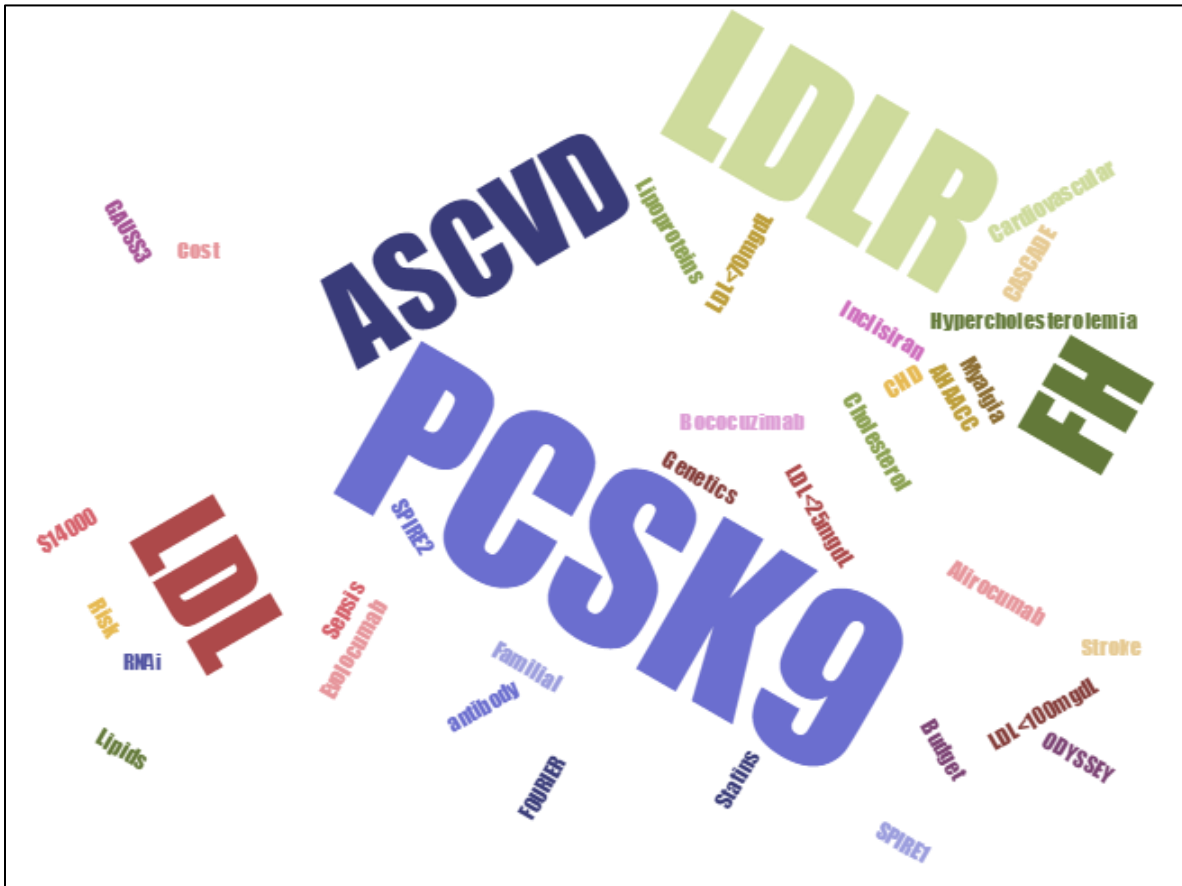


Lowering Cholesterol with PCSK9 Inhibitors



Zahid Ahmad, MD
Division of Nutrition and Metabolic Diseases

Internal Medicine Grand Rounds
March 31, 2017

This is to acknowledge that Dr. Ahmad's disclosures include research funding from NIH, FH Foundation, and Regeneron Pharmaceuticals; and honorarium (speaker bureaus, advisory boards) from Amgen, Inc., Regeneron Pharmaceuticals, and Sanofi.

