST-elevation acute coronary syndromes

NSTEMI - Non-ST-elevation myocardial infarction

PCI – Percutaneous coronary intervention

RCT – Randomized controlled trial

STEMI – ST-elevation myocardial infarction

UA – Unstable angina

1. INTRODUCTION

1.1. History of statins

Atherosclerosis was documented in the 19th century, but its pathological potential and mechanisms behind its development were not understood. Some of the first clues that pointed towards cholesterol and its association with atherosclerosis emerged in 1910 (1). In the second half of the 20th century, there was a light thrown upon the correlation between cholesterol and its role in the causality of cardiovascular disease. Several studies were performed in the 1950s which led to the emergence of *"Lipid Hypothesis"*. This hypothesis suggested that the correlation between elevated low-density lipoprotein (LDL) cholesterol were in fact causally related to coronary artery disease (CAD) and thus lowering it would reduce the events of myocardial infarction (MI) and other coronary events. Treatment consisted mainly of lowering cholesterol through several different dietary measures. Akira Endo, a Japanese microbiologist discovered in the 1970s the first products that had an inhibitory effect on 3-hydroxy-3-methyl-glutaryl-coenzyme A (HMG-CoA) reductase, including compactin. This product which was extracted from *Penicillium citrium* was shown to reduce plasma cholesterol levels in several animal models. Nevertheless, this discovery was brought to a halt when serious animal toxicity was observed. Lovastatin, the very first US Food and Drug Administration (FDA)-approved statin was shown to reduce LDL cholesterol in humans and was discovered in 1978 by Alberts, Chen, and others thus gaining approval in 1986. Lovastatin managed to sucessfully reduce plasma cholesterol in clinical practice and soon became a standard treatment modality for hypercholesterol (2). Shortly after the discovery and approval of lovastatin, the pharmaceutical development of simvastatin and other forms of statins ensued.

1.2. Statin pharmacodynamics and pharmacokinetics

The main mechanism of action of statins is characterized by the competitive binding and inhibition of the rate-limiting enzyme HMG-CoA reductase in hepatocytes, with a subsequent decrease of intrinsic biosynthesis of total cholesterol and lowering of LDL cholesterol in peripheral circulation. An increased expression of LDL receptors appears to be a result of decreased cholesterol synthesis (3). Furthermore, statins can be divided into lipophilic and hydrophilic groups, which will impact their pharmacokinetic properties. Lipophilic statins include lovastatin, simvastatin, fluvastatin, and atorvastatin. Rosuvastatin and pravastatin are included in the group of hydrophilic statins. There is a variable effect of absorption of statins in relation to food intake. Lovastatin has improved bioavailability when taken together with

food, whereas for atorvastatin, fluvastatin, and pravastatin the effect is decreased. Of all statins, the only ones where bioavailability is not affected by the timing of meals, are rosuvastatin and simvastatin, and thus not affecting the rate of cholesterol-lowering effect (4).

Following oral intake, the absorption of all statins is rapid, showing peak plasma concentrations within 4 hours (5).

Before entering the systemic circulation and eventually reaching the liver, statins need to pass through the barriers of the intestinal tract. Passage through hepatocytes occurs through active transport, or passively via the ATP-binding transport system. Metabolism of statins occurs mainly in the liver and to some degree in the kidney. Entry into these organs depends on the liposolubility of the statin and is mediated through two families of enzymes including the cytochrome P450 (CYP), and UDP-glucuronosyltransferase (UGT). Lipophilic statins use passive transport to enter the liver and are metabolized by CYP enzymes, while rosuvastatin and pravastatin which are hydrophilic enter the liver through active transport. Hydrophilic statins are metabolized less than lipophilic statins by the CYP enzymes (4).

Most of the endogenous cholesterol production occurs in the liver. This is a key fact and is important when we consider the relative hepatoselectivity of statins, and its effect in inhibiting HMG-CoA reductase, which is an important part of endogenous cholesterol production (5, 6).

As previously elaborated, statins exhibit variable degrees of liposolubility, and thus they differ in selectivity in regards to their uptake in different tissues, including the liver. Lipophilic statins enter hepatocytes by passive diffusion, but also in other nonhepatic cells. On the other hand, hydrophilic statins are significantly more selective for liver entry. The effect of pravastatin on smooth muscle cell proliferation is absent and is most probably due to the low penetrance of cells by this hydrophobic statin.

The active metabolites of lovastatin, simvastatin, and atorvastatin also contribute to the inhibitory effect on HMG-CoA reductase. These active metabolites mostly include 2-hydroxy- and 4-hydroxy-atorvastatin acid for atorvastatin, and for pravastatin, the metabolites include β-hydroxy acid and its 6-hydroxy, 6-hydroxymethyl, and 6-exomethylene derivatives (5).

The mode of excretion of statins is mainly through the biliary system (fecal excretion) after metabolism in the liver. Of note, hydrophilic statins such as rosuvastatin and pravastatin are partially excreted through kidneys (10% for rosuvastatin and 20% for pravastatin). Therefore, impaired liver function is a risk factor for statin-induced myopathy (4).

Pharmacological characteristics of statins that are used in contemporary clinical practice are summarized in **Table 1**.

Table 1. Pharmacological characteristics of statin types used in clinical practice

	Rosuvastatin	Pravastatin	Lovastatin	Fluvastatin	Pitavastatin	Simvastatin	Atorvastatin
Therapeutic dose (mg/daily)	5-40	2-80	10-80	20-80	1-4	5-40	10-80
Bioavailability (%)	20	17	<5	6	>60	<5	12
Active metabolites	Yes (minor)	No	Yes	No	No	Yes	Yes
% protein binding	89	50	>95	98	96	95	≥90
% fecal excretion	90	71	83	90	75	58	90
% renal excretion	10	20	10	<6	2	13	<2
Half-life (hours)	19	1-2	2	4.7	12	1-2	14
Liver metabolism	*CYP450 2C9, 2C19*	*Sulphation*	*CYP450 3A4*	*CYP450 2C9*	*CYP450 2C9*	*CYP450 3A4*	*CYP450 3A4*
Solubility	Hydrophilic	Hydrophilic	Lipophilic	Lipophilic	Lipophilic	Lipophilic	Lipophilic

1.3. Statin effects on lipids

As already mentioned, main purpose of statin use is to lower abnormal cholesterol and lipid levels, and ultimately lower the risk of cardiovascular disease, by the means of inhibit the HMG-CoA reductase, and also in part from its pleiotropic effects. HMG-CoA reductase catalyzes the conversion of HMG-CoA to mevalonate, an early rate-limiting step in biosynthesis of cholesterol. Statins reversely binds this enzyme, resulting in lowering hepatic cholesterol concentration. Inhibition of HMG-CoA reductase leads to an increase of LDL receptor expression on the surface of hepatocytes as a compensatory mechanism for the reduced intrinsic cholesterol production. This results in an enhanced uptake of LDL cholesterol from the peripheral circulation, thus, further lowering the circulating LDL cholesterol (3).

Additional effects of statin drugs include lowering triglycerides in patients with combined (mixed) dyslipidemia by reducing the production of apolipoprotein (apo) B100, thus providing an effective reduction in circulating levels of both LDL cholesterol and triglycerides (7, 8). By not fully elucidated mechanisms, statins also increase circulating levels of high-density lipoproteins (HDL) to varying degrees. It is suggested that this effect might be due to phosphorylation of peroxisomal proliferating activator receptor-α (PPAR- α) (9).

1.4. Pleiotropic effects of statins beyond lipid-lowering mechanisms

Other than the dominant lipid-lowering effects of statins, some additional properties have been observed and are termed as “pleiotropic” statin effects. These include augmentation of endothelial nitric oxide (eNOS), causing vasodilation, and improving endothelial function (10). Statins may also lead to a better outcome after percutaneous transluminal coronary angioplasty (PTCA), possibly due to inhibition of platelet function and attenuation of plaque vulnerability, but also by inhibiting myocyte infiltration and reduced secretion of metalloproteinases (3). Upregulation of eNOS leads to a potential reduction in the production of thromboxane A2 and altering cell membrane constituents of cholesterol. Reduction of cell membrane cholesterol composition reduces the chance for the thrombotic formation of these cells (11).

Statins act anti-inflammatory and were associated with beneficial effects in the setting of atrial fibrillation and outcomes after cardiac surgery which portends a lot of inflammatory stimuli. For example, in the primary prevention setting such as one in the JUPITER (Rosuvastatin to Prevent Vascular Events in Men and Women with Elevated C-Reactive

Protein) trial, rosuvastatin managed to reduce C-reactive protein (CRP) by 37% (12). In the acute setting, such as in MIRACL (Myocardial Ischemia Reduction With Aggressive Cholesterol Lowering) trial, atorvastatin lowered CRP by 83% (13, 14). Moreover, statins decrease levels of high-sensitivity CRP (hs-CRP) which is a clinical marker for systemic inflammation. Elevated hs-CRP levels are associated with an elevated risk of coronary artery disease (CAD) in otherwise healthy people. Hs-CRP binds to LDL cholesterol inside atherosclerotic plaques, activates complement cascade thus stimulating the progression of the atherosclerotic plaque. There has been shown a correlation between statin treatment and a decreased amount of hs-CRP in patients with hypercholesterolemia (11).

In addition to the endothelial protective effects, enhancing blood flow and contributing to plaque stability, statins also exert inhibition of vascular smooth muscle cells (VSMCs). Statins inhibit proliferation of VSMCs and induce apoptosis in this cell type (15). The mechanism behind this inhibition is thought to be partially from inhibiting isoprenoid, thus decreasing platelet-derived growth factor (PDGF)-induced DNA synthesis (16). It is indicated that the inhibition of smooth vascular muscle cells is due to cell cycle arrest between the G1 and S phase transition of the cell cycle (17, 18).

Another surprising property of statin is its use in local injections as a treatment of bone resorption after periodontitis. Periodontal inflammation and bone loss were significantly reduced in patients getting high dose atorvastatin injection (19).

1.5. Intravenous statins – future pathway to mitigate acute ischemic events?

Until now, periprocedural administration of high-dose statins concerning reperfusion intervention has been done orally. The question to be asked now is, whether intravenous administration of statins would be superior to oral administration. A recent study was conducted to investigate intravenous atorvastatin administration during experimentally-induced AMI, and to compare it with oral administration shortly after MI. This study involved animals that were divided into 3 arms; arm 1 received an intravenous bolus of atorvastatin during MI; arm 2 received a bolus of the intravenous vehicle during MI, and arm 3 got oral atorvastatin within 2 hours after MI. The results favoured intravenous atorvastatin administration in several endpoints, such as; a reduction of myocardial damage, enhanced cardiac function, as well as limitation of scar formation in comparison to the other groups. The lower extent of apoptosis and myocardial damage was demonstrated to a higher degree compared with those receiving

oral statin administration. Also, intravenous administration revealed a noteworthy higher myocardial salvage index compared to the other groups. Interestingly, the placebo group had a similar myocardial salvage index as the group receiving oral atorvastatin. Intravenous administration of atorvastatin after AMI was shown to reduce the adverse left ventricular remodeling to a higher extent as well (20). Mechanistic explanations were provided through observation that AMPK signaling pathway activation in cardiomyocytes mediated cardioprotective effects of intravenous atorvastatin during ischemia (21). Parenteral systems for statin delivery might be way in the future since oral statin administration in acute setting has downsides such as hepatic first-pass metabolism and degradation within the gastrointestinal tract thus limiting their bioavailability and in some patients with ACS that present with nausea and vomiting oral statin administration might not be feasible and effective (22).

1.6. Adverse effects of statins

The best documented and the most important adverse side effects are seen in observational studies and clinical trials. These include myopathy and slightly increased risk of new-onset diabetes. Beside myopathy and diabetes risk, there are other possible adverse effects like an increased risk of hemorrhagic stroke, impaired cognition and memory, cataract and effects on the kidneys. Although there is substantial and reliable evidence of an increased risk of hemorrhagic stroke, other risks have not been robustly documented.

Myopathy caused by statins is seen to be either a direct myotoxic effect that is dose-dependent and reversible or associated with an autoimmune reaction targeting HMG-CoA reductase, which is not dose-dependent nor is a resolution seen after discontinuation of the drug (23). Musculoskeletal side effects can be linked to most statins. Manifesting most commonly as myalgia, and less commonly as myositis which is linked with an increase of serum creatine kinase (CK). The most severe form of musculoskeletal side effect is rhabdomyolysis, exhibiting a CK rise more than 10x the upper limit of normal values, and the following associated myoglobinuria, renal impairment, and thus serum electrolyte imbalance. Generally, the incidence of the most severe musculoskeletal side effects is low (24).

Mechanisms underpinning statin-induced myotoxicity include hypothesis about muscle damage due to decreased *ubiquinone*, a protein important in stabilizing the cell membrane and which also is included in the mitochondrial respiratory chain; leading to elevated levels of sterols that may increase the damaging effects of statins in the myocytes or it may be explained

by *artrogen-1* overexpression, an important gene implicated in skeletal muscle atrophy (23). The change of cholesterol content in the cells alters the function of ion channels including calcium, resulting in damage and cell death. Direct drug-related toxic effects are rarely seen and when they occur they are almost exclusively related to high statin doses and more intense lipid-lowering therapies. It would be intuitive to expect that more potent statins would induce more toxic effects than less potent statins, but this has not been readily demonstrated. Myotoxicity increases substantially when combing other drugs which interact with statins, more specifically, inhibitors of cytochrome P450 (24).

Extremely rarely, patients on statin treatment may develop myopathy due to the development of an autoimmune reaction and this occurs in about 2-3 per 100,000 patients treated per year (25). This condition is also known as statin-induced necrotizing autoimmune myopathy (SINAM) and is characterized by proximal muscle weakness, necrosis detected on muscle biopsy, markedly elevated CK and the presence of autoantibodies against HMG-CoA reductase. As previously mentioned, this damage is not reversible and may require immunosuppressive therapy. In conclusion, clinical phenotypes of statin-induced myalgia and myositis can be summarized as rhabdomyolysis, myalgia and/or mild hyperCKemia, self-limited toxin statin myopathy, or immune-mediated necrotizing myopathy (26).

Statin treatment is associated with an increased risk and development of type 2 diabetes mellitus (T2DM), by affecting pancreatic beta cells and by increasing insulin resistance. The estimation of the overall increased risk is about 10-20 per 10,000 patients per year and it has been shown that treatment with atorvastatin and simvastatin was associated with a 14% increased risk of T2DM (27). The increased risk is especially linked to patients that have other risk factors, including elevated body-mass index (BMI), impaired fasting glucose, or high glycated hemoglobin (HbA1c) (23). As for myopathy, the risk is elevated with a higher dose and intensity of statin therapy, but in contrast to myopathy, the use of more potent statin drugs increases the risk of diabetes as well (23, 24). The exact pathogenic mechanism is not fully elucidated. Nevertheless, it has been suggested that it might be related to lowering of LDL cholesterol, or increased expression of LDL receptors on the pancreas, and thus damage of pancreatic tissue by entering of excess cholesterol (23). With regards to the lowering of cardiovascular risk with statin therapy, diabetic patients are the patient group that reap the highest therapeutic benefit. There is also no evidence that the use of statins worsens diabetic dysregulation. Cardiovascular protective benefits of statins used in patients with diabetes mellitus outweigh the adverse effects concerning diabetes (24).

An association between statin use and hemorrhagic stroke has been noted in observational studies. This is especially seen in patients with high blood pressure. Evidence shows that there is an estimated risk increase of 21 % and is mostly linked with previous cerebrovascular disease, and certain populations, like Asian people (23). Meta-analysis and randomized trials, have shown that although the risk of ischemic stroke was reduced, the development of hemorrhagic stroke was increased by using statins (28). Similarly, in patients with a history of cerebrovascular disease, statins significantly decreased the risk of ischemic stroke, however, that was partially offset by an increased risk of hemorrhagic stroke (29). On the other hand, a recent systematic review and meta-analysis showed that statin treatment was not associated with cerebral microbleeds (CMBs) overall, however, it might be associated with an increase in the risk of lobar CMB formation (30).

Finally, the most recent network and dose-response meta-analysis encompassing 62 primary prevention trials showed that the risk of adverse events attributable to statins was low and did not outweigh their advantages for the primary prevention of cardiovascular disease thus finally concluding that the benefit-to-harm balance of statins in this setting is generally favourable (31).

