Trial Designs

Impact of PCSK9 Inhibition on Coronary Atheroma Progression: Rationale and Design of GLAGOV (Global Assessment of Plaque Regression with a PCSK9 Antibody as Measured by Intravascular Ultrasound)

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RCT# NCT01813422

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Running title: PCSK9 inhibition and plaque progression-regression

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ABSTRACT

**Background:** Statin-mediated low-density lipoprotein cholesterol (LDL-C) lowering fails to prevent more than half of cardiovascular events in clinical trials. Serial plaque imaging studies have highlighted the benefits of aggressive LDL-C lowering, with plaque regression evident in up to two-thirds of patients with achieved LDL-C levels <70 mg/dL. Proprotein convertase subtilisin/kexin type 9 (PCSK9) inhibitors permit LDL-C-lowering by a further 54-75% in statin-treated patients. The impact of achieving very low LDL-C levels with PCSK9 inhibitors on coronary atherosclerosis has not been investigated.

**Aims:** To test the hypothesis that incremental LDL-C lowering with the PCSK9 inhibitor, evolocumab, will result in a significantly greater change from baseline in coronary atheroma volume than placebo in subjects receiving maximally tolerated statin therapy.

**Methods:** A phase 3, multi-center, double blind, randomized, placebo-controlled trial evaluating the impact of evolocumab on coronary atheroma volume as assessed by serial coronary intravascular ultrasound at baseline in patients undergoing a clinically-indicated coronary angiogram with angiographic evidence of coronary atheroma, and following 78-weeks of treatment. Subjects (n=968) were randomized 1:1 into 2 groups to either receive monthly evolocumab 420 mg or placebo subcutaneous injections.

**Conclusions:** The GLAGOV (Global Assessment of Plaque Regression with a PCSK9 Antibody as Measured by Intravascular Ultrasound) trial will explore whether greater degrees of plaque regression are achievable with ultra-high-intensity LDL-C lowering following combination statin-PCSK9 inhibitor therapy. GLAGOV will provide important mechanistic,
safety and efficacy data prior the eagerly anticipated clinical outcomes trials testing the PCSK9 inhibitor hypothesis (www.clinicaltrials.gov identifier NCT01813422).
INTRODUCTION

Animal, epidemiological and genetic data have established a causal role for low-density lipoprotein cholesterol (LDL-C) in atherosclerotic cardiovascular disease. Statins are widely used for LDL-C lowering, with unequivocal evidence of cardiovascular benefit in randomized clinical trials. More intensive statin therapy associates with greater reductions in atherosclerotic events and now represents the preferred strategy recommended by the ACC/AHA cholesterol lowering guidelines. In parallel, serial plaque imaging studies have demonstrated that high-intensity statins promote disease regression, further underscoring their benefits for patients with coronary artery disease.

The beneficial effects of statins, however, are limited by a considerable residual risk of cardiovascular events, statin intolerance and inertia in terms of dose maximization in clinical practice. These observations have stimulated interest in developing alternative lipid-modifying therapies. The incremental clinical benefit of adding ezetimibe to statin therapy has reaffirmed the LDL-C hypothesis and potential benefits of non-statin agents. The discovery of the role of proprotein convertase subtilisin/kexin type 9 (PCSK9) in LDL-C metabolism has led to the development of novel inhibitors with potent LDL-C lowering effects. Thus, PCSK9 inhibitors have the potential for profound cardiovascular benefit, particularly in high-risk patients.

Effects of lowering LDL-C on atheroma: findings from early generation imaging studies

For nearly 50 years, coronary angiography had been the preferred imaging modality for diagnosing and quantifying the extent of coronary artery disease. Clinical trials employing serial coronary angiography demonstrated direct relationships between achieved LDL-C levels and the progression of coronary lumen obstruction. Yet no large angiographic study demonstrated disease regression. Serial carotid B-mode ultrasonic imaging of intimal-medial thickness (cIMT)
represents a non-invasive means of assessing the potential anti-atherosclerotic effects of novel therapies, with evidence from population studies demonstrating associations between cIMT and cardiovascular events \(^{24,25}\). Serial cIMT trials provided the first robust evidence of the benefits of intensive versus moderate statin therapy on the artery wall \(^{26,27}\), with more recent studies extending these benefits to lower risk patients \(^{28}\). However there is significant interest for directly quantifying the volumetric extent of coronary atherosclerosis – the vascular bed responsible for the majority of cardiovascular events.