UT Southwestern
Medical Center

Zahid Ahmad, MD, is an Assistant Professor in the Department of Internal Medicine, Division of Nutrition and Metabolic Disease, Center for Human Nutrition, UT Southwestern Medical center. His clinical, research, and teaching efforts focus on lipid and lipoprotein disorders. He attends lipid clinics at UT Southwestern (Aston building) and Parkland Health and Hospital Systems. These clinics provide care for patients with severe elevations in LDL-C, severe hypertriglyceridemia (triglycerides > 1000 mg/dL), and statin intolerance. His research efforts include NIH-funded projects to study the genetics of familial hypercholesterolemia in minorities and collaborations with biotech companies for early phase studies of new lipid-lowering drugs.

Purpose & Overview

The purpose of this presentation is to familiarize the audience with a new class of lipid lowering drugs: PCSK9 inhibitors.

The presentation will discuss the unmet needs (i.e. the large group of patients who require lipid lowering therapies in addition to statins), the history and current status of PCSK9 inhibitors, and the cardiovascular outcomes data from studies of PCSK9 inhibitors.

Objectives

At the conclusion of this lecture, the listener should be able to...

1. Recognize that a large number of patients with coronary heart disease fail to achieve their LDL-C goals.
2. Appreciate that Familial Hypercholesterolemia (FH) is underdiagnosed in the US, and FH patients require lipid lowering therapies in addition to statins.
3. Understand the similarities and differences between the two approved PCSK9 inhibitors
4. Know the cardiovascular outcomes data for PCSK9 inhibitors

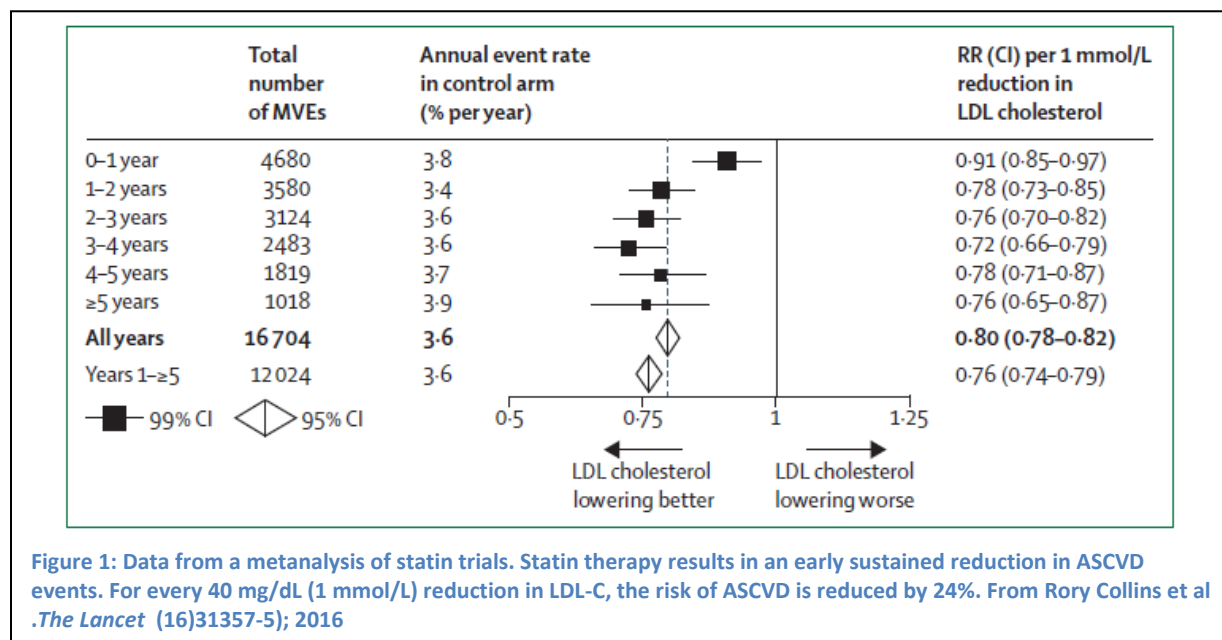
The protocol will discuss the unmet needs for patients with atherosclerotic cardiovascular disease (ASCVD) and familial hypercholesterolemia, the history and current status of PCSK9 inhibitors, and the cardiovascular outcomes data from studies of PCSK9 inhibitors.