1.7. PCSK9 inhibitors in managing dyslipidemia

Some novel classes of drugs need to be briefly mention due to their efficacy in managing LDL cholesterol. Of note, Proprotein Convertase Subtilisin/Kexin Type 9 (PCSK9)-inhibitors are a group of drugs that potently reduce serum cholesterol concentration and are used in the primary treatment of hypercholesterolemia, including both familial and non-familial hypercholesterolemia (32). Included in this drug group are evolocumab and alirocumab. These drugs are monoclonal antibodies, and they exert their mechanism of action by binding to endogenous PCSK9 enzyme with high affinity thus preventing its natural binding to LDL receptors. When PCSK9 has been pharmacologically blocked this results in less LDL receptor degradation on the surface of hepatocytes thus enabling more uptake of LDL cholesterol and clearance of LDL from peripheral circulation (33, 34).

PCSK9-inhibitors are used in combination with statins and are seen to reduce circulating LDL cholesterol concentrations by about 50% to 60%, on top of maximized statin background treatment, thereby significantly halving the occurrence of adverse cardiovascular events (32). Other than LDL cholesterol reduction, these drugs also effectively reduce total cholesterol, non-

HDL cholesterol, APOβ, and lipoprotein (a) (LPA), while increasing HDL cholesterol and apolipoprotein A-1 (33).

Pleiotropic effects of PCSK9 inhibitors have also been established. In vitro studies have shown that these effects include reduction of nuclear factor kappa-light-chain-enhancers of activated B-cells and inhibition of pro-inflammatory cytokines such as interleukin-1 (IL-1) and interleukin-6 (IL-6), as well as other pathologic mediators such as monocyte chemoattractant protein-1, and Toll-like receptor-4 (35). Clinical trials on this topic are ongoing (33).

1.8. ACUTE CORONARY SYNDROMES (ACS)

1.8.1. Acute coronary syndromes in general

Acute coronary syndrome (ACS) refers to a group of conditions characterized by sudden onset of acute chest pain, which raises the suspicion of heart disease. ACS is a syndrome where blood flow to the heart is limited due to the occlusion of one or more coronary arteries. The occlusion is most commonly caused by a ruptured atheroma, or less commonly atheroma erosion. (36, 37). Clinical consequences and severity of the condition usually depend on location and degree of obstruction within the coronary vessels. ACS is usually symptomatic and acute heart condition, while most common risk factors for the development of ACS include, obesity, smoking, arterial hypertension, dyslipidemia, diabetes mellitus, male sex, physical inactivity, poor nutrition, and a family history of early MI (38).

ACS is a spectrum of acute coronary disease that encompasses clinical entities such as unstable angina (UA), non-ST-segment elevation myocardial infarction (NSTEMI), and ST-segment elevation myocardial infarction (STEMI). These conditions involve acute coronary ischemia and are clinically distinguished based on presenting symptoms, electrocardiogram (ECG) changes suggestive of ischemia, and circulating levels of cardioselective enzymes reflecting myocardial injury such as cardiac troponins I or T (cTnI/cTnT) or high-sensitivity cardiac troponin (hs-cTn) laboratory assays. The importance of distinguishing these types of ACS is related to the type of treatment and evaluation of prognosis (37).

Diagnostic algorithm and triage in ACS based on symptoms/vital signs, ECG changes and initial troponin levels at presentation and dynamics of troponin change (cTn Δ during 1, 2 or 3 hours during admission) is presented in **Figure 1**.

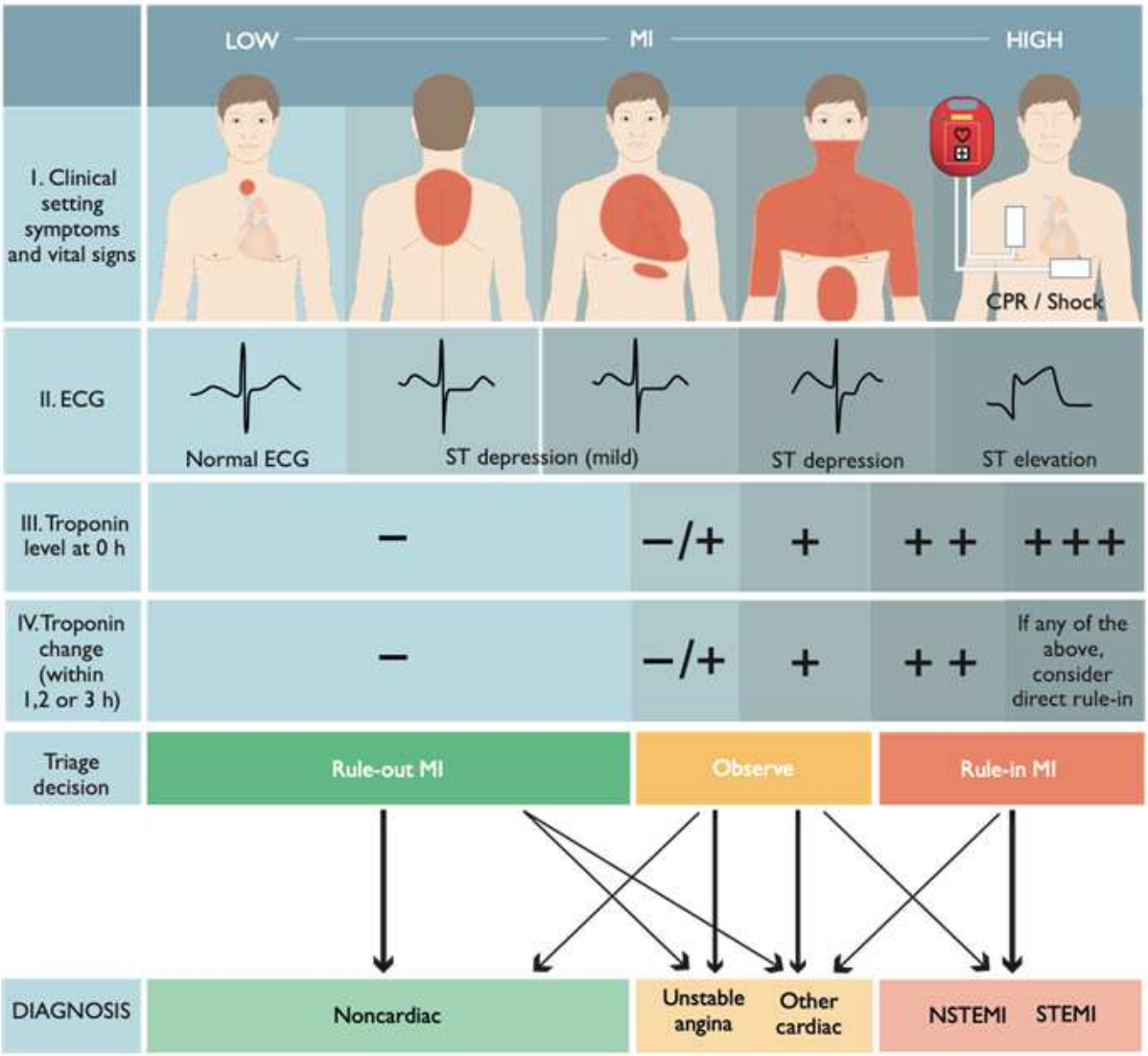

Figure 1. Diagnostic algorithm and triage in acute coronary syndrome (taken from Collet JP, Thiele H, Barbato E, Barthélémy O, Bauersachs J, Bhatt DL, et al. 2020 ESC Guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation. *Eur Heart J*. 2021;42:1289–367)

The incidence of ACS tends to hold a male-to-female ratio of about 3:2, and the mean age in the United States for the first index ACS event is 68 years (39). Estimations show that >780 000 persons will have an episode of ACS, and about 70% will present as NSTEMI. In Croatia, women tended to present with ACS about 7 years later than men and they represented nearly one-third of ACS patients (40). The underlying pathophysiology of ACS is reduced blood flow to certain parts of the myocardium causing ischemia (damage) or death and necrosis of myocytes (infarction). Most of the plaques precipitating ACS are not a result of a gradual

narrowing of the vessel lumen but rather a result of abrupt disruption of vulnerable atherosclerotic plaques (41).

The mismatch between increased oxygen demand and limited supply may also be caused by several non-atherosclerotic mechanisms that limit the blood flow through coronary vessels such as vasospastic angina, Takotsubo cardiomyopathy (stress-induced cardiomyopathy), microvascular angina, coronary artery embolism, spontaneous coronary artery dissection (SCAD), myocardial bridging, or coronary arteritis (42). Other systemic conditions that can precipitate non-obstructive forms of ACS (also known as type 2 MI) typically include hypotension, arterial hypertension (such as in hypertensive emergency), severe anemia, hypertrophic cardiomyopathy, tachycardia, or valvular pathologies such as aortic stenosis. Myocarditis, cardiotoxic drugs, and cardiac contusion are examples of non-ischemic myocardial injury that may lead to supply-demand mismatch and resulting ACS (39).

1.9. Acute coronary syndromes without persistent ST-segment elevation (NSTE-ACS)

In clinical practice, NSTE-ACS presentations are broadly defined as those that lack distinctive ECG features that are seen in STEMI such as ST-segment elevation (STEMI is an immediate electrocardiographic diagnosis). NSTE-ACS is further classified as NSTEMI and UA and these two are differentiated based on ECG changes and measured troponin values. However, one has to bear in mind that many ACS presentations despite the absence of ST-segment elevation on the 12-lead ECG can still signify severe coronary artery disease, and in fact, there are many *"atypical"* ECG patterns or "STEMI-equivalents" that are associated with poor prognosis and would require immediate reperfusion such as de Winter pattern, Wellens syndrome, hyperacute T waves, new-onset left bundle branch block (LBBB) including paced rhythm and right bundle branch block (RBBB) (43-46). About 30% of NSTEMIs are associated with total occlusion of a coronary artery thus these patients are at high risk of mortality and complications but are often not managed according to a STEMI-like pathway (47). This is why some of the cardiovascular scientists such as Smith, Aslanger, and Meyers call for the replacement of clinically established STEMI/NSTE-ACS dichotomy by the concept of occlusion *vs.* non-occlusion MI (OMI/NOMI paradigm) as this concept is more accurate in identifying coronary occlusion MI per ECG criteria (48-50). These important implications should be kept in mind but are out of the scope of this thesis.

As previously stated, most cases of NSTE-ACS are caused by the sudden rupture of the unstable atherosclerotic plaque within the coronary tree. Suspicion of ACS must systematically be evaluated by incorporating data obtained from patient history, symptoms and signs found on physical examination, 12-lead ECG tracings, and laboratory workup of cardioselective enzymes as previously shown in **Figure 1**. The most common chief complaint is substernal central chest pain that is pressurizing in character and that might radiate to either of the shoulder, neck, or jaw, occurring at rest or with minimal physical exertion thus lasting more than 10 minutes. Other presenting symptoms may include dyspnea, and less common, nausea, abdominal pain, syncope, and/or diaphoresis. Atypical symptoms may be seen in the following patients; patients older than 75 years of age, women, diabetic patients, and those with impaired renal function. These atypical symptoms include epigastric pain, indigestion, stabbing or pleuritic pain, or increasing dyspnea in the absence of chest pain (39, 51).

In NSTE-ACS, the 12-lead ECG may show a heterogeneous and broad variety of ECG patterns, such as ST-segment depression, T-wave flattening (applanation), or inversion, biphasic T-waves or it may have minimal non-specific changes or even seem completely normal. In such cases, sequential recording of multiple ECG tracings during the period of the next few hours might be helpful since it might require some time for these changes to be captured on the ECG. A transient ST-segment elevation may also be seen but it does not persist. Another important part of the diagnosis of NSTE-ACS is the positivity (rise) of cardioselective enzymes such as cardiac troponins which indicate a myocardial injury and are helpful in differentiating NSTEMI from unstable angina (UA) (52).

1.9.1. NSTEMI

NSTEMI is caused by complete or partial occlusion of a coronary artery, causing downstream myocardial ischemia with cardiomyocyte necrosis. In approximately 20% to 35% of cases the flow-limitation in NSTEMI is caused by the total vessel obstruction (52-54, 47). Similarly, the proportion of patients having three-vessel disease and/or left main stenosis in NSTE-ACS is substantial (55). Diagnosis of NSTEMI is based on the patient's symptoms, ECG changes, and cardiac troponin levels. The presence of elevated cardiac troponins in the setting of NSTEMI is essential to distinguish it from unstable angina, and this has downstream implications for the patient treatment and prognosis/risk stratification (56).

The current standard of care (SoC) medical treatment for NSTEMI should be personalized by making individual patient risk stratification concerning ischemic and bleeding risk (37). SoC treatment for early hospital care includes the administration of oxygen if peripheral oxygen saturation (SpO2) is <90% or a patient experiences respiratory distress or has other high-risk features for hypoxemia. Nitrates are indicated for continuous ischemic pain, heart failure, or hypertension, however, should be avoided when right ventricular (RV) infarction is suspected or if the patient is hypotensive. Analgesic treatment may include intravenous morphine sulfate for continuous ischemic chest pain despite maximally tolerated anti-ischemic medication. Beta-adrenergic blockers are administrated within 24 h in the absence of contraindications. In the case of contraindication for beta-blockers, calcium channel blockers are the alternative drug in the absence of contraindication. Ivabradine can also be used for acute heart rate lowering, however, this is off-label indication at the current time (57). Immediate cholesterol management with statins is the last pharmacological cornerstone in the treatment of NSTEMI in the absence of contraindications and should be initiated as soon as possible regardless of baseline cholesterol levels (IA class of recommendations in ESC NSTE-ACS guidelines and IB class of recommendation in American AHA/ACC guidelines (37, 39).

Timely and effective antithrombotic treatment is a paramount pharmacological intervention in NSTE-ACS and ACS in general and it is critical in improving outcomes and survival in this patient population. By providing antithrombotic medications we achieve an effective platelet inhibition, blunt further thrombus formation, and provide systemic anticoagulation that can allow for the restoration of vessel patency and thrombotic resolution (58).

A loading dose of aspirin (usually 150-300 mg) is the mainstay of antiplatelet therapy, followed by a lower maintenance dose (usually 75-100 mg per day). A second antiplatelet agent in the form of P_2Y_{12} receptor antagonist is added to aspirin, and according to current ESC NSTE-ACS guidelines, this second agent should not be administered routinely in a patient presenting with NSTE-ACS until the coronary anatomy is known and in whom the early invasive management is planned (37). Together, aspirin and P_2Y_{12} receptor antagonist constitute what is also known as a dual antiplatelet treatment or DAPT. P_2Y_{12} receptor antagonists include older agents such as clopidogrel, or newer agents such as ticagrelor or prasugrel. Prasugrel should be considered in preference to ticagrelor in NSTE-ACS patients who proceed to percutaneous coronary intervention (PCI).

In addition to antiplatelet treatment, parenteral anticoagulation is recommended in all patients at the time of NSTE-ACS diagnosis and especially during revascularization procedures

according to both ischaemic and bleeding risks (IA class recommendation). Unfractionated heparin (UFH) is the drug of choice and can be given in a combination with GP IIb/IIIa inhibitor, especially in those NSTE-ACS patients undergoing PCI (IA class recommendation). Other anticoagulation alternatives (with weaker levels of recommendations in the current ESC guidelines) in selected cases could include fondaparinux, bivalirudin, or low-molecular-weight heparins such as enoxaparin. Regardless of the clinical scenario, a precision balancing and estimation of individual patient's bleeding and ischemic risk is warranted at all times.

A brief graphic summary of pharmacologic antithrombotic treatments in NSTE-ACS encompassing antiplatelet and anticoagulation drugs is shown in **Figure 2**.