**Insights from serial coronary intravascular ultrasonography**

Intravascular ultrasound (IVUS) is a high-resolution, cross-sectional imaging modality enabling volumetric quantification of atheroma across long vascular segments. Serial IVUS evaluation permits assessment of the effect of novel therapies on coronary atherosclerosis \(^{29}\), demonstrating the benefits of intensive statin therapy on coronary plaque burden. The REVERSAL (Reversal of Atherosclerosis with Aggressive Lipid Lowering) trial compared the effects of atorvastatin 80 mg and pravastatin 40 mg on coronary atheroma progression \(^{15}\). While ongoing disease progression (ΔPAV from baseline: +1.6\%) was noted in the pravastatin group (achieved LDL-C 110 mg/dL), no significant change in disease burden (ΔPAV from baseline: -0.4\%) was evident in the atorvastatin group (achieved LDL-C 79 mg/dL) following 18-months. ASTEROID (A Study to Evaluate the Effect of Rosuvastatin on Intravascular Ultrasound-Derived Coronary Atheroma Burden) investigated the impact of high-intensity statin therapy with rosuvastatin 40 mg for 24 months in patients with coronary artery disease. With an achieved LDL-C of 61 mg/dL, ASTEROID demonstrated significant plaque regression (ΔPAV from baseline: -0.98\%), with evidence of reductions in plaque burden in the majority of individuals \(^{16}\). SATURN (Study of Coronary Atheroma by Intravascular Ultrasound: Effect of Rosuvasstatin versus Atorvastatin)
sought to directly compare the efficacy of two high-intensity statin regimens (rosuvastatin 40 mg vs. atorvastatin 80 mg) on plaque burden over 24-months. Achieving low LDL-C levels with rosuvastatin (63 mg/dL) and atorvastatin (70 mg/dL) associated with significant atheroma regression (rosuvastatin group: ΔPAV from baseline = -1.22%; atorvastatin group: ΔPAV from baseline = 0.99%) (Figure 1).

Radiofrequency (virtual histology, VH) analysis from SATURN patients revealed a serial reduction in the fibro-fatty plaque component, consistent with high-intensity statin-induced plaque delipidation, and an increase in plaque calcium. This was further supported by a pooled analysis of nearly 3,500 patients demonstrating pro-calcific effects of statins, especially following high-intensity statin therapy. These findings suggest that plaque delipidation is likely associated with fibrous healing, which ultimately calcifies. These composition changes are likely to reflect plaque stabilization, further highlighting the beneficial impact of potent LDL-C lowering on the arterial wall. Accumulating evidence has established the relationship between plaque burden, progression and cardiovascular events. Findings from serial coronary IVUS studies have proven to complement those of cardiovascular outcome trials involving the same agent. Accordingly, there continues to be interest in understanding the impact of novel therapies, not only on cardiovascular events, but also on the underlying biology within the arterial wall.

**Targeting proprotein convertase subtilisin/kexin type 9 for cholesterol lowering**

Circulating LDL particles are removed from the circulation primarily via their interaction with LDL receptors on the liver surface. Within the liver, the particle is removed and degraded, while the receptor shuttles back to the liver surface, enabling ongoing LDL removal from the circulation. Proprotein convertase subtilisin/kexin type 9 (PCSK9) binds to the LDL receptor,
facilitating its destruction within hepatocytes and preventing its recirculation to the hepatocyte surface. Mutations in the PCSK9 gene, later determined to be gain-of-function mutations, result in a form of autosomal dominant hypercholesterolemia, accompanied by markedly elevated LDL-C levels and premature atherosclerotic disease. In contrast, loss-of-function mutations of the PCSK9 gene associates with lifelong lower LDL-C levels, by up to 40% lower than those without the mutation, accompanied by a significantly lower risk of atherosclerotic events. In fact, individuals with 2 loss-of-function mutations involving the PCSK9 gene harbor extremely low LDL-C levels (<20 mg/dL), and seem not to display any deleterious effects as a result of this genotype-phenotype combination. Furthermore, the observation that statins, by virtue of lowering circulating and intrahepatocyte cholesterol levels, increase the expression of both LDL receptors and PCSK9 could explain the reason why an apparent ceiling effect of LDL-C lowering is observed with increasing statin doses. These mechanisms underscore why concomitant PCSK9 inhibition during statin therapy results in incremental LDL-C reductions compared with statin therapy alone.