Unmet Needs

Lipid lowering drugs are the second most commonly prescribed medication class in the United States (US) (1) with statins being the most commonly prescribed agent within this class. The widespread use of statins owes to overwhelming evidence of benefit and safety as well as awareness campaigns directed both at physicians and consumers.

High potency statins (such as atorvastatin 40-80 mg/daily or rosuvastatin 20-40 mg/daily) can reduce LDL-C by 50% or more. For every 40 mg/dL reduction in LDL-C (1 mmol/L), the risk of an atherosclerotic cardiovascular disease (ASCVD) event is reduced by 24% with statin therapy (Figure 1).

Several non-statin lipid lowering drugs also reduce LDL-C: niacin, bile-acid sequestrants, and ezetimibe. Of these, ezetimibe was recently shown to reduce the risk of atherosclerotic events by 6-7% when administered in addition to statin therapy (IMPROVE-IT study) (2).



Current Lipid-lowering Drugs are not sufficient to get all high-risk patients to goal.

Although statins effectively treat the vast majority of dyslipidemias, a few challenges remain. First, patients with known ASCVD often do not achieve goal LDL-C levels. In fact only 30% of patients with coronary heart disease (CHD) achieve LDL-C < 70 mg/dL (3).

Second, some subgroups at high risk for CHD - such as patients with severe elevations in cholesterol due to heterozygous familial hypercholesterolemia (FH) - present treatment challenges and often fail to reach their low-density-lipoprotein cholesterol (LDL-C) goals (4, 5). Additionally, such patients require high doses of statins placing them at increased risk for adverse events.

FH is a genetic disorder characterized by elevated low-density lipoprotein cholesterol (LDL-C), a markedly elevated risk of atherosclerotic cardiovascular disease (ASCVD), and a 50% chance of inheritance among offspring.(6) Recent epidemiological(7, 8) and genetic studies(9, 10) support a prevalence of FH of approximately 1 in 200 in the general community; if these prevalence figures hold true for the US population, as many as 1.5 million Americans may have FH, a substantially higher figure than suggested by earlier estimates.(11) Despite data confirming that prompt detection and treatment of FH reduces the risk of premature coronary heart disease (CHD) and death,(12) the majority of FH patients worldwide remain unidentified(8) and, of those diagnosed, most fail to receive appropriate treatment.(7)

We recently reported data from the FH Foundation's US FH patient registry. For patients seen at lipid centers across the US, we found that the mean treated LDL-C was 134 mg/dL (pretreatment 239 mg/dL). Only 26% of patients were able to achieve an LDL-C < 100 mg/dL (a typical goal for FH patients) and only 45% achieved a 50% LDL-C reduction (a goal advocated by the 2013 ACC/AHA Cholesterol Guidelines Committee). Our findings mirror those from other countries with nationwide screening programs to identify FH patients: only 15-33% of treated FH patients achieve their LDL-C goal despite being treated with statins (4, 13, 14).