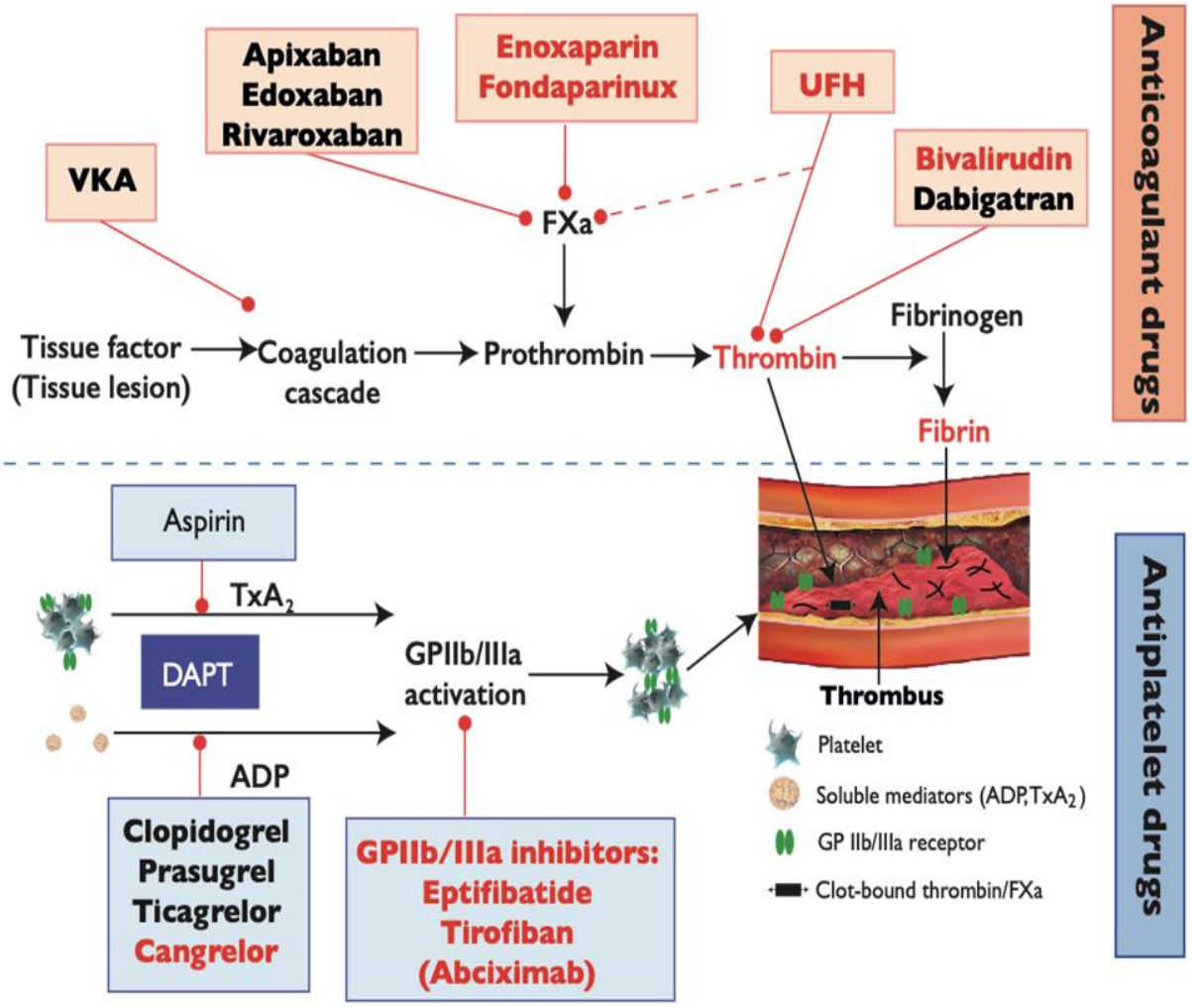

Figure 2. Antithrombotic treatments* and pharmacological targets in non-ST-segment elevation acute coronary syndrome (taken from Collet JP, Thiele H, Barbato E, Barthélémy O, Bauersachs J, Bhatt DL, et al. 2020 ESC Guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation. *Eur Heart J.* 2021;42:1289–367)

*Drugs with oral administration are shown in black letters and drugs with preferred parenteral administration are shown in red.

Invasive management in the form of coronary angiography among patients with NSTE-ACS helps to clarify whether presumed anginal chest pain is a consequence of myocardial ischemia that is caused by the culprit coronary lesion and significant stenosis. Once diagnostic coronary angiography is performed and coronary anatomy of a patient with NSTE-ACS is visualized, if the culprit lesion is identified then it should be treated either with PCI or coronary artery bypass graft (CABG) surgery depending on the lesion morphology and the patient risk

profile. Several percutaneous revascularization strategies are available depending on clinical scenarios: selective invasive, early invasive, and immediate invasive approach.

Immediate invasive strategy (<2 h from hospital admission) which includes coronary angiography and revascularization is indicated in very high-risk NSTE-ACS patients with at least one very high-risk criterion, with the aim of prompt vessel reperfusion.

Early invasive strategy with coronary angiography should be performed within 24 h of hospital admission in high-risk patients as shown in **Figure 3.**

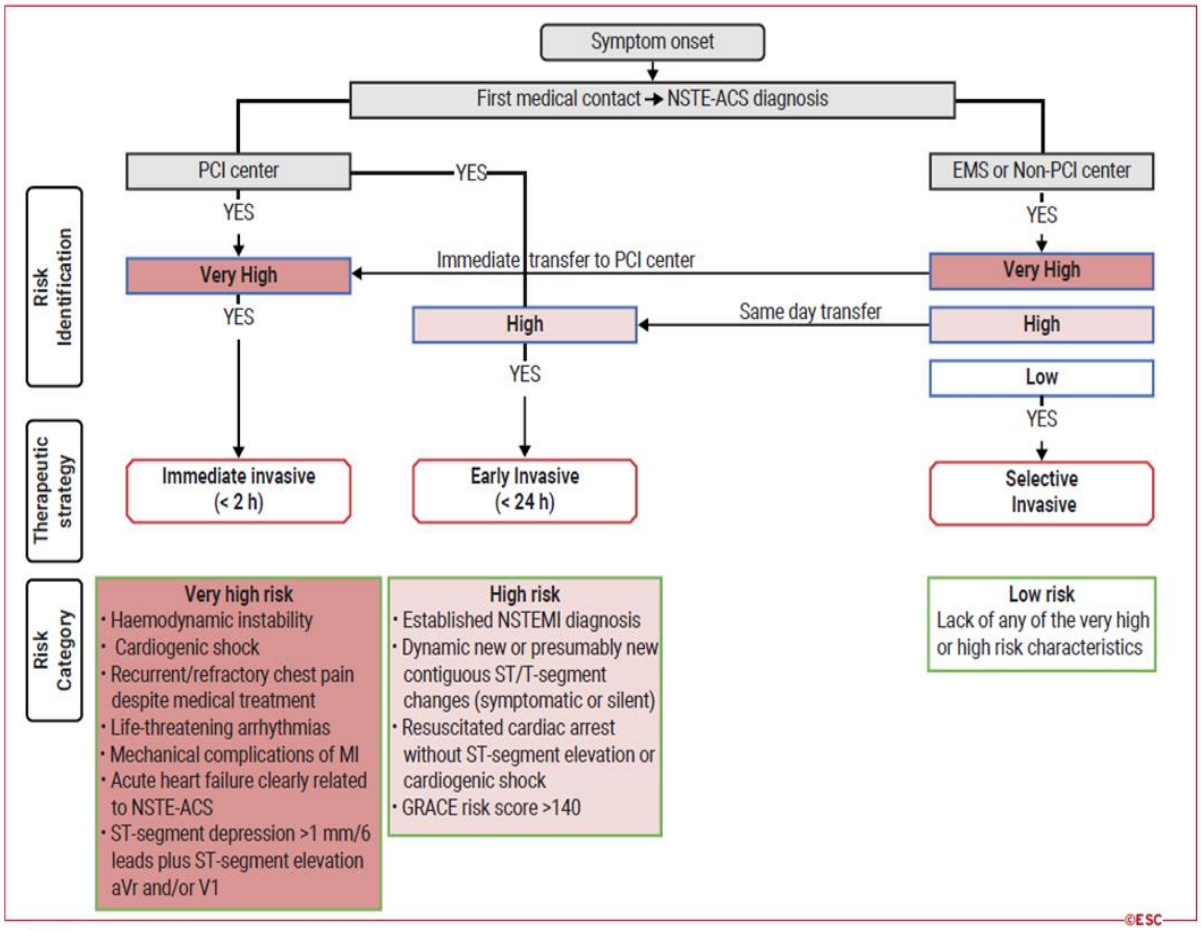

Figure 3. Non-ST-segment elevation acute coronary syndrome percutaneous treatment strategy and timing according to initial risk stratification (taken from Collet JP, Thiele H, Barbato E, Barthélémy O, Bauersachs J, Bhatt DL, et al. 2020 ESC Guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation. *Eur Heart J*. 2021;42:1289–367)

Abbreviations: EMS = emergency medical services; GRACE = Global Registry of Acute Coronary Events; MI = myocardial infarction; NSTE-ACS = non-ST-segment elevation acute coronary syndrome; NSTEMI = non-ST-segment elevation myocardial infarction; PCI = percutaneous coronary intervention.

1.9.2. Unstable angina (UA)

Unstable angina is a part of the ACS and NSTE-ACS continuum as it is clinically characterized by the ischemia of myocardium at rest or with minimal physical exertion resulting from impeded blood flow to the heart due to the formation of non-occlusive thrombus in the lumen of a coronary artery, just like in some cases of NSTEMI. Other etiologies excluding atherothrombosis might involve vasospasm, and hypertensive heart disease due to increased cardiac afterload. In the presence of a narrowed coronary artery lumen, an increase in oxygen demand of the myocardium may result in anginal symptoms due to ischemia without cardiomyocyte cell damage (so-called functional ischemia). Factors that are known to increase myocardial oxygen demand and precipitate acute chest pain in the setting of existing fixed coronary stenosis limiting oxygen supply are as follows: tachyarrhythmias (especially atrial fibrillation with fast ventricular rate), fever (infection), arterial hypertension, cocaine use, aortic stenosis, AV shunt, anemia, thyrotoxicosis, pheochromocytoma, and chronic heart failure. In contrast to NSTEMI, patients with UA do not experience myocardial necrosis and have a lower risk of death (37, 59).

Diagnosis of UA is dominantly clinical and follows the same diagnostic approach as for NSTEMI with respect to the evaluation of patient history, physical signs and symptoms, and possibly electrocardiogram findings. In most cases of UA, ECG changes are non-specific or ECG is fully normal while cardiac troponin levels are within the normal range. In clinical practice, it is the elevation of cardioselective enzymes that differentiates NSTEMI from UA.

Similarly, as in the setting of NSTEMI, treatment for UA is directed against the propagation of intracoronary thrombus, and towards the restoration of balance between myocardial oxygen supply and demand. Initial treatment is similar to that of NSTEMI and includes antithrombotic and anticoagulation treatment as well as lipid management with statins, management of hypoxemia (if present), and analgesic management of pain. Therapeutic intervention for UA is in the vast majority of cases confined to non-invasive (conservative) medical management that is less intensive than in patients with NSTEMI, however, select patients with protracted and refractory chest pain or high-risk features calculated by risk stratification scoring systems might benefit from more aggressive interventions to restore coronary blood flow such as PCI or CABG (37).

Finally, patients with the absence of high-risk features and with no recurrence of symptoms should be managed according to the 2019 European Society of Cardiology (ESC) Guidelines for diagnosis and management of chronic coronary syndrome (60).

1.10. Role of statins in NSTE-ACS as per current international guidelines

As long as there are no contraindications, all patients presenting with NSTE-ACS should be started on a cholesterol-lowering regimen with statins or continued for those that already have a prescription for statins. Statin management for these patients is important for the risk reduction of recurrent coronary events, recurrent vessel revascularization, stroke, and its use also appears to be associated with lower overall mortality risk (39). Patients that are considered to be at high-risk of CV events have shown to have the greatest benefit from using atorvastatin and other high-intensity statins, as it has been proven in seminal studies (61, 62). High-potency statins in high doses (20-40 mg for rosuvastatin and 40-80 mg for atorvastatin) can reduce LDL cholesterol by more than 50 %, in comparison to moderate- or low-intensity statins (39). Regardless of the initial LDL cholesterol concentrations at the time of ACS presentation, statin therapy should be initiated as early as possible to mitigate the risk of future adverse cardiovascular events. High-dose atorvastatin is known to reduce the risk of events before, during, and after the invasive procedure in both patients that have never been prescribed statins, and those that are currently on chronic therapy with these cholesterol-lowering drugs. The goal of statin treatment in those with established CAD (which automatically classifies them at very high risk for CV events) is to lower LDL concentration to less than 1.4 mmol/L (<55 mg/dL). If the baseline serum concentration of LDL cholesterol is 1.8 to 3.5 mmol/L (70 to 135 mg/dL), the treatment goal should be to lower the serum level LDL cholesterol level by a minimum of 50 % (37).

1.11. The use of rosuvastatin loading in acute coronary syndrome and PCI setting.

The role of rosuvastatin in acute coronary syndromes and NSTE-ACS has been sporadically evaluated and no large trials have been performed in this setting. Similarly, the optimal timing of rosuvastatin administration with respect to PCI procedure is not clearly defined. For these reasons and due to the scarcity of evidence in this domain, this thesis was designed to evaluate and to accumulate existing data from available randomized evidence to answer the question of whether rosuvastatin, the most potent commercially available oral statin compound, would be able to impact on short-term morbidity and mortality outcomes in NSTE-ACS.

Lipid-lowering, anti-inflammatory and positive immunomodulatory effects of rosuvastatin, administered in high-dose demonstrated a potent reduction of pro-inflammatory

mediators and blood viscosity in patients with ACS (63-65) while also nephroprotective effects of rosuvastatin in terms of prevention of contrast-induced acute kidney injury were demonstrated in elderly patients with ACS (66). A single high loading dose of rosuvastatin 2-4 hours before PCI in NSTE-ACS was associated with a significant attenuation of the post-procedural increase in hs-CRP and IL-6, as well as troponin I and CK-MB thus demonstrating cardioprotective effects of rosuvastatin in this setting (67). High-dose rosuvastatin loading before PCI in NSTE-ACS can also reduce periprocedural myocardial injury and periprocedural inflammation cytokine release (68).

Finally, there is a solid body of clinical evidence that corroborates the efficacy of rosuvastatin in LDL cholesterol lowering and cardioprotection, and based on limited randomized trial data, early use of high-dose rosuvastatin in patients with NSTE-ACS should be systematically evaluated and large scale trials should be initiated in this setting in the future. There is also a sound biological and mechanistic plausibility that lipid-lowering, anti-inflammatory, and other pleiotropic effects of rosuvastatin might provide beneficial effects in the acute setting of NSTE-ACS and confer cardioprotection that might translate to improved clinical outcomes, especially in the short-term period and if the patient is reperfused with PCI.

Due to the paucity of evidence in this setting, the goal of this thesis and the current study was to analyze literature and perform a meta-analysis of randomized clinical trials that investigated the use of early high-dose rosuvastatin loading in patients with NSTE-ACS with planned invasive management to determine its potential impact on short-term clinical outcomes such as major adverse cerebrovascular and cardiovascular events (MACCE), myocardial infarction and all-cause death at 30 days.

2. OBJECTIVES

2.1. Aims of the study

The present study aimed to investigate the effects of early high-dose rosuvastatin loading (pretreatment) *vs.* placebo or no loading across randomized controlled trials that enrolled patients presenting with acute coronary syndromes without persistent ST-segment elevation (NSTE-ACS) scheduled to undergo percutaneous coronary intervention (PCI) concerning the following short-term outcomes:

a) Major adverse cerebrovascular and cardiovascular events (MACCE) at 30 days
b) Recurrent myocardial infarctions (MIs) at 30 days
c) All-cause mortality at 30 days
d) MACCE at 30 days among patients of which all received PCI (100% of PCI receipt)

2.2. Hypotheses

Regarding the prespecified aims of the study, the following hypotheses were proposed:

a) Early rosuvastatin loading will be associated with a lower likelihood of MACCE at 30 days among patients with NSTE-ACS compared to placebo or no loading.
b) Early rosuvastatin loading will be associated with a lower likelihood of recurrent MI at 30 days in patients with NSTE-ACS compared to placebo or no loading.
c) Early rosuvastatin loading will be similar to placebo or no loading in patients with NSTE-ACS with respect to the outcome of all-cause mortality at 30 days.
d) Early rosuvastatin loading will be associated with a lower likelihood of MACCE at 30 days among patients with NSTE-ACS that all received PCI compared to placebo or no loading.

3. PATIENTS AND METHODS

3.1. Study design

This diploma thesis was designed as a systematic review of the literature and meta-analysis of randomized controlled trials (RCTs) investigating the impact of early high-dose rosuvastatin loading in statin-naive patients with NSTE-ACS undergoing PCI on the short-term (30 days) outcomes of MACCE, myocardial infarction, and all-cause death. No prespecified protocol was registered before performing this analysis and no Ethics Committee approval from the University of Split School of Medicine was required for the study of this design. This study was carried out under the Department of Pathophysiology, University of Split School of Medicine.

3.2. Search strategy

The search strategy was developed by the student mentor (JAB) while the search of electronic databases was independently carried out by the student (MLO) and student mentor (JAB). Electronic databases included in the search were the National Library of Medicine – PubMed, Ovid MEDLINE, Cochrane Central Register of Controlled Trials, Ovid Journals (full text), and SCOPUS. These databases were manually searched to obtain full records of original articles (RCTs) that investigated the use of high-dose rosuvastatin loading in NSTE-ACS patients. The search was limited to records published in relevant peer-reviewed journals in the English language from 2000 until 2021 that included adult human subjects. The date of the last search was performed on August 10th, 2021. No grey literature search was performed and no external authors were contacted to provide additional data or to obtain additional studies. Both the student and mentor independently performed the literature search, deleted duplicate records, screened available titles and abstracts for relevance and classified obtained studies as *„excluded“* or requiring further assessment. Such studies were labeled as *„potential for inclusion“*. Finally, prespecified eligibility and exclusion criteria were applied consistently among potentially inclusive studies. If there was a discrepancy between the two investigators concerning the search strategy, this was resolved by the joint discussion involving the opinion of the external expert from the Department of Pathophysiology, University of Split School of Medicine.