Pre-clinical PCSK9 knockout models demonstrated 2- to 3-fold increments in the number of LDL receptors, accompanied by up to 50% lower LDL-C levels. These observations stimulated the exploration of approaches to either reduce PCSK9 levels or function, including the development of antisense oligonucleotides, peptide mimetics, LDL-R antagonists, and small molecule inhibitors. However, the development of parenteral monoclonal antibodies has emerged as the most clinically advanced form of PCSK9 inhibition to date, and these antibodies are now within the advanced stages of clinical testing, with 2 compounds (evolocumab and alirocumab) recently receiving FDA-approval, and evolocumab also receiving Canadian and European approval.
Clinical Development of Evolocumab

Evolocumab (Amgen, Thousand Oaks, CA) is a fully human IgG2 monoclonal antibody with high binding affinity for PCSK9. *In vitro* assays demonstrated that evolocumab protects the LDL receptor from PCSK9-mediated degradation and subsequently reverses PCSK9-mediated decreases in LDL particle uptake. Based on phase II dose-ranging studies, a 140 mg dose every 2 weeks (Q2W) or a 420 mg monthly (QM) were identified as the optimal dosing regimens to maximally reduce LDL-C and provide clinically equivalent time-averaged pharmacodynamic effects.\textsuperscript{45-49}

The efficacy and safety of evolocumab has been evaluated in more than 6,000 subjects to date.\textsuperscript{45-47, 49-54} Evolocumab consistently reduced LDL-C levels by 54-75\% compared with placebo and by 35-45\% compared with ezetimibe across a range of hyperlipidemic (non-familial and heterozygous familial) populations with cardiovascular disease or high risk patients. Significant improvements in other lipid parameters, including lipoprotein (a) [Lp(a)], were also observed. These lipid changes were consistent regardless of the therapeutic setting. In homozygous familial hypercholesterolemic subjects, evolocumab reduced LDL-C by approximately 30\% compared with placebo.\textsuperscript{54} suggesting that many of these patients have some form of functional LDL receptor. From a safety perspective, reported adverse events were similar in evolocumab and control groups across all studies. No relationships were observed between the subject incidence of adverse events and the dose or dose frequency. The overall incidence of serious adverse events leading to discontinuation of evolocumab was low and balanced between treatment and control groups. Assessment of muscle-related adverse events and abnormalities in creatine kinase and liver transaminases did not reveal a safety concern. Anti-evolocumab binding antibodies were rare; no neutralizing anti-evolocumab antibodies were
identified. Pooling of data from long term treated patients suggested potentially less cardiovascular events, and possibly more neurocognitive adverse events. The implications of these findings are being evaluated in the large clinical outcomes trial, FOURIER (ClinicalTrials.gov Identifier: NCT01764633)

METHODS

Study rationale and objectives

GLAGOV (Global Assessment of Plaque Regression with a PCSK9 Antibody as Measured by Intravascular Ultrasound; NCT01813422) will evaluate the effect of evolocumab on the change in burden of coronary atherosclerosis as measured by the IVUS-derived percent atheroma volume (PAV) in subjects undergoing a clinically indicated coronary angiogram with evidence of coronary artery disease who are receiving maximally tolerated statin therapy. The primary objective of GLAGOV is to test the hypothesis that LDL-C lowering with a monthly subcutaneous injection of evolocumab 420 mg for 78-weeks will result in a significantly greater change from baseline in PAV than placebo in subjects taking background statin therapy. The primary efficacy endpoint is the change in PAV by IVUS imaging, performed at baseline and at the end of the 78-week treatment period. Secondary endpoints include the percentage of patients demonstrating PAV regression (defined as any reduction from baseline), the nominal change in total atheroma volume (TAV) by IVUS imaging from baseline to 78-weeks, and the percentage of patients demonstrating TAV regression (defined as any reduction from baseline). Exploratory objectives include an assessment of the effect of evolocumab on coronary atheroma composition, an assessment of the effects of evolocumab on the change and percent change upon a range of lipoprotein parameters including non-high-density lipoprotein (non-HDL) cholesterol, apolipoprotein B (ApoB), total cholesterol/HDL-C ratio, ApoB/Apolipoprotein A-1 (ApoA1)
ratio, Lp(a), triglycerides (TG), very low-density lipoprotein cholesterol (VLDL-C), LDL-C, HDL-C, PCSK9 levels and high-sensitivity (hs) C-Reactive protein (CRP). The incidence of major adverse cardiovascular events will also be explored in the total patient cohort as a function of changes in IVUS and lipoprotein variables. In addition, prespecified exploratory analyses will include sex-based analysis of the effects of evolocumab on IVUS variables, the efficacy of evolocumab in patients with diabetes, as well as a range of metabolomic and lipidomic biomarker discovery analyses. GLAGOV is fully funded by Amgen Inc. No financial or editorial support was used to write this paper. The authors are solely responsible for the design and conduct of this study, all study analyses, the drafting and editing of the paper and its final contents.