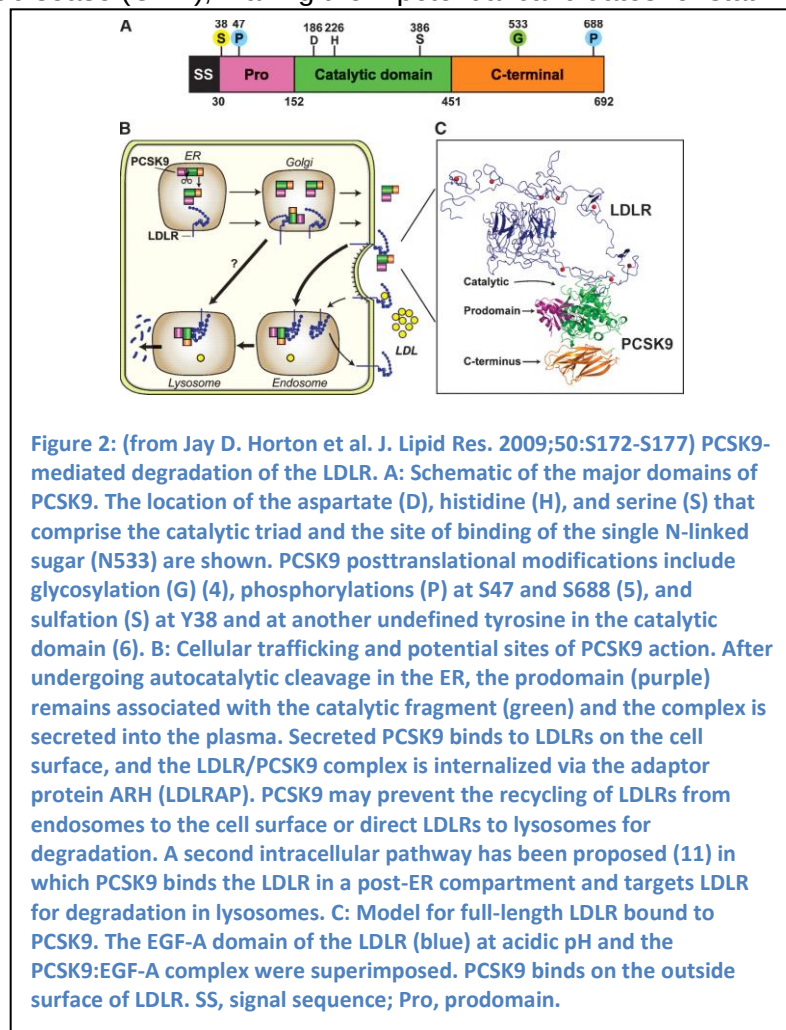
Third, 5-10% of statin users develop intolerance (15). According to the Centers for Disease Control and Prevention (CDC) data from 2003, 37% of the US population has at least two risk factors for coronary heart disease (CHD), making them potential candidates for statin use. If 5-10% of these individuals experience statin-induced muscular symptoms, then the estimated potential prevalence of statin myopathy is 5 -10 million in the US (16).

Proprotein convertase subtilisin/kexin 9 (PCSK9)

The road from discovery to drug development was rapid for PCSK9 inhibitors.

In 2003, a gain-of-function mutation in *PCSK9* was discovered to cause familial hypercholesterolemia in a French FH family (17). Additional families were soon identified in Utah, confirming *PCSK9* as the causative gene.

Work by Helen Hobbs, MD, Jonathan Cohen, PhD, and Jay Horton, MD, helped elucidate the human genetics and physiological roles for *PCSK9*. Perhaps most importantly, in 2005, Hobbs et al identified loss-of-function mutations in *PCSK9* resulting in



low cholesterol and an 88% reduced risk of coronary heart disease (18).

Proprotein convertase subtilisin/kexin 9 (PCSK9) is a member of the proprotein convertase family of zymogens and modulates plasma LDL-C levels by promoting degradation of LDL receptors (LDLRs). PCSK9 is expressed at highest levels in the liver, small intestine, brain, and kidney (19). PCSK9 expressed from the liver is secreted into the circulation and associates with LDLRs. In the presence of PCSK9, LDLRs are targeted for degradation rather than recycling to the hepatocyte cell surface (Figure 2) (20).