3.3. Inclusion and exlusion criteria, PICOS

To be eligible for potential inclusion, obtained studies had to satisfy a number of inclusion criteria according to PICOS questions, as follows:

1. **Patient population:** statin-naive patients with NSTE-ACS including its clinical subtypes – unstable angina and NSTEMI, scheduled to undergo PCI
2. **Intervention:** patients with NSTE-ACS had to receive a high-dose loading of rosuvastatin (defined as at least 20 mg of rosuvastatin dose prior to PCI) on top of the guideline-directed standard of care treatment that is administered in NSTE-ACS
3. **Comparison:** patients in the control group would need to not receive rosuvastatin (or any other statin) loading (pretreatment) before PCI or would need to be given a placebo pill added to standard of care treatment
4. **Outcome:** the primary outcomes of interest were MACCE at 30 days, myocardial infarction at 30 days, and all-cause death at 30 days. MACCE was defined as a composite endpoint of all-cause death, non-fatal myocardial infarction, and non-fatal stroke, as reported in the studies.
5. **Study design:** studies had to be designed and executed as RCTs to be considered for the potential inclusion in the analysis.

Studies were considered for potential inclusion only if the length of follow-up was designed to capture at least a 30-day period following PCI.

We excluded studies in the following circumstances:

1. If the study had a non-RCT design (*i.e.* observational and/or non-randomized study)
2. If the study did not report on any of the prespecified outcomes of interest or reported number of events regarding the primary outcome in both experimental and control groups or if the study did not provide basic data on study length, description of the main baseline characteristics relevant for the studied population such as age, sex, PCI receipt, periprocedural characteristics, statin type (rosuvastatin) and statin dose
3. If the study enrolled patients with stable coronary artery disease, *i.e.* patients with stable angina or those with chronic coronary syndromes (CCS)
4. If the study enrolled patients that were not naive to statin treatment (*i.e.* patients already on the current statin treatment or with a positive medical history of statin treatment)
5. If the study was not designed to investigate high-dose statin loading before PCI

6. If the study investigated loading with some other statin other than rosuvastatin (*i.e.* atorvastatin, simvastatin, fluvastatin, etc.)
7. If the study was a duplicate report without additional or updated outcome data

3.4. Data items and extraction

Both the student (MLO) and mentor independently extracted data from the included studies by using pre-designed, piloted extraction forms containing baseline study information such as author's first and last name, study design, the total number of patients, and a number of patients stratified by experimental/control group, rosuvastatin dose and timing/route of administration prior to PCI, sex distribution in the experimental and control group, the mean age of experimental and control group, description of the control treatment (placebo or no-statin), percentage of PCI procedures performed in the whole study sample and prespecified primary outcomes of interest as elaborated previously. For each study, we also extracted the prevalence of comorbidities including arterial hypertension, diabetes mellitus, smoking as well as the history of previous MI or PCI. Furthermore, the prevalence of multivessel coronary artery disease (defined as significant stenosis in at least two epicardial coronary vessels as determined by diagnostic angiography). Baseline pharmacotherapy in the experimental and control group was registered with respect to the use of beta-blockers, ACE inhibitors or ARBs, acetylsalicylic acid (ASA), P_2Y_{12} receptor antagonist, and GPIIb/IIIa inhibitors. Finally, post-PCI maintenance antithrombotic and lipid-lowering treatment was also described for each included study.

3.5. Risk of Bias (RoB) assessment

Cochrane's Risk of Bias (RoB) tool, as recommended by the Cochrane Collaboration (69), has been used to assess the individual risk of bias of each included study. RoB assessment included an evaluation of sequence generation of the allocation, allocation concealment, blinding of participants, personnel and outcome assessors, incomplete outcome data, selective outcome reporting, and other sources of bias. RoB was independently performed by the student (MLO) and mentor (JAB) while potential discrepancies were resolved by consultation with the third investigator from the Department of Pathophysiology, University of Split School of Medicine.

3.6. Statistical analysis (quantitative synthesis)

Data analysis was performed by adhering to Cochrane Collaboration recommendations and PRISMA statement (70).

Odds ratio (OR) with 95% confidence intervals (95% CI) was used as the main summary measure for effect estimates on prespecified dichotomous outcomes. Random-effects model with Mantel-Haenszel statistical method was applied for the meta-analysis. Meta-analysis was performed by using Review Manager software (RevMan, version 5.4, The Cochrane Collaboration, 2020). Chi-square test of heterogeneity and Higgins I^2 statistic of inconsistency were used to assess heterogeneity across studies. Studies with an I^2 statistic of 25% to <50% were considered to have low heterogeneity; 50% to 75% - moderate heterogeneity, and those with I^2 statistic >75% were considered to have a high heterogeneity. *P*-values were two-tailed and results were considered statistically significant if $P<0.05$ at all instances.

4. RESULTS

4.1. Study inclusion and risk of bias assessment

A total of 144 records were screened after duplicate records were removed. Out of these records, 95 were excluded because they did not pertain to acute coronary syndromes but other forms of cardiovascular disease. Finally, full texts were obtained for 27 records and were analyzed for potential inclusion in qualitative synthesis and meta-analysis. This resulted in five (5) randomized controlled trials being included in the data analysis as shown in **Figure 4**.

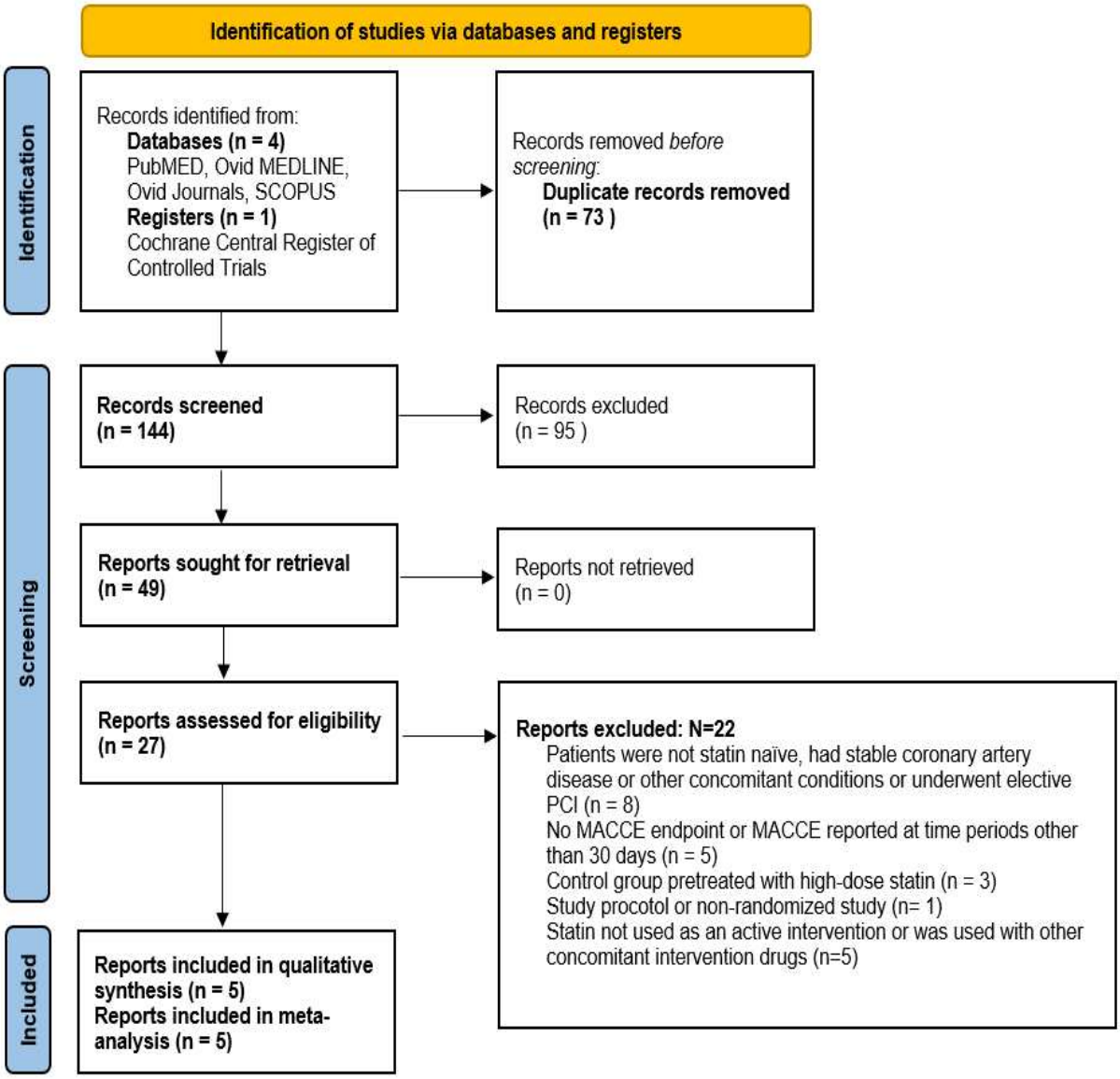

Figure 4. PRISMA flow diagram of study inclusion

Risk of bias assessment (RoB) was independently performed for each of included five RCTs by the principal thesis author (MLO) and thesis mentor (JAB). Summary of the RoB for each included trial is presented in **Figure 5** while the percentage of low, unclear, or high risk of bias judgments across included trials is shown in **Figure 6**. This analysis revealed that most trials had unclear risk concerning selection bias (random sequence generation and allocation concealment) while in two out of five studies blinding procedure (performance bias) was not explicitly defined or treatments were dispensed as an open-label. Included trials generally had a low risk of bias with respect to detection, attrition, reporting, and other potential biases.

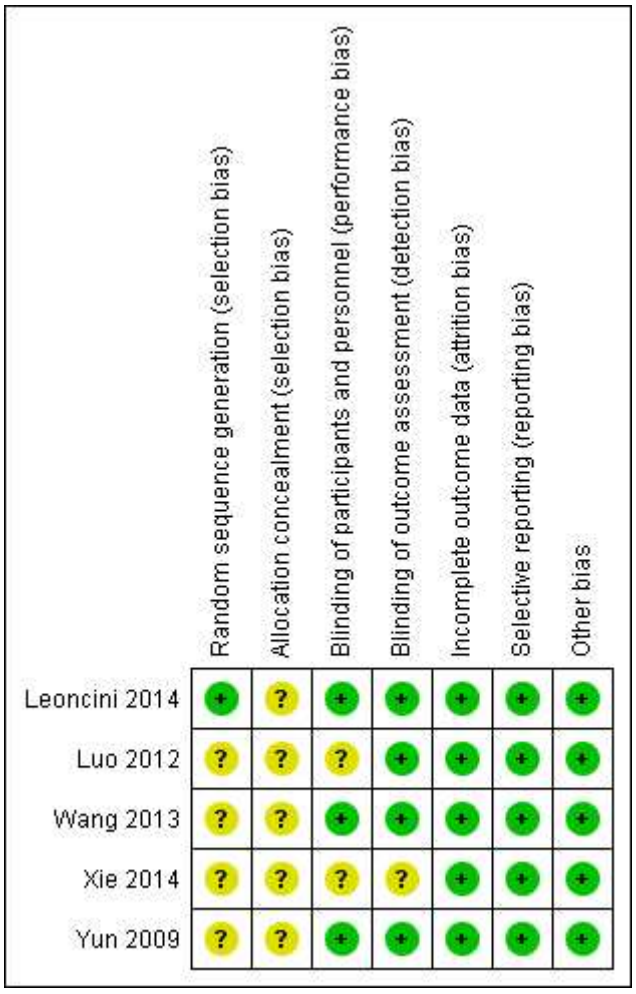

Figure 5. Risk of bias (RoB) summary including authors' judgements about each RoB item for each included study

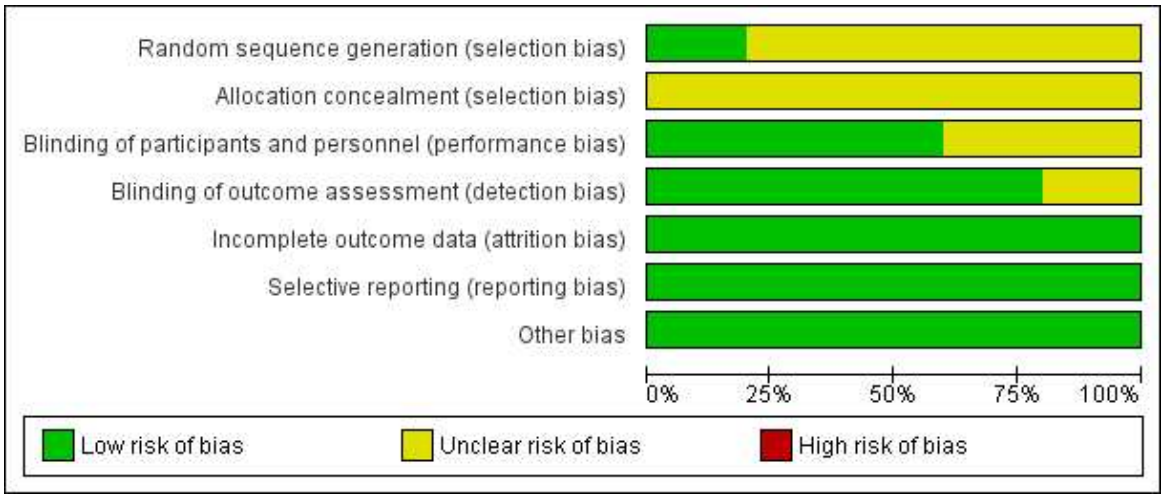

Figure 6. Risk of bias (RoB) graph of review authors' judgements about each RoB item presented as percentages across all included studies

As shown in **Table 2**. all included trials were designed as randomized controlled trials that studied the use of at least 20 mg dose of rosuvastatin administered *per os* before planned PCI as an experimental intervention added to standard of care antithrombotic treatment for NSTE-ACS. On the other hand, control intervention consisted of no statin treatment or placebo pill added to standard of care antithrombotic treatment. MACCE at 30 days was a prespecified primary endpoint in 4 out of five trials while 1 trial provided all data required for the calculation of MACCE at 30 days (Leoncini et al.). All trials included were free from pharmaceutical industry-related funding while four out five trials reported receiving full funding, or at least partial grants from the government, universities, or private foundations. One trial did not state sources of funding within the manuscript (Luo et al.). Four trials were conducted in China, while one was conducted in Italy.

Table 2. Baseline characteristics of included studies per PICOS model

Study	Intervention *vs.* comparator	Primary outcome measured	Study design, single center or multicenter	ITT analysis / sample size calculation / COI declared	Funding	Country
Yun et al. 2009	40 mg *per os* before PCI *vs.* no statin treatment	MACCE at 30 days	**RCT** *(open label)* Single center	No / Yes / No	Supported by grants from the Wonkwang University in 2008 and the Cardiovascular Research Foundation Asia	China
Luo et al. 2012	20 mg *per os* 12 hours before PCI + 20 mg *per os* at 2 hours before PCI *vs.* no statin treatment	MACCE at 30 days	**RCT** *(blinded to outcomes)* Single center	No / No / No	Funding not stated in the manuscript	China
Wang et al. 2013	20 mg *per os* at 2-4 hours before PCI *vs.* placebo	MACCE at 30 days	**RCT** *(DB)* Single center	No / No / No	Qingdao Science and Technology Support Program [2012-1-3-1-(2)-nsh]	China
Xie et al. 2014	20 mg *per os* 12 hours + 20 mg *per os* at 2 hours before PCI *vs.* placebo	MACCE at 30 days	**RCT** *(unclear blinding)* Single center	No / Yes / No	Study was suppored by the Science and Technology Development Plan of Yulin (grant no. Yu Ke Ji 20141002)	China
Leoncini et al. 2014	40 mg *per os* on admission following by 20 mg/day *per os vs.* no statin treatment	Incidence of contrast-induced AKI	**RCT** *(open label)* Single center	No / Yes / No	Study supported by the Centro Cardiopatici Toscani, Italy	Italy

Abbreviations: AKI-acute kidney injury; **COI**-conflict of interest; **DB**-double blinded; **ITT**-intention-to-treat; **MACCE**-major adverse cardiovascular and cerebrovascular events; **PCI**-percutaneous coronary intervention;

As shown in **Table 3**, all trials included patients with acute diagnoses of unstable angina (UA) and NSTEMI. In two trials (Yun et al. and Wang et al.) the large majority of patients had UA as a diagnosis at enrollment while the trial by Leoncini almost exclusively enrolled patients with NSTEMI. Two trials (Luo et al. and Xie et al.) did not make a distinction between UA and NSTEMI in their study. Furthermore, 100% of patients received PCI in four out of five trials

while in a trial by Leoncini et al. two-thirds of patients underwent PCI. Both rosuvastatin loading and control cohorts were well-matched with respect to age (mean age of 63±11 vs. 63±11 years, respectively). Most of the enrolled patients with NSTE-ACS were male (71.2% in the rosuvastatin loading cohort and 68.2% in the control cohort) and the size of trials ranged from a total of 67 to 504 enrolled patients.