Study population

A total of 968 patients aged ≥18 years with at least one >20% visual lumen stenosis within a native epicardial coronary artery, during a clinically indicated coronary angiogram, and meeting the entry criteria outlined in Tables 1 and 2 were randomized and received at least one dose of investigational product. Randomization of patients occurred between 9th May 2013 and 12th January 2015. The baseline clinical characteristics of patients enrolled in GLAGOV are summarized in Table 3. Similar to prior serial IVUS trials, the GLAGOV cohort is of a similar age with a preponderance of males, Caucasians and a high cardiovascular risk factor burden. Table 4 summarizes the baseline measures of lipid and inflammatory parameters in the overall cohort, in patients who had been treated with at least 4 weeks of a stable dose of atorvastatin.

Study design, drug administration and visit schedule

GLAGOV is a Phase III, multi-center, double blind, randomized, placebo-controlled study. Following informed consent, patients undergoing a clinically indicated coronary angiogram with
an acceptable IVUS, stable background statin therapy and a LDL-C level meeting inclusion
criterion (Table 1) were eligible for randomization. At time of randomization, subjects needed to
be on stable, optimized background statin therapy that was expected to be unchanged for the
duration of study participation (up to 18 months). Optimal statin therapy was defined as an
effective dose of atorvastatin (or equivalent titrated) to achieve target LDL-C (reduction or goal)
as defined by regional guidelines (i.e. atorvastatin: 20-80 mg; simvastatin: 40-80 mg;
rosuvastatin: 5-40 mg; pravastatin: 80 mg; lovastatin: 80 mg; pitavastatin: 4 mg). Where locally
approved, highly effective statin therapy, defined as at least atorvastatin 40 mg daily or
equivalent, was recommended. For subjects with LDL-C > 100 mg/dL (2.6 mmol/L) not
receiving highly-effective statin therapy (defined as atorvastatin ≥ 40 mg daily or equivalent), the
investigator needed to attest that higher dose statin therapy was not appropriate for that subject
(i.e. higher dose not tolerated, dose not available in that country, other significant clinical
concern). Subjects not on a maximally tolerated dose of atorvastatin (or equivalent), entered a
two to four week lipid stabilization period for initiation or titration of atorvastatin with a
maximum of one titration step to achieve optimization. However, the protocol allowed the
enrollment of statin intolerant patients (≤10% of total study population), as the effect of PSCK9-
induced LDL-C-lowering on changes in coronary atheroma volume in this special patient
subgroup was felt to be an important sub-investigation. No further alterations in lipid-lowering
therapies were permitted once subjects are randomized. Subjects were randomized in a 1:1
allocation into 2 treatment groups: evolocumab 420 mg SC monthly or placebo SC monthly.
Study visits will occur during screening, Day 1 (randomization) and weeks 4, 12, 24, 36, 52, 64,
76 (last dose of investigational product, IP) and 78 (time of final IVUS procedure). Subjects
initially self-administered study drug while under observation at the study site, thereafter
subjects self-administered at home, unless they chose to visit the study site for the study drug to be administered by site-based personnel. During each study visit, compliance, vital signs, adverse events and concomitant medications were recorded and laboratory tests were performed. The last study drug was to be administered at week 76, final IVUS performed at week 78 and end-of-study visit will occur at week 80. Sites will contact subjects at week 80 to assess for any potential adverse effects. Cardiovascular events (death, myocardial infarction, hospitalization for unstable angina, stroke, transient ischemic attack, hospitalization for heart failure) are adjudicated by an independent clinical events committee along with formal review of the accumulating data by an independent data monitoring committee. Subjects enrolled in GLAGOV were consented to provide additional blood samples for the possibility of future pharmacogenomic, metabolomic and lipidomic biomarker discovery analyses. The study design and treatment schema is summarized in Figure 2. Figure 3 broadly summarizes the number of patients screened and finally enrolled in GLAGOV.

Acquisition and analysis of intravascular ultrasound imaging

Intravascular ultrasonographic coronary imaging was acquired and analyzed in the same manner as undertaken in prior clinical trials assessing coronary atheroma progression-regression with IVUS \(^{15-17, 35, 38, 55-59}\). IVUS examinations were performed in the longest and least angulated coronary artery containing no lumen stenosis >50\% throughout a target segment of at least 40mm in length, had not undergone prior revascularization, and was not the culprit vessel responsible for a previous myocardial infarction. The target vessel for imaging had also to be considered unlikely to require revascularization during the course of the study. Following anticoagulation and administration of intracoronary nitroglycerin (100–300 mg), the imaging catheter was advanced as distally as possible within the vessel. Patients were imaged with
either a 40 MHz Atlantis SR Pro (Boston Scientific Inc., Maple Grove, MN, USA) or a 45 MHz Revolution (Volcano Corporation, San Diego, CA, USA) catheter. Continuous images were acquired while the catheter was withdrawn back to the aorta by a motor drive at a constant speed of 0.5mm per second. Images were burned on DVD and sent to the Atherosclerosis Imaging Core Laboratory at the Cleveland Clinic Coordinating Center for Clinical Research (C5R) for analysis.