Table 3. Clinical characteristics of included studies indicating study size, follow-up period, distribution of NSTE-ACS subtypes, mean age, sex distribution, PCI receipt proportion, and number of patients evaluated *vs.* randomized to treatment

Study	**Unstable angina** N/Total N (%)	**NSTEMI** N/Total N (%)	**PCI** (%)	**Age** R *vs.* C (years)	**Percent of men** R *vs.* C	**Total N** (R/C)	**N evaluated to N randomized**	**FU Reported**
Yun et al. 2009	330/445 (74.2%)	115/445 (25.8%)	100%	64±10 *vs.* 63±11	60% *vs.* 62%	445 (225/220)	445/510 87%	30 days
Luo et al. 2012	67/67 (100%)*		100%	58±12 *vs.* 61±9	90% *vs.* 78%	67 (31/36)	67/78 86%	30 days
Wang et al. 2013	105/125 (84.0%)	20/125 (16.0%)	100%	65±10 *vs.* 65±12	65% *vs.* 65%	125 (62/63)	125/167 75%	30 days
Xie et al. 2014	159/159 (100%)*		100%	62±11 *vs.* 60±11	75% *vs.* 70%	159 (79/80)	159/218 73%	24 hours 30 days
Leoncini et al. 2014	39/504 (7.7%)	465/504 (92.3%)	66%	66±12 *vs.* 66±14	66% *vs.* 66%	504 (252/252)	504/543 93%	30 days 6 months

*Authors didn't discriminate patients with NSTEMI and unstable angina in the study

Abbreviations: **C**-control group; **FU**-follow-up; **NSTEMI**-non-ST-elevation myocardial infarction; **PCI**-percutaneous coronary intervention; **R**-rosuvastatin loading group

4.2. Comorbidities, angiographic disease burden and concomitant cardiovascular treatment at baseline

Overall, more than half of patients with NSTE-ACS across included trials had arterial hypertension, about one-quarter had diabetes mellitus while more than one-third were smokers. Similarly, about 14% of patients experienced myocardial infarction previously while ≈ 11% of patients had a history of percutaneous revascularization. As shown in **Figure 7**, the comorbidity burden was well-balanced between the control and rosuvastatin loading cohorts.

Prevalence of multivessel disease (defined as significant stenosis involving ≥2 epicardial coronary vessels, as assessed by diagnostic coronary angiography) across included trials of patients with NSTE-ACS was substantial (**Figure 8**). In both control and rosuvastatin loading cohorts, more than half of patients had severe angiographic disease burden (ranging from the average 34.4% in a trial by Wang et al. to 68% in a trial by Xie et al).

Regarding the concomitant cardiovascular pharmacotherapy at baseline, both control and rosuvastatin loading cohorts were well-balanced. Of note, the use of beta blockers, ACE inhibitors or ARBs, ASA, P_2Y_{12} inhibitors and glycoprotein IIb/IIIa inhibitors was similar between both groups (**Figure 9**).

In most instances, concomitant antithrombotic treatment or DAPT consisted of 300 mg ASA (aspirin) and clopidogrel (300 to 600 mg dose) administered before PCI, with elective use of GPIIb/IIIa inhibitors and heparin or its derivatives at the discretion of the operator. The post-PCI maintenance DAPT was uniform across trials and dominantly consisted of 100 mg ASA and 75 mg of clopidogrel per day (**Table 4**).

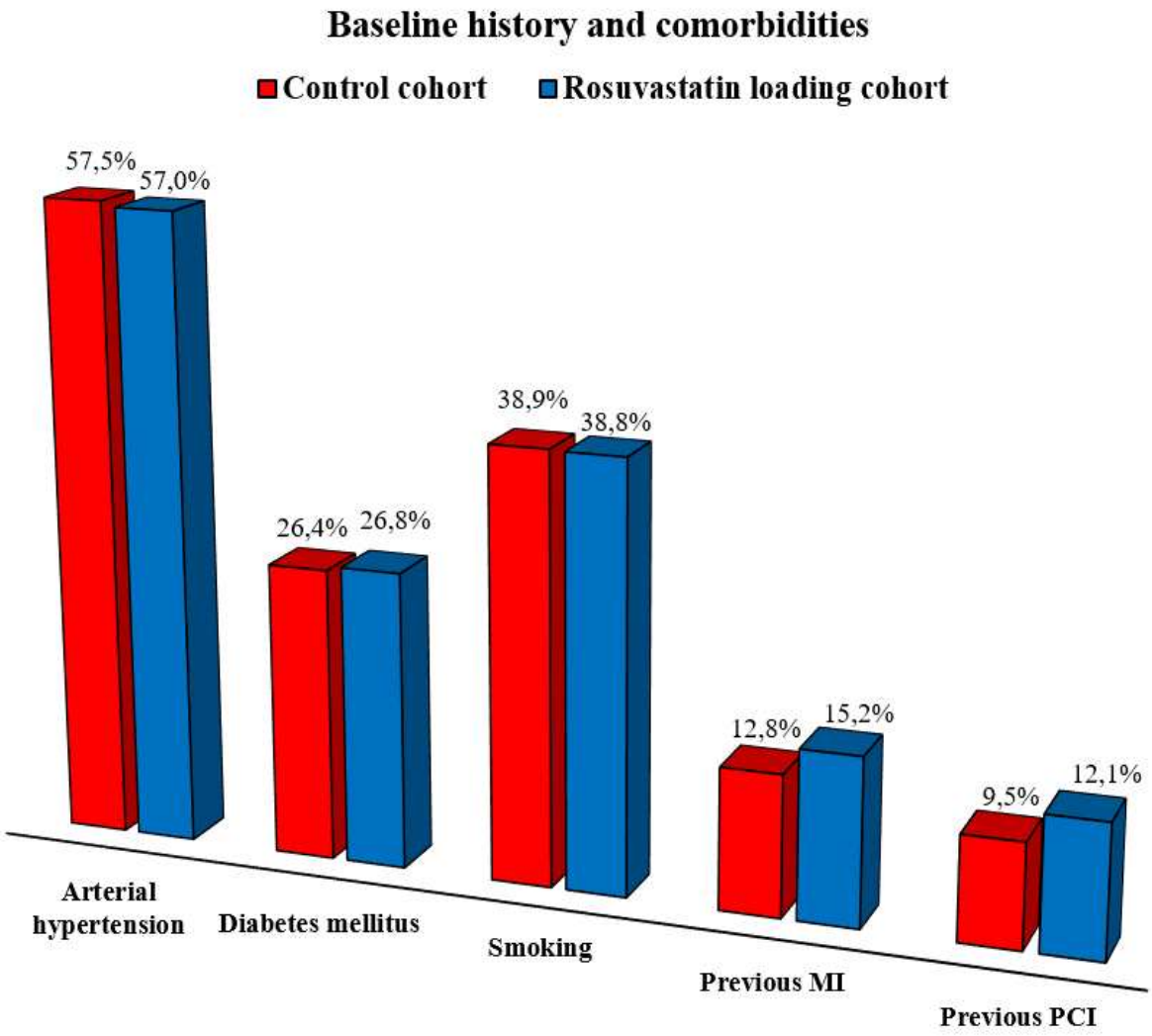

Figure 7. Prevalence of comorbidities such as arterial hypertension, diabetes mellitus, smoking, and history of myocardial infarction (MI) or percutaneous coronary intervention (PCI) according to treatment group

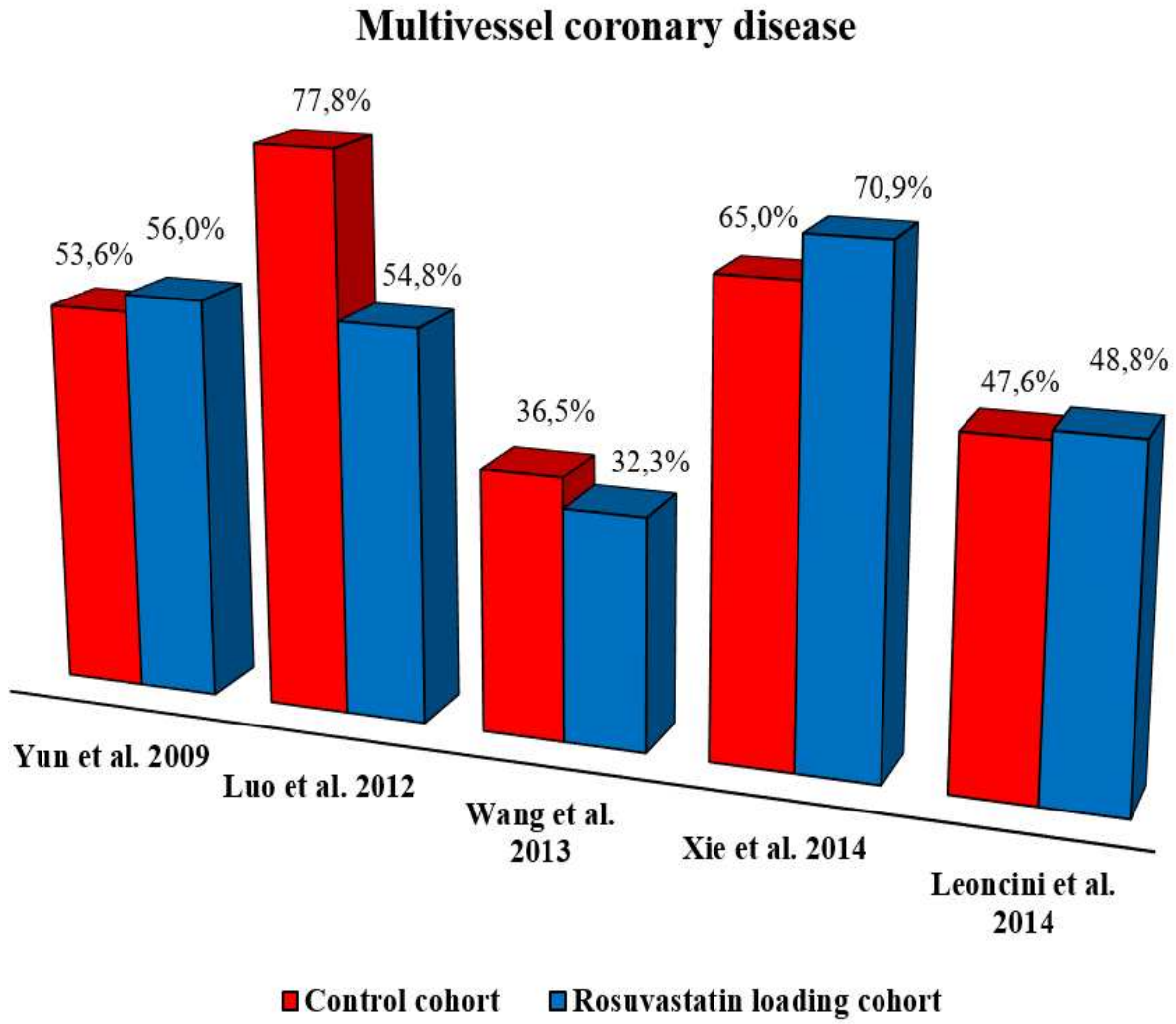

Figure 8. Prevalence of multivessel disease as determined by diagnostic coronary angiography across included studies and according to treatment group

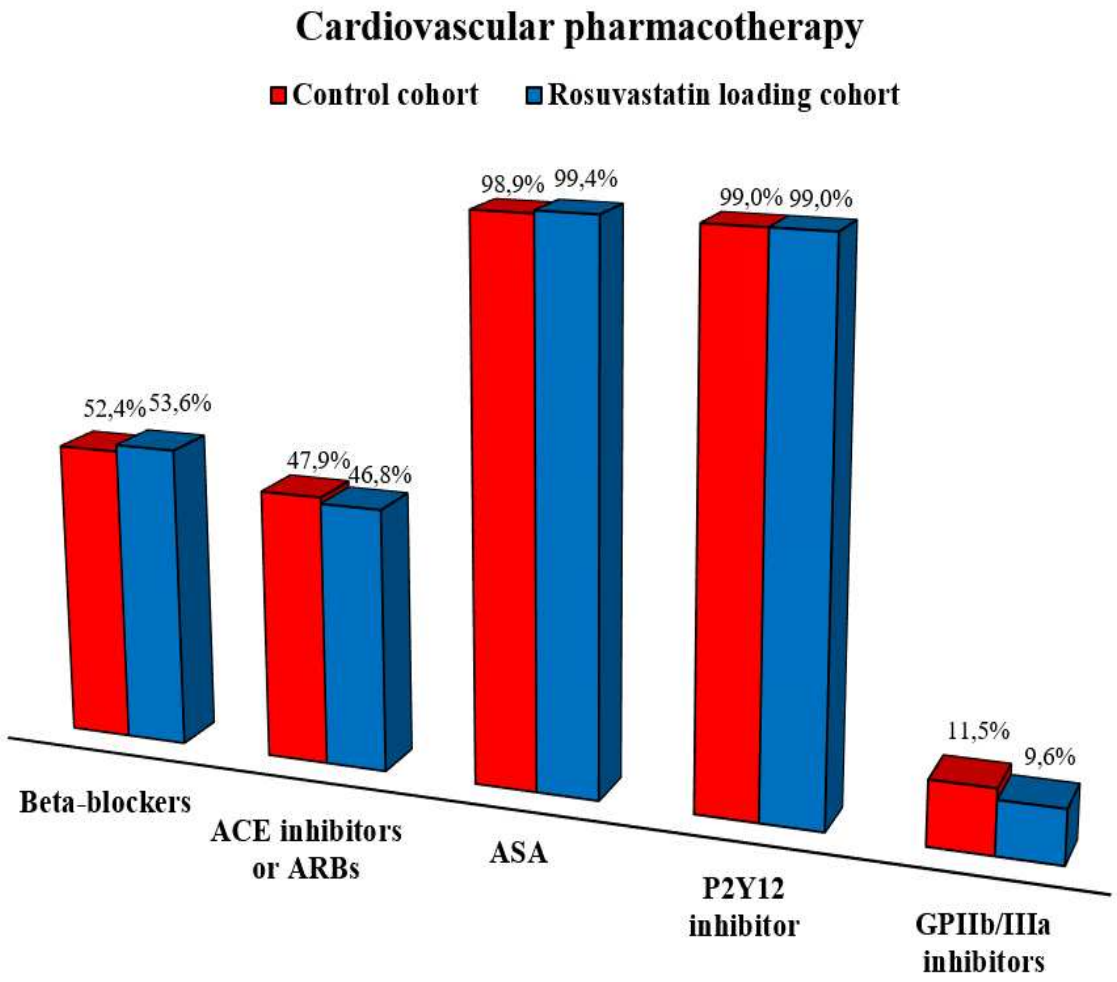

Figure 9. In-hospital use of concomitant cardiovascular pharmacotherapies including beta-blockers, angiotensin-converting enzyme (ACE) inhibitors, angiotensin receptor blockers (ARBs), acetylsalicylic acid (ASA), P_2Y_{12} receptor inhibitors (dominantly clopidogrel), and glycoprotein IIb/IIIa (GPIIb/IIIa) inhibitors at baseline

Table 4. Baseline characteristics of included studies according to baseline statin and antithrombotic treatment

Study	Timing and dose of statin administration	Concomitant antithrombotic medications for ACS prior to PCI	Post-PCI maintenance pharmacotherapy
Yun et al. 2009	**40 mg *per os* before PCI Mean time 16±5 h prior to PCI** (range 7-25 h)	• Aspirin 300 mg • Clopidogrel 300 mg • GPI at operator's discretion	• Aspirin 200 mg/day • Clopidogrel 75 mg/day • Rosuvastatin 10 mg/day
Luo et al. 2012	**20 mg *per os* 12 hours before angiography; further 20 mg *per os* at 2 hours before angiography** (average timing not disclosed)	• Aspirin and clopidogrel co-administered as standard treatment for ACS prior to angiography • Doses not disclosed in the manuscript	• Aspirin 100 mg/day • Clopidogrel 75 mg/day • Rosuvastatin 10 mg/day
Wang et al. 2013	**20 mg *per os* at 2-4 hours before angiography** (average timing not disclosed)	• Aspirin 300 mg • Clopidogrel 300 mg • GPI at operator's discretion	• Aspirin 100 mg/day • Clopidogrel 75 mg/day • Rosuvastatin 10 mg x 1 • Low-molecular-weight heparin subcutaneously for 3 to 5 days after PCI
Xie et al. 2014	**20 mg *per os* at 12 hours before PCI; further 20 mg *per os* at 2 hours before PCI** (average timing not disclosed)	• Unknown • Ony antithrombotic regimen administered after PCI were disclosed	• Aspirin 100 mg/day • Clopidogrel 75 mg/day • Rosuvastatin 10 mg/day
Leoncini et al. 2014	**40 mg *per os* loading on admission following by 20 mg/day *per os* prior to coronary angiography** Median time 22.5 h (IQR 14-43 h) prior to angiography	• Aspirin (dose not disclosed) • Clopidogrel 600 mg loading dose • Unfractionated heparin (dose not disclosed)	• Aspirin 100 mg/day • Clopidogrel 75 mg/day • Rosuvastatin 20 mg/day (or 10 mg/day for patients with eGFR <30 mL/min./m^2) • Atorvastatin 40 mg/day for the control group

Abbreviations: DAPT-dual antiplatelet treatment; **eGFR**-estimated glomerular filtration rate; **GPI-**glycoprotein IIb/IIIa inhibitor; **NSTE-ACS**-non-ST-elevation acute coronary syndrome encompassing unstable angina and NSTEMI; **NSTEMI**-non-ST-elevation myocardial infarction; **PCI**-percutaneous coronary intervention; **STEMI**-ST-elevation myocardial infarction

4.3. Effects of interventions

4.3.1. MACCE at 30 days

All trials contributed to effect estimates with an overall of 1300 patients with NSTE-ACS. High-dose rosuvastatin loading *vs.* no loading or placebo before PCI was associated with an overall 59% reduction in the likelihood of MACCE at 30 days (OR 0.41, 95% CI 0.27–0.60) and this effect was significant (Z=4.52, P<0.001) (**Figure 10**). No significant heterogeneity was detected across analyzed trials (Tau^2=0.00, I^2=0 %, P=0.910).

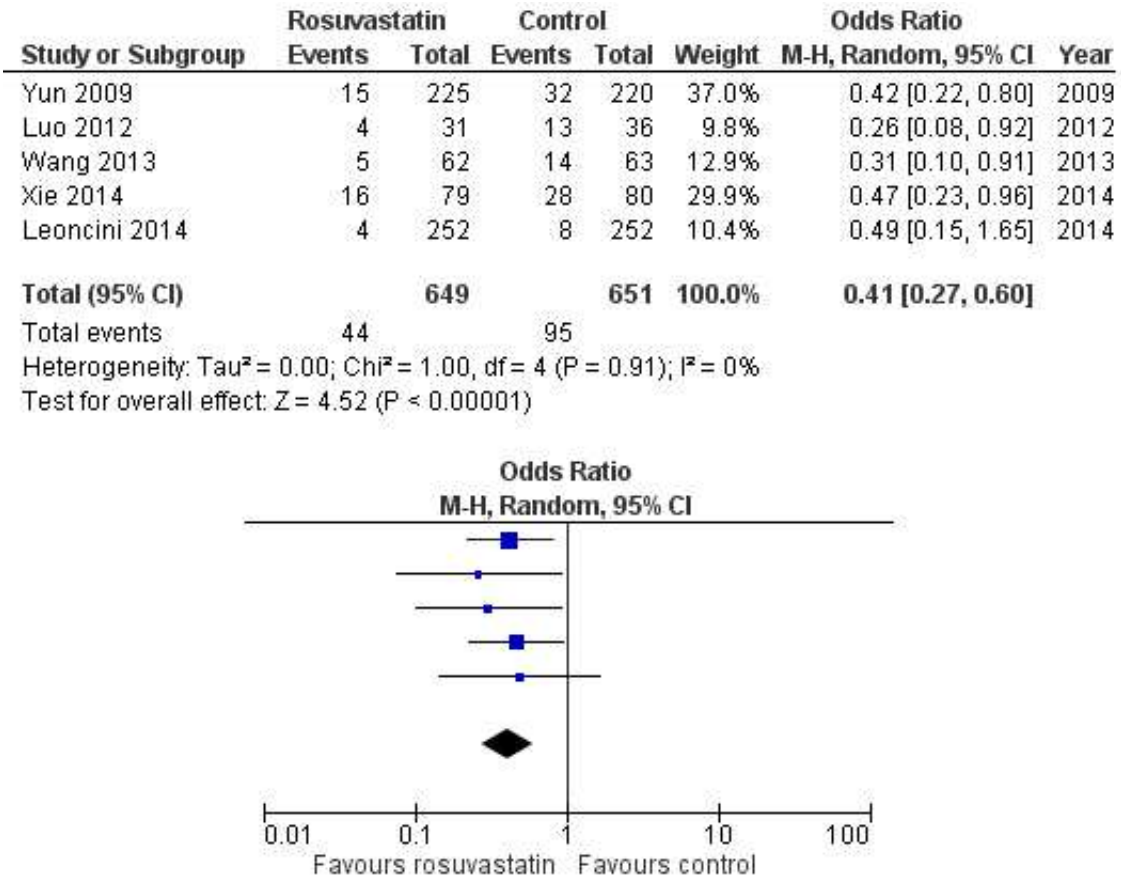

Figure 10. Odds ratio (OR) for the likelihood of experiencing MACCE at 30 days in NSTE-ACS if early high-dose rosuvastatin loading was used *vs.* if it was not (control)

4.3.2. Myocardial infarction at 30 days

All trials contributed to effect estimates with an overall of 1300 patients with NSTE-ACS. High-dose rosuvastatin loading *vs.* no loading or placebo before PCI was associated with an overall 56% reduction in the likelihood of myocardial infarction at 30 days (OR 0.44, 95% CI 0.29–0.67) and this effect was significant (Z=3.85, *P*<0.001) (**Figure 11**). No significant heterogeneity was detected across analyzed trials (Tau^2=0.00, I^2=0 %, *P*=0.810).

	Rosuvastatin		Control			Odds Ratio	
Study or Subgroup	**Events**	**Total**	**Events**	**Total**	**Weight**	**M-H, Random, 95% CI**	**Year**
Yun 2009	14	225	26	220	37.9%	0.50 [0.25, 0.98]	2009
Luo 2012	3	31	11	36	9.1%	0.24 [0.06, 0.97]	2012
Wang 2013	5	62	14	63	14.7%	0.31 [0.10, 0.91]	2013
Leoncini 2014	2	252	5	252	6.4%	0.40 [0.08, 2.06]	2014
Xie 2014	15	79	24	80	32.0%	0.55 [0.26, 1.14]	2014
Total (95% CI)		**649**		**651**	**100.0%**	**0.44 [0.29, 0.67]**	
Total events	39		80				

Heterogeneity: Tau² = 0.00; Chi² = 1.59, df = 4 (P = 0.81); I² = 0%
Test for overall effect: Z = 3.85 (P = 0.0001)

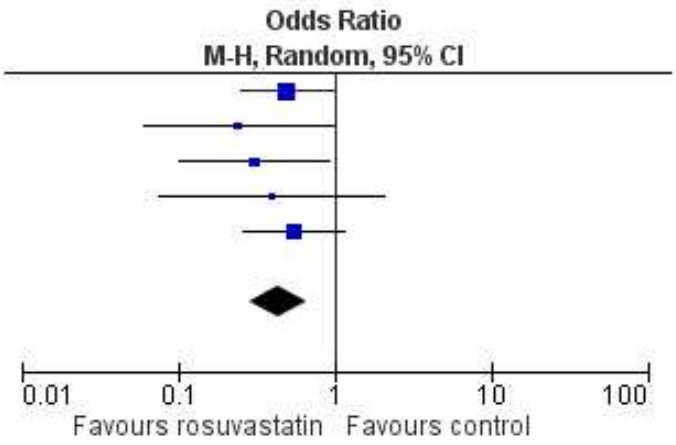

Figure 11. Odds ratio (OR) for the likelihood of experiencing myocardial infarction at 30 days in NSTE-ACS if early high-dose rosuvastatin loading was used *vs.* if it was not (control)

4.3.3. All-cause death at 30 days

Due to zero all-cause death events in three trials, only 2 trials provided estimates for the endpoint of all-cause mortality at 30 days thus accruing 8 events in 949 patients with NSTE-ACS (**Figure 12**). High-dose rosuvastatin loading *vs.* no loading or placebo before PCI was associated with a non-significant 56% reduction in the likelihood of all-cause death at 30 days (OR 0.44, 95% CI 0.09–2.03; *Z*=1.06, *P*=0.290). No significant heterogeneity was detected across included trials (Tau^2=0.00, I^2=0 %, *P*=0.370).

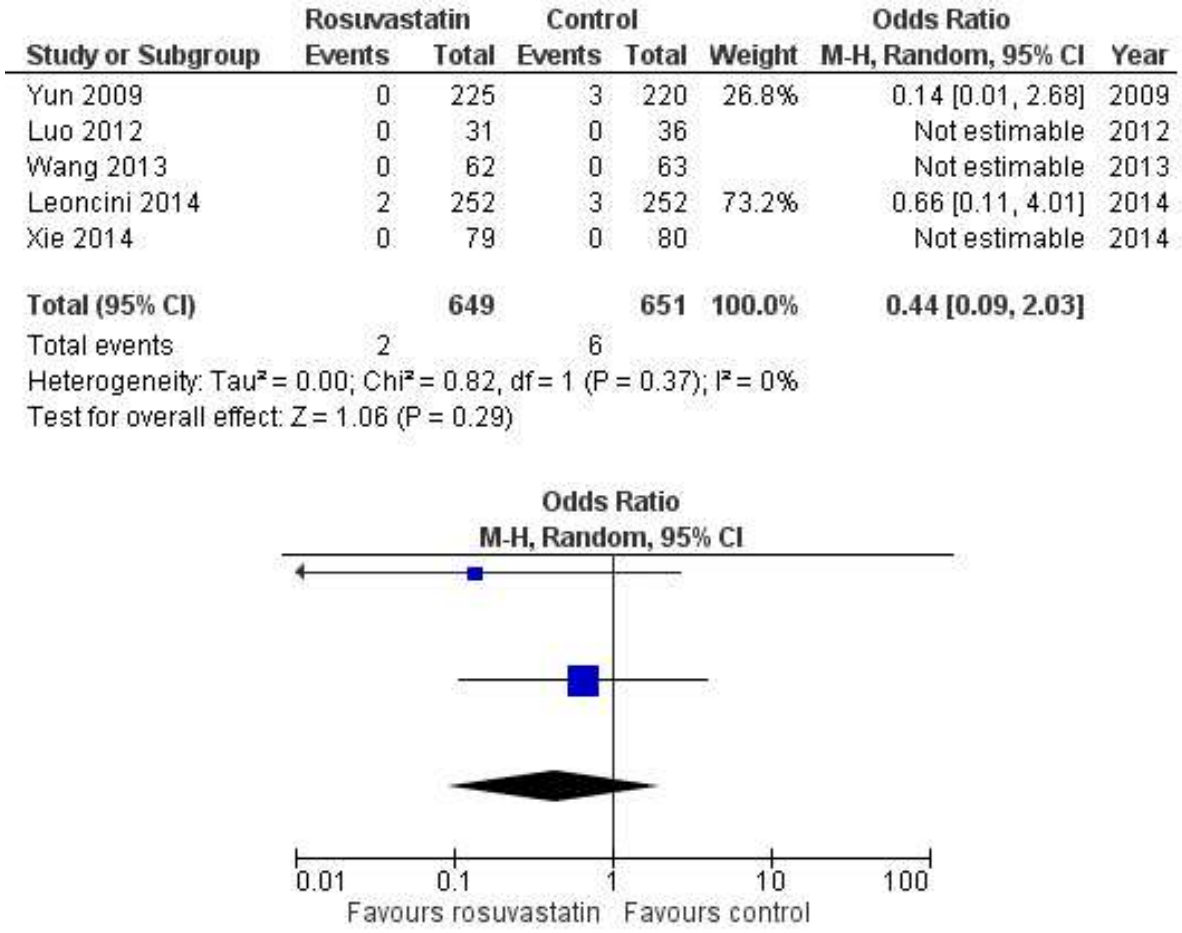

Figure 12. Odds ratio (OR) for the likelihood of all-cause death at 30 days in NSTE-ACS if early high-dose rosuvastatin loading was used *vs.* if it was not (control)

Abbreviations: NSTE-ACS-non-ST-elevation acute coronary syndrome

4.3.4. MACCE at 30 days in trials with 100% of PCI receipt

Four trials contributed to effect estimates with an overall of 796 patients with NSTE-ACS treated with PCI thus generating 130 events. High-dose rosuvastatin loading *vs.* no loading or placebo before PCI was associated with an overall 62% reduction in the likelihood of MACCE at 30 days (OR 0.38, 95% CI 0.25–0.57) and this effect was significant (Z=4.62, *P*<0.001) (**Figure 13**). No significant heterogeneity was detected across analyzed trials (Tau^2=0.00, I^2=0 %, *P*=0.840).

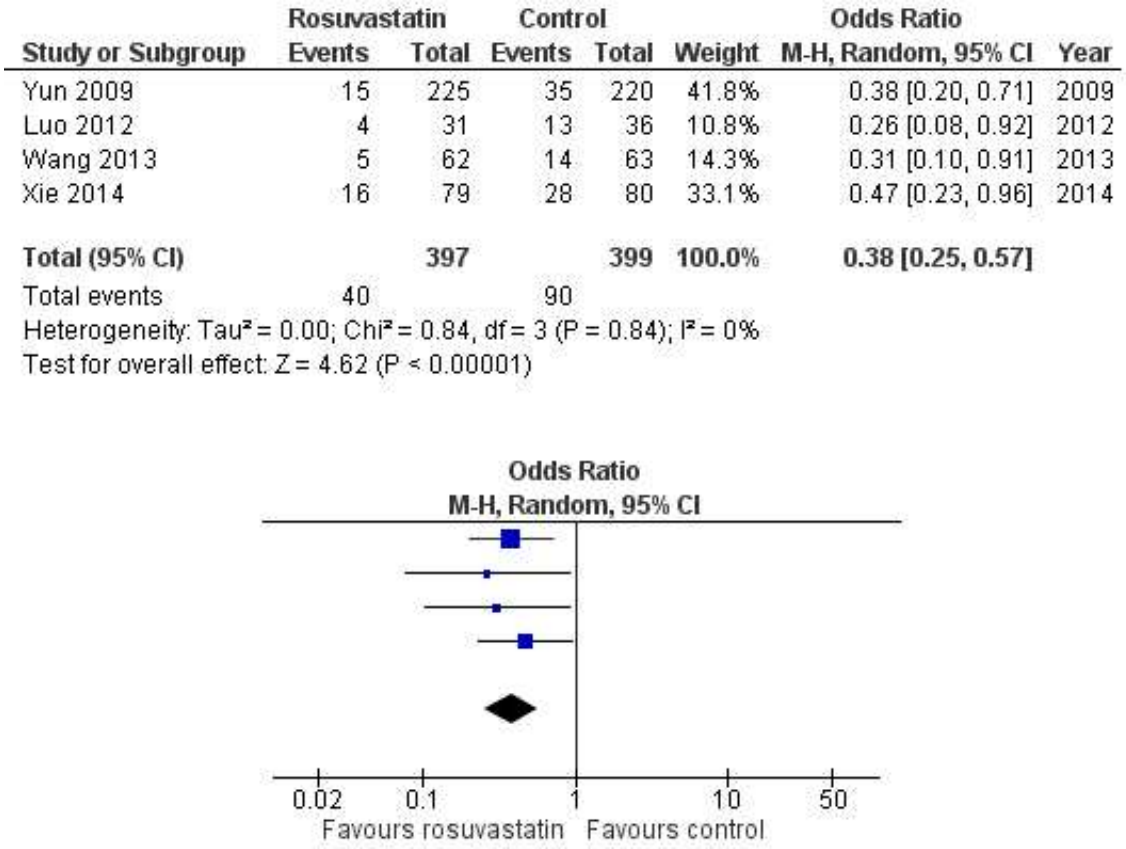

Figure 13. Odds ratio (OR) for the likelihood of experiencing MACCE at 30 days in NSTE-ACS undergoing PCI if early high-dose rosuvastatin loading was used *vs.* controls

5. DISCUSSION

The results of this meta-analysis derived from randomized data show that high-dose rosuvastatin loading, compared to no rosuvastatin loading or placebo, before PCI in statin-naive patients with NSTE-ACS was associated with a significant reduction in the likelihood of short-term adverse events such as major adverse cerebrovascular and cardiovascular events and recurrent myocardial infarctions during the 30-day follow-up. This intervention, however, did not impact all-cause mortality. Our findings were further reinforced when similar results were obtained by including only studies in which all patients with NSTE-ACS received PCI. Of note, both experimental and control cohorts were well-balanced in terms of baseline comorbidities and personal history of MI or PCI. Similarly, both cohorts followed the same post-PCI maintenance antithrombotic and lipid-lowering therapy and were generally well-matched in terms of cardiovascular pharmacotherapy received during the index hospitalization.