Technicians blinded to the treatment status of the patient and the timing of each individual pullback, will perform the analysis. A matched arterial segment will be determined for measurement containing the same proximal and distal side branch. Cross-sectional images, spaced 1mm apart, will be selected for measurement. The leading edges of the lumen and the external elastic membrane (EEM) will be defined by manual planimetry (Figure 3). In each image, the plaque area will be calculated as the area between the two leading edges. Additional measurements recorded for each image include the maximum and minimum plaque thickness and the degree of calcification. Two measures of atheroma burden will be calculated for each patient. The PAV is calculated as the proportion of the EEM volume occupied by atherosclerotic plaque:

\[
P_{AV} = \frac{(EEM_{area} - Lumen_{area})}{EEM_{area}} \times 100
\]

The TAV is calculated as the summation of plaque areas in each measured cross sectional image within the segment, and subsequently normalized by the median number of images analyzed in the entire cohort to account for heterogeneity in segment length between subjects:

\[
T_{AV_{normalized}} = \frac{(EEM_{area} - Lumen_{area})}{\text{Number of Images in Pullback}} \times \text{Median number of images in cohort}
\]
This adjustment allows for each subject to be equally represented in the statistical analysis. In addition to absolute changes in measures of atheroma burden, the percentage of subjects that demonstrate plaque regression, defined as any reduction in PAV or TAV from baseline, will also be calculated.

Sample size determination and statistical analysis

The assumptions for sample size calculation are based on insights from SATURN\textsuperscript{17,60}, which indicates that every 1 mg/dL reduction in LDL-C is estimated to associate with change of 0.03026 in PAV at week 104. In GLAGOV, the assumed treatment effect is change of at least 0.706 in PAV at week 78, which is derived from an expected treatment effect of \textgreater 31 mg/dL reduction in LDL-C from baseline to week 78. The assumed common standard deviation (SD) is 2.9. Thus, assuming 25\% of randomized subjects will not be included in the primary analysis, the planned sample size of 950 subjects would provide approximately 712 subjects in the primary analysis to ensure 90\% power to test the study hypothesis. The sample size calculation was performed using a 2-sided t-test with a 0.05 significance level.

DISCUSSION

GLAGOV will be the first large-scale serial coronary imaging trial to formally test the extension of the LDL hypothesis outlined in Figure 1; that is if achieved LDL-C levels are further reduced to approximately 40 mg/dL, even further degrees of atheroma regression than those demonstrated in SATURN might be achieved within the coronary arterial wall. GLAGOV’s results will be available prior to the results of larger clinical outcomes trials testing the effects of various PCSK9 inhibitors. Although not a substitute for demonstrating reductions in clinical events, the consistency between serial coronary imaging trials and clinical trials evaluating the same compound, should provide the medical community with an early indication of the potential
benefits of adding PCSK9 inhibitors to statins in patients with coronary disease. Additional insights into the relationship between on-treatment lipoprotein and inflammatory parameters with changes in measures of plaque burden and composition (Virtual Histology) will be sought. Although GLAGOV is not formally powered to test between-group differences in clinical events, the availability of clinically adjudicated events will nevertheless provide further intriguing insights of the interaction between potent LDL-C lowering, arterial wall changes and atherosclerotic cardiovascular events.

Testing the GLAGOV hypothesis in a coronary artery disease population on maximally tolerated statins (harboring baseline LDL-C, HDL-C, triglycerides and hsCRP levels of 92.6 mg/dL, 46 mg/dL, 120 mg/dL and 1.6 mg/L respectively) will provide unique mechanistic information relevant to current clinical practice. Clinical guidelines stress the importance of optimizing statin intensity or achieving low LDL-C targets in the setting of established atherosclerotic disease\textsuperscript{14, 61}. However there remains a large gap between societal recommendations and contemporary clinical prescribing patterns\textsuperscript{62}. Furthermore, considerable residual cardiovascular risk is apparent in patients already prescribed statin therapies, or in those achieving lower LDL-C levels\textsuperscript{18}. Given the strength and consistency of the direct relationship between achieved LDL-C levels and cardiovascular risk, this creates opportunities for novel LDL-lowering therapies to impart incremental clinical benefits despite statin therapy\textsuperscript{21}.

Clinical trials employing serial coronary IVUS have been pivotal in outlining the anti-atherosclerotic effects of LDL-C lowering within the arterial wall, in particular highlighting the ability to halt atheroma progression and even induce regression following high-intensity statin therapies. These observations with serial coronary IVUS corroborate with lower rates of atherosclerotic events evident following intense LDL-C lowering. PCSK9 inhibitors represent a
new frontier of potent LDL-C lowering therapies. GLAGOV will explore whether greater
degrees of plaque regression can be achieved with ultra-potent LDL-C lowering following
combination statin-PCSK9 inhibitor therapy with evolocumab, compared with statin therapy
alone. These data will serve as an important prelude to the eagerly anticipated clinical outcomes
trials testing the PCSK9 inhibitor hypothesis.

TRANSPARENCY

Funding declaration

GLAGOV is sponsored by Amgen, Inc.

Author declarations

R.S., J.Y., H.K., S.W., and R.S are employees and stockholders of Amgen, Inc. L.C serves as a
consultant for Amgen and Sanofi-Aventis, S.E.N has received research support to perform
clinical trials through the Cleveland Clinic Coordinating Center for Clinical Research (C5R)
from Pfizer, AstraZeneca, Novartis, Roche, Daiichi-Sankyo, Takeda, Sanofi-Aventis,
Resverlogix and Eli Lilly; and is a consultant/advisor for many pharmaceutical companies but
requires them to donate all honoraria or consulting fees directly to charity so that he receives
neither income nor a tax deduction. J.J.P.K reports consulting fees from Amgen during the
conduct of the study; consulting fees from Cerenis, The Medicines Company, CSL Behring,
Regeneron, Eli Lilly, Esperion, AstraZeneca, Pronova, Boehringer Ingelheim, Catabasis,
Novartis, Merck, Isis Pharmaceuticals, Kowa, Sanofi, Gempshire, and Cymabay. W.K has
received consulting fees from Novartis, Pfizer, The Medicines Company, Amgen, AstraZeneca,
MSD, GSK, and Sanderling Ventures, honorarium (lectures) from AstraZeneca, Novartis, MSD,
Amgen and Actavis, research contracts with Abbott, Roche Diagnostics, Beckmann and
Singulex, and participates in several clinical trials (PEGASUS, LEADER, CANTOS and
FOURIER). C.M.B has received grant/research support from Abbott Diagnostic, Amarin,
Amgen, Eli Lilly, Esperion, Novartis, Pfizer, Otsuka, Regeneron, Roche Diagnostic, Sanofi-
Synthelabo, Takeda, NIH, the AHA, and the ADA (all significant, paid to his institution). He
also serves as a consultant to Abbott Diagnostics, Amarin, Amgen, Astra Zenea, Eli Lilly,
Esperion, Genzyme, Matinas BioPharma Inc, Merck, Novartis, Pfizer*, Regeneron, Roche and
Sanofi-Synthelabo. T.J.A reports research collaborations with Amgen, Merck and Pfizer. S.J.N
reported receiving research support from AstraZeneca, Novartis, Eli Lilly, Anthera, LipoScience,
Roche, and Resverlogix; and receiving honoraria from or serving as a consultant to AstraZeneca,
Roche, Esperion, Abbott, Pfizer, Merck, Takeda, LipoScience, Omthera, Novo-Nordisk, Sanofi-
Aventis, Atheronova, Anthera, CSL Behring, and Boehringer Ingelheim. All other authors have
no declarations to make.

Acknowledgements
No editorial or writing assistance in the preparation of this manuscript was sought.
REFERENCES


Figure legends

Figure 1
Median changes in percent atheroma volume (PAV) vs. average on-treatment LDL-C in serial coronary IVUS trials. Dotted blue line shows a projected outcome of the degree of plaque regression in those patients receiving evolocumab in GLAGOV. SATURN: The Study of Coronary Atheroma by Intravascular Ultrasound: Effect of Rosuvastatin vs. Atorvastatin; ASTEROID: A Study to Evaluate the Effect of Rosuvastatin on Intravascular Ultrasound-Derived Coronary Atheroma Burden; ILLUSTRATE: Investigation of Lipid Level Management Using Coronary Ultrasound To Assess Reduction of Atherosclerosis by CETP Inhibition and HDL Elevation; REVERSAL: Reversal of Atherosclerosis with Aggressive Lipid Lowering; STRADIVARIUS: Strategy To Reduce Atherosclerosis Development InVolving Administration of Rimonabant—the Intravascular Ultrasound Study; CAMELOT: Comparison of Amlodipine vs. Enalapril to Limit Occurrences of Thrombosis.