The timing of statin loading in current NSTE-ACS guidelines has not been explicitly stated. In both European and US-based guidelines, statins hold IA class of recommendation and are recommended to be initiated as early as possible, in the absence of contraindications and regardless of the baseline cholesterol levels (37, 39). It is unclear how early should *„as early as possible"* be. Furthermore, there is a scarcity of large randomized studies examining the use of rosuvastatin, as the most potent statin, in the setting of NSTE-ACS. In contrast, the role of atorvastatin, the most potent lipophilic statin, in STEMI has been well-documented and investigated. High-dose atorvastatin loading improved microvascular myocardial perfusion among STEMI patients undergoing PCI in the STATIN STEMI trial (Efficacy of High-Dose AtorvaSTATIN Loading Before Primary Percutaneous Coronary Intervention in ST-Elevation Myocardial Infarction), however, without impact on short-term mortality (71). On the other hand, the largest-to-date randomized trial on the use of atorvastatin in ACS – Statins Evaluation in Coronary Procedures and Revascularization (SECURE-PCI) showed that loading doses of atorvastatin in ACS and planned invasive management did not reduce MACE at 30 days (72) although prespecified subanalysis of this trial that examined only those patients that received PCI showed that periprocedural atorvastatin loading significantly reduced the relative risk of MACE in STEMI by 41% while this effect was not significant in NSTE-ACS population (73). Interestingly, this beneficial effect of atorvastatin was consistent and preserved regardless of the timing of atorvastatin administration, even if it was administered only within 2 hours before PCI. Thus, the evidence base on the early use of atorvastatin in ACS, particularly in STEMI is robust, while this is not the case with rosuvastatin, especially in the NSTE-ACS setting.

In the present meta-analysis, five randomized trials cumulatively enrolled 1300 patients with NSTE-ACS accruing a total of 139 adverse events registered during the 30-day follow-up

(44 events in rosuvastatin loading cohort *vs.* 95 events in the control cohort). The first month after ACS, as well as in NSTE-ACS, is also regarded as *„vulnerable period"* in which intense antithrombotic and lipid-lowering treatment is mandatory in patients that underwent PCI to avoid post-procedural complications such as stent thrombosis, target lesion revascularization failure, recurrent MI, arrhythmias and possibly death. A large study from the US registry showed that the death rate within the 30-days post PCI is ≈2% and 58% of these deaths were attributed to cardiac while 42% to non-cardiac causes (74). Furthermore, less than half of 30-day deaths were attributed to PCI-related complications. Similarly, the rate of unplanned 30-day readmission after PCI was 7.2% in the large United States Nationwide Readmissions Database (75). According to our data, 8 all-cause deaths were registered in 1300 patients with NSTE-ACS during the 30-day follow-up thus showing that the event rate of death in this population during this period was very low (0.6%). Likely, the minuscule rate of death events and overall limited sample size of the whole population did not allow for the detection of potential mortality benefits conferred by high-dose rosuvastatin loading in NSTE-ACS. This could partially explain the result of why rosuvastatin did not impact mortality outcomes in this study. On the other hand, MACCE events were dominantly driven by a much higher incidence of recurrent MIs that contributed to 119 out of 139 events (86% of all adverse events). In this aspect, rosuvastatin loading was associated with a robust and highly significant reduction in the likelihood of recurrent MI post-PCI as it halved rates of MI at 30 days (56% reduction in the likelihood of MI).

The early use of high-dose rosuvastatin prior to PCI in NSTE-ACS patients, such as examined in this thesis, might be of large importance for several reasons.

First, many patients with NSTE-ACS, despite not meeting „STEMI criteria" will be discovered to have severe coronary artery disease as about one-third of all NSTEMI patients will have a fully occluded coronary artery at the time of presentation (47). The problem with this notion in clinical practice is that these patients, although at high risk of mortality and poor outcomes, might not be managed in a timely fashion or similar enough as patients managed through STEMI-like pathway although they essentialy share the same prognosis.

Secondly, between 40% to even up to 70% of all NSTEMI cases will be complicated by the finding of multivessel coronary disease when angiography is performed (76) and this should not come as a surprise given the high presence of comorbidity burden in this population (*e.g.* smoking, arterial hypertension, dyslipidemia, diabetes mellitus, etc.). A similar finding was confirmed in this thesis as the presence of multivessel disease in included NSTE-ACS trials ranged from an average of 34% to even 68% while one-quarter of patients had diabetes mellitus

with more than one-half having documented arterial hypertension. Therefore, a significant proportion of NSTE-ACS will have a high-risk disease profile and might be predisposed to poor outcomes in a short term.

Thirdly, according to the latest practice guidelines, the concomitant antithrombotic management of NSTE-ACS should not be fully executed until coronary angiography is performed and the P_2Y_{12} inhibitor should be only administered after this step has been performed. Not giving such a drug during the early unstable phase of the NSTE-ACS event might even make the role of high-dose rosuvastatin loading even more important in this population concerning the provision of early cardioprotection during the index event. Furthermore, under the assumption that all ACS-oriented therapies work synergistically and complement each other, rosuvastatin loading might be an important piece of the puzzle if we embrace the concept that the whole is greater than the sum of the parts in the ACS.

Finally, from the pharmacological standpoint, rosuvastatin was associated with a greater reduction in systemic and microvascular inflammation compared to atorvastatin in patients with ACS and this might likely translate to better clinical outcomes (77), however, exact mechanistic pathways supporting such causality are not elucidated. It is well-known, however, that mitigating systemic inflammatory response in ACS is related to improved clinical outcomes since hs-CRP levels tightly correlate to prognosis in the ACS population as they predict new MACE and cardiovascular and all-cause mortality events (78). High-dose rosuvastatin was also associated with delayed ventricular remodeling, inhibition of the malignant arrhythmogenic remodeling of the heart, and improvement in systolic function among patients with ACS (79). In Statin Contrast-Induced Nephropathy Prevention (PRATO-ACS) trial, high-dose rosuvastatin was administered during admission to statin-naive patients with NSTE-ACS who were scheduled for early invasive approach and this resulted in the significant reduction of contrast-induced acute kidney injury and improved short-term outcomes (80).

Taken together, due to robust lipid-lowering, anti-inflammatory, anti-remodeling, and nephroprotective effects elicited by rosuvastatin it remains biologically plausible as a concept that high-dose rosuvastatin loading likely contributes to cardioprotection and improved short-term clinical outcomes among patients with NSTE-ACS undergoing PCI.

Limitations of the present meta-analysis are that included trials were generally of smaller size and the number of certain events of interest such as all-cause mortality were too low to detect a meaningful difference between experimental and control cohorts and this might have been mitigated if large-scale studies were available. Furthermore, the majority of trials were conducted in China thus possibly contributing to geographical bias. Likewise, no grey

literature search was performed for studies not included in large public databases. For these reasons, there is a possibility that these results might not be entirely generalizable to other populations and healthcare systems. On the other hand, presented results were based on data exhibiting a low degree of heterogeneity, and conservative effect estimates were employed by using random-effects rather than fixed-effects method. Future randomized studies should be designed with larger patient recruitment investigating the early use of high-dose rosuvastatin in NSTE-ACS and STEMI for which the largest evidence gap exists.

In conclusion, our data support the notion that early high-dose rosuvastatin loading should be initiated immediately at the first medical contact with a patient diagnosed with NSTE-ACS if no contraindications exist and early invasive management is planned. This intervention seems to provide a large benefit-to-harm ratio concerning short-term outcomes following PCI, dominantly by halving the likelihood of recurrent myocardial infarction with a very low rate of side-effects and no significant safety concerns during the short-term follow-up.

6. CONCLUSIONS

Based on the quantitative and meta-analytic synthesis of obtained data from randomized controlled trials investigating the use of early high-dose rosuvastatin loading in statin-naive patients with NSTE-ACS, we can conclude the following:

1. Early high-dose rosuvastatin loading before scheduled PCI in patients with NSTE-ACS was significantly associated with a 59% reduction in the likelihood of MACCE at 30 days, compared to no statin loading strategy or placebo.
2. Early high dose rosuvastatin loading before scheduled PCI in patients with NSTE-ACS was significantly associated with a 56% reduction in the likelihood of myocardial infarction at 30 days, compared to no statin loading strategy or placebo.
3. Early high-dose rosuvastatin loading before scheduled PCI in patients with NSTE-ACS does not seem to confer any mortality benefits and was similar to placebo or no loading strategy concerning this outcome.
4. Among patients with NSTE-ACS of whom all received PCI, early high-dose rosuvastatin loading *vs.* no loading strategy or placebo before PCI was significantly associated with a 62% reduction in the likelihood of MACCE at 30 days.
5. Taken together, early high-dose rosuvastatin loading in NSTE-ACS patients scheduled to undergo PCI seems to be an effective strategy that substantially reduces adverse short-term outcomes such as MACCE and myocardial infarction at 30 days, compared to no statin loading strategy or placebo, however, this intervention did not reduce all-cause mortality.

7. REFERENCES

1. Endo A. A historical perspective on the discovery of statins. Proc Jpn Acad Ser B Phys Biol Sci. 2010;86:484–93.
2. Hajar R. Statins: Past and present. Heart Views. 2011;12:121.
3. Sirtori CR. The pharmacology of statins. Pharmacol Res. 2014;88:3–11.
4. M MAS, I S, J S, S H, S T. Effects of different types of statins on lipid profile: a perspective on asians. International Journal of Endocrinology and Metabolism. 2017;e43319.
5. Schachter M. Chemical, pharmacokinetic and pharmacodynamic properties of statins: an update. Fundam Clin Pharmacol. 2005;19:117–25.
6. Lennernäs H, Fager G. Pharmacodynamics and pharmacokinetics of the HMG-CoA reductase inhibitors. Similarities and differences: Similarities and Differences. Clin Pharmacokinet. 1997;32:403–25.
7. Mangravite LM, Thorn CF, Krauss RM. Clinical implications of pharmacogenomics of statin treatment. Pharmacogenomics J. 2006;6:360–74.
8. Stein EA MD, PhD, Lane M MS, Laskarzewski P PhD. Comparison of statins in hypertriglyceridemia. Am J Cardiol. 1998;81:66B-69B.
9. Wierzbicki AS, Poston R, Ferro A. The lipid and non-lipid effects of statins. Pharmacol Ther. 2003;99:95–112.
10. Förstermann U, Li H. Therapeutic effect of enhancing endothelial nitric oxide synthase (eNOS) expression and preventing eNOS uncoupling: Prevention of eNOS uncoupling. Br J Pharmacol. 2011;164:213–23.
11. Liao JK, Laufs U. Pleiotropic effects of statins. Annu Rev Pharmacol Toxicol. 2005;45:89–118.
12. Ridker PM, Danielson E, Fonseca FAH, Genest J, Gotto AM Jr, Kastelein JJP et al. Rosuvastatin to prevent vascular events in men and women with elevated C-reactive protein. N Engl J Med. 2008;359:2195–207.
13. Kinlay S, Schwartz GG, Olsson AG, Rifai N, Leslie SJ, Sasiela WJ et al. High-dose atorvastatin enhances the decline in inflammatory markers in patients with acute coronary syndromes in the MIRACL study. Circulation. 2003;108:1560–6.
14. Schwartz GG, Olsson AG, Ezekowitz MD, Ganz P, Oliver MF, Waters D et al. Effects of atorvastatin on early recurrent ischemic events in acute coronary syndromes: the MIRACL study: a randomized controlled trial. JAMA. 2001;285:1711–8.

15. Erl W. Statin-induced vascular smooth muscle cell apoptosis: a possible role in the prevention of restenosis? Curr Drug Targets Cardiovasc Haematol Disord. 2005;5:135–44.
16. Liao JK. Isoprenoids as mediators of the biological effects of statins. J Clin Invest. 2002;110:285–8.
17. Chan K-C, Wang C-J, Ho H-H, Chen H-M, Huang C-N. Simvastatin inhibits cell cycle progression in glucose-stimulated proliferation of aortic vascular smooth muscle cells by up-regulating cyclin dependent kinase inhibitors and p53. Pharmacol Res. 2008;58:247–56.
18. Fouty BW, Rodman DM. Mevastatin can cause G1 arrest and induce apoptosis in pulmonary artery smooth muscle cells through a p27Kip1-independent pathway. Circ Res. 2003;92:501–9.
19. Suresh S, Narayana S, Jayakumar P, Sudhakar U, Pramod V. Evaluation of anti-inflammatory effect of statins in chronic periodontitis. Indian J Pharmacol. 2013;45:391–4.
20. Mendieta G, Ben-Aicha S, Gutiérrez M, Casani L, Aržanauskaitė M, Carreras F et al. Intravenous statin administration during myocardial infarction compared with oral post-infarct administration. J Am Coll Cardiol. 2020;75:1386–402.
21. Mendieta G, Ben-Aicha S, Casani L, Badimon L, Sabate M, Vilahur G. Molecular pathways involved in the cardioprotective effects of intravenous statin administration during ischemia. Basic Res Cardiol. 2019;115:2.
22. Korani S, Bahrami S, Korani M, Banach M, Johnston TP, Sahebkar A. Parenteral systems for statin delivery: a review. Lipids Health Dis. 2019;18:193.
23. Pinal-Fernandez I, Casal-Dominguez M, Mammen AL. Statins: pros and cons. Med Clín (Engl Ed). 2018;150:398–402.
24. Ramkumar S, Wilson A. Statins: Review of safety and potential side effects. Heart Lung Circ. 2015;24:S190.
25. Sharma P, Timilsina B, Adhikari J, Parajuli P, Dhital R, Tachamo N. Statin-induced necrotizing autoimmune myopathy: an extremely rare adverse effect from statin use. J Community Hosp Intern Med Perspect. 2019;9:503–6.
26. Selva-O'Callaghan A, Alvarado-Cardenas M, Pinal-Fernández I, Trallero-Araguás E, Milisenda JC, Martínez MÁ et al. Statin-induced myalgia and myositis: an update on pathogenesis and clinical recommendations. Expert Rev Clin Immunol. 2018;14:215–24.

27. Danaei G, García Rodríguez LA, Fernandez Cantero O, Hernán MA. Statins and risk of diabetes: an analysis of electronic medical records to evaluate possible bias due to differential survival. Diabetes Care. 2013;36:1236–40.
28. Cholesterol Treatment Trialists' (CTT) Collaborators. The effects of lowering LDL cholesterol with statin therapy in people at low risk of vascular disease: meta-analysis of individual data from 27 randomised trials. Lancet. 2012;380:581–90.
29. Vergouwen MDI, de Haan RJ, Vermeulen M, Roos YBWEM. Statin treatment and the occurrence of hemorrhagic stroke in patients with a history of cerebrovascular disease. Stroke. 2008;39:497–502.
30. Katsanos AH, Lioutas V-A, Charidimou A, Catanese L, Ng KKH, Perera K et al. Statin treatment and cerebral microbleeds: A systematic review and meta-analysis. J Neurol Sci. 2021;420:117224.
31. Cai T, Abel L, Langford O, Monaghan G, Aronson JK, Stevens RJ et al. Associations between statins and adverse events in primary prevention of cardiovascular disease: systematic review with pairwise, network, and dose-response meta-analyses. BMJ. 2021;374:n1537.
32. Sabatine MS. PCSK9 inhibitors: clinical evidence and implementation. Nat Rev Cardiol. 2019;16:155–65.
33. Pasta A, Cremonini AL, Pisciotta L, Buscaglia A, Porto I, Barra F et al. PCSK9 inhibitors for treating hypercholesterolemia. Expert Opin Pharmacother. 2020;21:353–63.
34. Handelsman Y, Lepor NE. PCSK9 inhibitors in lipid management of patients with diabetes mellitus and high cardiovascular risk: A review. J Am Heart Assoc. 2018;7: e008953.
35. Karagiannis AD, Liu M, Toth PP, Zhao S, Agrawal DK, Libby P et al. Pleiotropic anti-atherosclerotic effects of PCSK9 InhibitorsFrom molecular biology to clinical translation. Curr Atheroscler Rep. 2018;20:20.
36. Ibanez B, James S, Agewall S, Antunes MJ, Bucciarelli-Ducci C, Bueno H et al. 2017 ESC Guidelines for the management of acute myocardial infarction in patients presenting with ST-segment elevation: The Task Force for the management of acute myocardial infarction in patients presenting with ST-segment elevation of the European Society of Cardiology (ESC). Eur Heart J. 2018;39:119–77.
37. Collet J-P, Thiele H, Barbato E, Barthélémy O, Bauersachs J, Bhatt DL et al. 2020 ESC Guidelines for the management of acute coronary syndromes in patients presenting without persistent ST-segment elevation. Eur Heart J. 2021;42:1289–367.

38. Mirza AJ, Taha AY, Khdhir BR. Risk factors for acute coronary syndrome in patients below the age of 40 years. Egypt Heart J. 2018;70:233–5.
39. Amsterdam EA, Wenger NK, Brindis RG, Casey DE Jr, Ganiats TG, Holmes DR Jr et al. 2014 AHA/ACC guideline for the management of patients with non-ST-elevation acute coronary syndromes: A report of the American college of cardiology/American heart association task force on practice guidelines. J Am Coll Cardiol. 2014;64:e139–228.
40. Novak K, Vrdoljak D, Jelaska I, Borovac JA. Sex-specific differences in risk factors for in-hospital mortality and complications in patients with acute coronary syndromes: An observational cohort study. Wien Klin Wochenschr. 2017;129:233–42.
41. Kumric M, Borovac JA, Martinovic D, Ticinovic Kurir T, Bozic J. Circulating biomarkers reflecting destabilization mechanisms of coronary artery plaques: Are we looking for the impossible? Biomolecules. 2021;11:881.
42. Waterbury TM, Tarantini G, Vogel B, Mehran R, Gersh BJ, Gulati R. Non-atherosclerotic causes of acute coronary syndromes. Nat Rev Cardiol. 2020;17:229–41.
43. Tzimas G, Antiochos P, Monney P, Eeckhout E, Meier D, Fournier S et al. Atypical electrocardiographic presentations in need of primary percutaneous coronary intervention. Am J Cardiol. 2019;124:1305–14.
44. Borovac JA, Orsolic A, Miric D, Glavas D. The use of Smith-modified Sgarbossa criteria to diagnose an extensive anterior acute myocardial infarction in a patient presenting with a left bundle branch block. J Electrocardiol. 2021;64:80–4.
45. Macias M, Peachey J, Mattu A, Brady WJ. The electrocardiogram in the ACS patient: high-risk electrocardiographic presentations lacking anatomically oriented ST-segment elevation. Am J Emerg Med. 2016;34:611–7.
46. Lawner BJ, Nable JV, Mattu A. Novel patterns of ischemia and STEMI equivalents. Cardiol Clin. 2012;30:591–9.
47. Tziakas D, Chalikias G, Al-Lamee R, Kaski JC. Total coronary occlusion in non ST elevation myocardial infarction: Time to change our practice? Int J Cardiol. 2021;329:1–8.
48. Aslanger EK, Meyers HP, Bracey A, Smith SW. The STEMI/NonSTEMI Dichotomy needs to be replaced by Occlusion MI vs. Non-Occlusion MI. Int J Cardiol. 2021;330:15.

49. Pendell Meyers H, Bracey A, Lee D, Lichtenheld A, Li WJ, Singer DD, et al. Accuracy of OMI ECG findings versus STEMI criteria for diagnosis of acute coronary occlusion myocardial infarction. Int J Cardiol Heart Vasc. 2021;33(100767):100767.
50. Aslanger EK, Meyers PH, Smith SW. STEMI: A transitional fossil in MI classification? J Electrocardiol. 2021;65:163–9.
51. DeVon HA, Mirzaei S, Zègre-Hemsey J. Typical and atypical symptoms of acute coronary syndrome: Time to retire the terms? J Am Heart Assoc. 2020;9:e015539.
52. Chang H, Min JK, Rao SV, Patel MR, Simonetti OP, Ambrosio G et al. Non-ST-segment elevation acute coronary syndromes: targeted imaging to refine upstream risk stratification. Circ Cardiovasc Imaging. 2012;5:536–46.
53. Ayad SW, El Zawawy TH, Lotfy MI, Naguib AM, El Amrawy AM. Incidence and impact of totally occluded culprit coronary artery in patients with non-ST segment elevation myocardial infarction acute coronary syndrome. Egypt Heart J. 2021;73:36.
54. Tziakas D, Chalikias G, Al-Lamee R, Kaski JC. Non ST-elevation myocardial infarction (NSTEMI) patients with total coronary artery occlusion: More than meets the eye. Int J Cardiol. 2021;333:52.
55. Mitsis A, Gragnano F. Myocardial infarction with and without ST-segment elevation: A contemporary reappraisal of similarities and differences. Curr Cardiol Rev. 2021;17:e230421189013.
56. Cohen M, Visveswaran G. Defining and managing patients with non-ST-elevation myocardial infarction: Sorting through type 1 vs other types. Clin Cardiol. 2020;43:242–50.
57. Niccoli G, Borovac JA, Vetrugno V, Camici PG, Crea F. Ivabradine in acute coronary syndromes: Protection beyond heart rate lowering. Int J Cardiol. 2017;236:107–12.
58. Werf F. Successes and future objectives in acute coronary syndrome. Nature Reviews Cardiology. 2014;11:624–625.
59. Basra SS, Virani SS, Paniagua D, Kar B, Jneid H. Acute coronary syndromes: unstable angina and non-ST elevation myocardial infarction. Cardiol Clin. 2014;32:353–70.
60. Knuuti J, Wijns W, Saraste A, Capodanno D, Barbato E, Funck-Brentano , et al. 2019 ESC Guidelines for the diagnosis and management of chronic coronary syndromes. Eur Heart J. 2020;41:407–77.

61. Cannon CP, McCabe CH, Belder R, Breen J, Braunwald E. Design of the pravastatin or atorvastatin evaluation and infection therapy (PROVE IT)-TIMI 22 trial. Am J Cardiol. 2002;89:860–1.
62. Schwartz GG. Effects of atorvastatin on early recurrent ischemic events in acute coronary SyndromesThe MIRACL study: A randomized controlled trial. JAMA. 2001;285:1711.
63. Jung L-Y, Lee S-R, Jung J-M, Kim Y-S, Lee S-H, Rhee K-S et al. Rosuvastatin reduces blood viscosity in patients with acute coronary syndrome. Korean Circ J. 2016;46:147–53.
64. Sexton TR, Wallace EL, Macaulay TE, Charnigo RJ, Evangelista V, Campbell CL et al. The effect of rosuvastatin on thromboinflammation in the setting of acute coronary syndrome. J Thromb Thrombolysis. 2015;39:186–95.
65. Link A, Ayadhi T, Böhm M, Nickenig G. Rapid immunomodulation by rosuvastatin in patients with acute coronary syndrome. Eur Heart J. 2006;27:2945–55.
66. Tropeano F, Leoncini M, Toso A, Maioli M, Dabizzi L, Biagini D et al. Impact of rosuvastatin in contrast-induced acute kidney injury in the elderly: Post hoc analysis of the PRATO-ACS trial. J Cardiovasc Pharmacol Ther. 2016;21:159–66.
67. Wang Z, Dai H, Xing M, Yu Z, Lin X, Wang S et al. Effect of a single high loading dose of rosuvastatin on percutaneous coronary intervention for acute coronary syndromes. J Cardiovasc Pharmacol Ther. 2013;18:327–33.
68. Gao Y, Jia Z-M, Sun Y-J, Zhang Z-H, Ren L-N, Qi G-X. Effect of high-dose rosuvastatin loading before percutaneous coronary intervention in female patients with non-ST-segment elevation acute coronary syndrome. Chin Med J (Engl). 2012;125:2250–4.
69. Higgins JPT, Thomas J, Chandler J, Cumpston M, Li T, Page MJ, et al., editors. Cochrane handbook for systematic reviews of interventions. Wiley; 2019.
70. Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group. Preferred reporting items for systematic reviews and meta-analyses: The PRISMA statement. PLoS Med. 2009;6:e1000097.
71. Kim J-S, Kim J, Choi D, Lee CJ, Lee SH, Ko Y-G et al. Efficacy of high-dose atorvastatin loading before primary percutaneous coronary intervention in ST-segment elevation myocardial infarction. JACC Cardiovasc Interv. 2010;3:332–9.
72. Berwanger O, Santucci EV, de Barros e Silva PGM, Jesuíno I de A, Damiani LP, Barbosa LM et al. Effect of loading dose of atorvastatin prior to planned percutaneous coronary

intervention on major adverse cardiovascular events in acute coronary syndrome: The SECURE-PCI randomized clinical trial. JAMA. 2018;319:1331.

73. Lopes RD, de Barros e Silva PGM, de Andrade Jesuíno I, Santucci EV, Barbosa LM, Damiani LP et al. Timing of loading dose of atorvastatin in patients undergoing percutaneous coronary intervention for acute coronary syndromes: Insights from the SECURE-PCI randomized clinical trial. JAMA Cardiol. 2018;3:1113.
74. Aggarwal B, Ellis SG, Lincoff AM, Kapadia SR, Cacchione J, Raymond RE et al. Cause of death within 30 days of percutaneous coronary intervention in an era of mandatory outcome reporting. J Am Coll Cardiol. 2013;62:409–15.
75. Sykes R, Mohamed MO, Kwok CS, Mamas MA, Berry C. Percutaneous coronary intervention and 30-day unplanned readmission with chest pain in the United States (Nationwide Readmissions Database). Clin Cardiol. 2021;44:291–306.
76. Baumann AAW, Mishra A, Worthley MI, Nelson AJ, Psaltis PJ. Management of multivessel coronary artery disease in patients with non-ST-elevation myocardial infarction: a complex path to precision medicine. Ther Adv Chronic Dis. 2020;11:2040622320938527.
77. Kumar B, Shah MAA, Kumar R, Kumar J, Memon A. Comparison of atorvastatin and rosuvastatin in reduction of inflammatory biomarkers in patients with acute coronary syndrome. Cureus. 2019;11:e4898.
78. Mani P, Puri R, Schwartz GG, Nissen SE, Shao M, Kastelein JJP et al. Association of initial and serial C-reactive protein levels with adverse cardiovascular events and death after acute coronary syndrome: A secondary analysis of the VISTA-16 trial: A secondary analysis of the VISTA-16 trial. JAMA Cardiol. 2019;:314–20.
79. Guo J, Zhang W-Z, Zhao Q, Wo J-S, Cai S-L. Study on the effect of different doses of rosuvastatin on ventricular remodeling in patients with acute coronary syndrome after emergency percutaneous coronary intervention. Eur Rev Med Pharmacol Sci. 2017;21:4457–63.
80. Leoncini M, Toso A, Maioli M, Tropeano F, Villani S, Bellandi F. Early high-dose rosuvastatin for contrast-induced nephropathy prevention in acute coronary syndrome: Results from the PRATO-ACS Study (Protective Effect of Rosuvastatin and Antiplatelet Therapy On contrast-induced acute kidney injury and myocardial damage in patients with Acute Coronary Syndrome). J Am Coll Cardiol. 2014;63:71–9.

8. SUMMARY

Objectives: The present study aimed to examine the impact of early high-dose rosuvastatin loading (pretreatment) *vs.* no loading or placebo on short-term adverse outcomes at 30 days in statin-naive patients with acute coronary syndromes without persistent ST-segment elevation (NSTE-ACS) undergoing percutaneous coronary intervention (PCI).

Patients and methods: Quantitative analysis and meta-analysis were performed by including five randomized controlled trials (RCTs) that examined the use of rosuvastatin in NSTE-ACS patients undergoing PCI. Primary outcomes of interest were outcomes at 30 days including major adverse cerebrovascular and cardiovascular events (MACCE), myocardial infarction (MI), and all-cause death. Secondarily, we sought to determine the impact of the same intervention on MACCE in patients with NSTE-ACS of whom all received PCI. Odds ratio (OR) with 95% confidence intervals (95% CI) was used as the main summary measure while a random-effects model with the Mantel-Haenszel method was used for the meta-analysis.

Results: A total of 5 RCTs enrolling 1300 patients contributed to observed effect estimates. More than two-thirds of patients were male with a mean age of 63±11 years. Both rosuvastatin loading and control cohorts were generally well-balanced concerning baseline comorbidities, angiographic disease burden, and concomitant cardiovascular pharmacotherapy. Early high-dose rosuvastatin loading, compared to no loading or placebo, was associated with a significant reduction in the likelihood of MACCE (OR 0.41, 95% CI 0.27-0.60; $P<0.001$) and myocardial infarction (OR 0.44, 95% CI 0.29-0.67; $P<0.001$) without significant impact in reduction of all-cause mortality (OR 0.44, 95% CI 0.09-2.03; $P=0.290$). Finally, in trials in which all NSTE-ACS patients received PCI, rosuvastatin loading was associated with a 62% reduction in the likelihood of MACCE at 30 days (OR 0.38, 95% CI 0.25-0.57; $P<0.001$).

Conclusion: Early high-dose rosuvastatin loading strategy was associated with a significant reduction of MACCE and recurrent MIs among statin-naive patients with NSTE-ACS undergoing PCI. Rosuvastatin loading did not affect short-term mortality in this setting.

9. CROATIAN SUMMARY

Naslov rada: Rana uporaba visoke doze rosuvastatina u bolesnika koji se prezentiraju s akutnim koronarnim sindromom bez perzistentne elevacije ST segmenta

Ciljevi: Glavni cilj ove studije je bio istražiti utjecaj strategije rane uporabe visokih doza rosuvastatina (pretretman ili engl. *loading*), u usporedbi strategije bez *loadinga* rosuvastatinom ili uporabom placeba, na kratkoročne nepovoljne ishode u bolesnika s akutnim koronarnim sindromom bez perzistentne elevacije ST segmenta (NSTE-ACS), a kod kojih je planirana perkutana koronarna intervencija (PCI).

Pacijenti i metode: Kvantitativna analiza i meta-analiza su izvršene uključivanjem pet randomiziranih kliničkih studija koje su istraživale ranu uporabu rosuvastatina u bolesnika sa NSTE-ACS, a u kojih je planirana PCI. Glavni ishodi od interesa su bili mjereni unutar 30 dana od revaskularizacije, a uključivali su velike nepovoljne cerebrovaskularne i kardiovaskularne događaje (MACCE), infarkt miokarda (MI) i smrt zbog svih uzroka. Sekundarno, istražena je povezanost navedene intervencije sa ishodom MACCE-a u studijama koje su uključile bolesnike sa NSTE-ACS, a koji su svi primili PCI. Omjer izgleda (OR) sa 95%-tnim intervalima pouzdanosti (95% CI) je korišten kao glavna mjera ishoda, a model s nasumičnim učincima i Mantel-Haenszel algoritmom je korišten za meta-analizu.

Rezultati: Analizirano je 5 randomiziranih kliničkih studija koje su uključile ukupno 1300 bolesnika. Više od dvije trećine bolesnika su bili muškarci, a prosječna dob bila je 63±11 godina. Eksperimentalna i kontrolna skupina su bile dobro ujednačene što se tiče komorbiditeta, angiografske težine koronarne bolesti i kardiovaskularne farmakoterapije. Rana uporaba visokih doza rosuvastatina, u usporedbi sa strategijom bez loadinga ili korištenjem placeba, bila je povezana sa značajno nižim izgledom za MACCE (OR 0,41, 95% CI 0,27-0,60; P<0,001), MI (OR 0,44, 95% CI 0,29-0,67; P<0,001), ali bez značajnog učinka na smanjenje smrti zbog svih uzroka (OR 0,44, 95% CI 0,09-2,03; P=0,290). Konačno, u studijama u kojima su svi bolesnici sa NSTE-ACS primili PCI, rana uporaba rosuvastatina je bila povezana sa značajnim 62%-tnim smanjenjem izgleda za MACCE unutar 30 dana (OR 0,38, 95% CI 0,25-0,57; P<0.001).

Zaključci: Rano korištenje visokih doza rosuvastatina je povezano sa značajnim smanjenjem velikih nepovoljnih cerebrovaskularnih i kardiovaskularnih događaja te infarkta miokarda tijekom 30-dnevnog perioda od revaskularizacije u populaciji bolesnika sa NSTE-ACS, a kod kojih je planirana PCI. Rano korištenje visokih doza rosuvastatina nije imalo značajnog učinka na smanjenje smrtnosti zbog svih uzroka u navedenoj skupini bolesnika.

10. CURRICULUM VITAE

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Borovac JA, **Leth-Olsen M**, Kumric M, D'Amario D, Schwarz K, Glavas D, Bozic J. Efficacy of high-dose atorvastatin or rosuvastatin loading in patients with acute coronary syndrome undergoing percutaneous coronary intervention: a meta-analysis of randomized controlled trials with GRADE qualification of available evidence. *Eur J Clin Pharmacol*. 2021. Online ahead of print. DOI: https://doi.org/10.1007/s00228-021-03196-9