Figure 2
Study design and treatment schema of GLAGOV

Figure 3
Cohort diagram summarizing screen failures/exclusions

Figure 4
Cross-sectional image of a coronary artery imaged with intravascular ultrasound (a), depicting measurements obtained by Core Laboratory technicians (b). The inner circle signifies the leading edge of the lumen, whereas the outer circle represents the leading edge of the external elastic membrane (EEM).
Figure 5

A motorized pullback of the IVUS imaging catheter is performed beginning at a distal fiduciary branch (site C) and ending at a proximal branch (site A). The pathway of the catheter is illustrated in the angiogram in the top panel. The analysis technician locates the distal branch in the IVUS image, C, and obtains cross-sections every 1 mm until a proximal branch is reached, A. An intermediate cross-section is illustrated in B (adapted and modified from Nissen SE et al. JAMA 2003;290:2292-2300 with permission from the author).
Table 1: Inclusion criteria

- Men or women >18 years of age
- Clinically indicated coronary angiogram, evidence of coronary disease
- Stable statin dose for ≥4 weeks prior to screening*
- LDL-C criteria met within 4 weeks of screening visit, or if applicable, at the end of lipid stabilization period:
  - LDL-C ≥80 mg/dL, **OR**
  - LDL-C ≥60 but <80 mg/dL in the presence of 1 major or 3 minor risk factors

Major risk factors (1 required)

- Non-coronary atherosclerotic vascular disease as evidenced by documented peripheral arterial disease, documented abdominal aortic aneurysm, or documented cerebrovascular disease
- Documented myocardial infarction or hospitalization for unstable angina within the last 2 years
- Documented type 2 diabetes mellitus

Minor risk factors (3 required)

- Cigarette smoking (current)
- Hypertension (BP ≥140/90 mmHg or current use of anti-hypertensive medications)
- Low HDL-C (men: <40 mg/dL; women <50 mg/dL)
- Family history of premature coronary heart disease (1st degree male relative <55 years of age or 1st degree female relative <65 years of age)
- Age (men ≥50 years; women ≥55 years)
- hs-CRP ≥2 mg/L

BP=blood pressure; hs-CRP=high-sensitivity C-Reactive protein

*Subjects already taking statin therapy, regulatory-approved sustained-release niacin (e.g., Niaspan) or ezetimibe at initial screening must have been on a stable dose for at least 4 weeks prior to the lipid panel used for the screening LDL-C. Subjects not currently taking lipid-regulating therapy can be screened but must enter the study via a lipid stabilization period, OR, subjects who are intolerant to statins (limited to no more than approximately 10% of total planned enrollment) must meet specific statin intolerance entry criteria.
Table 2: Exclusion criteria

- Clinically significant heart disease which, in the opinion of the Principal Investigator, is likely to require coronary bypass surgery, percutaneous coronary intervention, cardiac transplantation, surgical valve repair and/or replacement during the course of the study.
- Heart failure of New York Heart Failure Association class III or IV or last known left ventricular ejection fraction <30%.
- Coronary artery bypass surgery <6 weeks prior to the qualifying IVUS.
- Cardiac arrhythmia within 3 months prior to randomization that is not controlled by medication.
- Uncontrolled hypertension at day 1, defined as a resting systolic blood pressure of ≥180 mm Hg.
- Triglyceride level >400mg/dL at screening.
- Type 1 diabetes mellitus or poorly controlled type 2 diabetes (HbA1c >9%) at screening.
- Thyroid stimulating hormone (TSH) < lower limit of normal (LLN) or TSH >1.5x upper limit of normal (ULN).
- Estimated glomerular filtration rate (eGFR) <30 ml/min/1.73m².
- Aspartate aminotransferase (AST) or alanine aminotransferase (ALT) >2x ULN.
- Creatine kinase >3x ULN.
- Use of cholesterylester transfer protein (CETP) inhibition treatment within 12 months prior to randomization.
- Any prior use of PCSK9 inhibitor therapy.
- Consumption of any of the following drugs for more than 2 weeks in the last 3 months prior to LDL-C screening: systemic cyclosporine, systemic steroids, isotretinoin.
- History of malignancy (except non-melanoma skin cancers, cervical in-situ carcinoma, breast ductal carcinoma in situ, or stage 1 prostate carcinoma).
- Known major active infection, or major hematologic, renal, metabolic, gastrointestinal or endocrine dysfunction.
- Baseline IVUS does not meet IVUS Core Lab technical standards.
- Female subjects cannot be pregnant or breast feeding. Premenopausal females must be willing to use at least 1 highly effective method of birth control during treatment and for an additional 15 weeks after the end of treatment.

HbA1c = glycosylated hemoglobin
Table 3: Clinical characteristics of patients

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Cohort (n=968)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)</td>
<td>59.8±9.2</td>
</tr>
<tr>
<td>Men (%)</td>
<td>698 (72.1)</td>
</tr>
<tr>
<td>Caucasians (%)</td>
<td>908 (93.8)</td>
</tr>
<tr>
<td>Prior myocardial infarction or hospitalization for UAP (within 2 yrs) (%)</td>
<td>280 (28.9)</td>
</tr>
<tr>
<td>Cerebrovascular disease (%)</td>
<td>42 (4.3)</td>
</tr>
<tr>
<td>Peripheral arterial disease (%)</td>
<td>21 (2.2)</td>
</tr>
<tr>
<td>Abdominal aortic aneurysm (%)</td>
<td>7 (0.7)</td>
</tr>
<tr>
<td>Type 2 diabetes mellitus (%)</td>
<td>199 (20.6)</td>
</tr>
<tr>
<td>Current cigarette smoking (%)</td>
<td>235 (24.3)</td>
</tr>
<tr>
<td>Hypertension (%)</td>
<td>795 (82.1)</td>
</tr>
<tr>
<td>Family history of premature coronary heart disease (%)</td>
<td>329 (34.0)</td>
</tr>
<tr>
<td>Low HDL-C (Men: &lt;40 mg/dL, women: &lt;50 mg/dL) (%)</td>
<td>321 (33.2)</td>
</tr>
<tr>
<td>hsCRP ≥2 mg/L (%)</td>
<td>395 (41.8)</td>
</tr>
<tr>
<td>Congestive cardiac failure (%)</td>
<td>123 (12.7)</td>
</tr>
<tr>
<td>Atrial fibrillation/flutter (%)</td>
<td>69 (7.1)</td>
</tr>
<tr>
<td>Statin intolerance (%)</td>
<td>27 (2.8)</td>
</tr>
</tbody>
</table>

Continuous data are presented as mean±SD and categorical data as N (%)
hsCRP=high-sensitivity C-Reactive protein
### Table 4: Baseline risk factor control

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total cholesterol (mg/dL)</td>
<td>166.2 (34.1)</td>
</tr>
<tr>
<td>LDL-C (mg/dL)</td>
<td>92.6 (27.2)</td>
</tr>
<tr>
<td>HDL-C (mg/dL)</td>
<td>46.0 (12.8)</td>
</tr>
<tr>
<td>Triglycerides (mg/dL)</td>
<td>120 (89, 166)</td>
</tr>
<tr>
<td>ApoA (mg/dL)</td>
<td>140.0 (25.6)</td>
</tr>
<tr>
<td>ApoB (mg/dL)</td>
<td>81.5 (20.0)</td>
</tr>
<tr>
<td>Lp(a) (nmol/L)</td>
<td>32 (12, 152)</td>
</tr>
<tr>
<td>hsCRP (mg/L)</td>
<td>1.61 (0.79, 3.38)</td>
</tr>
<tr>
<td>Glucose (mg/dL)</td>
<td>105.7 (27.4)</td>
</tr>
<tr>
<td>HbA1c (%)</td>
<td>5.9 (0.78)</td>
</tr>
<tr>
<td>Systolic blood pressure (mmHg)</td>
<td>130.5 (15.2)</td>
</tr>
<tr>
<td>Diastolic blood pressure (mmHg)</td>
<td>77.4 (9.8)</td>
</tr>
</tbody>
</table>

Data are presented as mean±SD or median (IQR)

HbA1c = glycosylated hemoglobin
Figure 1
Figure 2

Screening and placebo run-in period
1. Clinically indicated coronary angiogram
2. IVUS based on coronary angiogram results
3. Subcutaneous injection of 3 mL placebo

Up to 4 week lipid stabilization period
Assigned to background statin therapy

Randomization 1:1 to study drug

Placebo SC every month

Evolocumab 420 mg SC every month

Max. 6 weeks

2–4 weeks

Day 1 Week 4 Week 12 Week 24 Week 36 Week 52 Week 64 Week 76* Week 78* Week 80 EOS

Study visits:
Study drug was administered monthly, at home or in the clinic.

EOS, end of study; IVUS, intravascular ultrasound; SC, subcutaneously
*Last dose of study drug
*Last IVUS procedure
Figure 3

Screened (N = 2628)

Excluded (n = 1658)
- Consent (n = 22)
- Unavailability/unreliability (n = 123)
- IVUS and angiographic criteria (n = 725)
- Lipid/biochemical parameters (n = 417)
- Protocol exclusion criteria (n = 235)
- Pre-study statin dosing (n = 25)
- Other (n = 111)

Randomized (n = 970)

Received ≥ 1 dose of study drug (n = 968)
Figure 4
Figure 5