Based on the phenotype of humans with PCSK9 deficiency – hypocholesterolemia without any apparent health consequences (18)- several pharmaceutical companies established development programs for PCSK9 inhibitors. These inhibitors mostly interfere with the PCSK9-LDLR interaction at the LDLR epidermal growth factor-like repeat A (EGF-A) region. Blocking this interaction increases active LDL uptake and reduces circulating cholesterol (21, 22).

Two PCSK9 inhibitors are FDA approved, but the indications can be variably interpreted

Two PCSK9 monoclonal antibodies have obtained approval by the US FDA: evolocumab (Amgen) and alirocumab (Sanofi/Regeneron). Per their package inserts, both are “indicated as an adjunct to diet and *maximally tolerated statin therapy* for the treatment of adults with *heterozygous familial hypercholesterolemia (FH)* or *clinical atherosclerotic cardiovascular disease*, who require *additional lowering of LDL-cholesterol (LDL-C)*.” Evolocumab has an additional indication for homozygous FH patients.

This labeled indication included terminology that can be variably interpreted and has created confusion between providers and payers:

- “Maximally tolerated statin” may mean
 - High dose/potency statin (e.g. atorva 80 mg daily)
 - Highest dose of statin that patient can handle (e.g. rosuva 5 mg once weekly)
 - No statin if patient cannot tolerate
- ASCVD is not fully defined.
 - Per the 2013 AHA/ACC Cholesterol treatment guidelines, ASCVD includes
 - Coronary Heart Disease
 - Acute coronary syndrome
 - History of myocardial infarction
 - Stable or unstable angina
 - Coronary (or other) revascularization
 - Stroke or TIA
 - Peripheral arterial disease
 - Some unclear categories:
 - Diabetics, who are usually considered to be at very high ASCVD risk
 - Patients with abnormal Coronary Artery Calcium scores but no ASCVD events.
- No standard FH diagnostic criteria exist in the US. Possible ways to diagnose FH include
 - Genetic confirmation, which is only done in 5% of all cases in the US.
 - “Clinically” diagnosed” with elevated LDL-C levels (usually > 190 mg/dL) plus a suspicious family history.
 - Complex scoring systems, such as the Dutch Lipid Clinic Network Scoring criteria, stratify patients into “unlikely, possible, probable, or definite” FH.

- Who needs “additional lowering of LDL-C?”
 - The AHA/ACC guidelines and the American Diabetes Association recommend at least 50% reduction in LDL-C
 - The National Lipid Association, International Atherosclerosis Society, European Society for Cardiology/European Atherosclerosis Society, and the American Academy of Clinical Endocrinologists still advocate for an LDL-C goal of < 70 mg/dL for patients with ASCVD
 - The National Lipid Association recommends an LDL-C goal of < 100 mg/dL for FH patients

Both approved agents lower LDL-C robustly and consistently in all patient populations.

Evolocumab and Alirocumab have been administered to over 30,000 patients in nearly 40 published clinical trials. Both drugs lower LDL-C by 50-60% with minimal adverse events.

Alirocumab has even been tried in a few FH patients with mutations in *PCSK9*, and the drug lowered LDL-C by 70%.

What are the differences between the two drugs?

Both drugs are quite effective at lowering LDL-C. Both have roughly the same cost (\$14,000-15,000 per year). A few differences, mostly minor, exist.

Alirocumab is available in two doses that are administered SQ every 2 weeks. The 75mg/mL dose reduces LDL-C by 50% on average and the 150 mg/mL dose reduces LDL-C by 60% on average. Patients who do not achieve adequate reduction on the 75 mg/mL dose can be titrated up to the 150 mg/mL dose.

Evolocumab is available in a 140 mg/ml dosage every 2 weeks or a once monthly dosage administered through an on-body infuser.

Alirocumab must be kept refrigerated, while Evolocumab can be left at room temperature for 28 days prior to administration. For patients who travel or do not have access to refrigeration, Evolocumab may be a better choice.

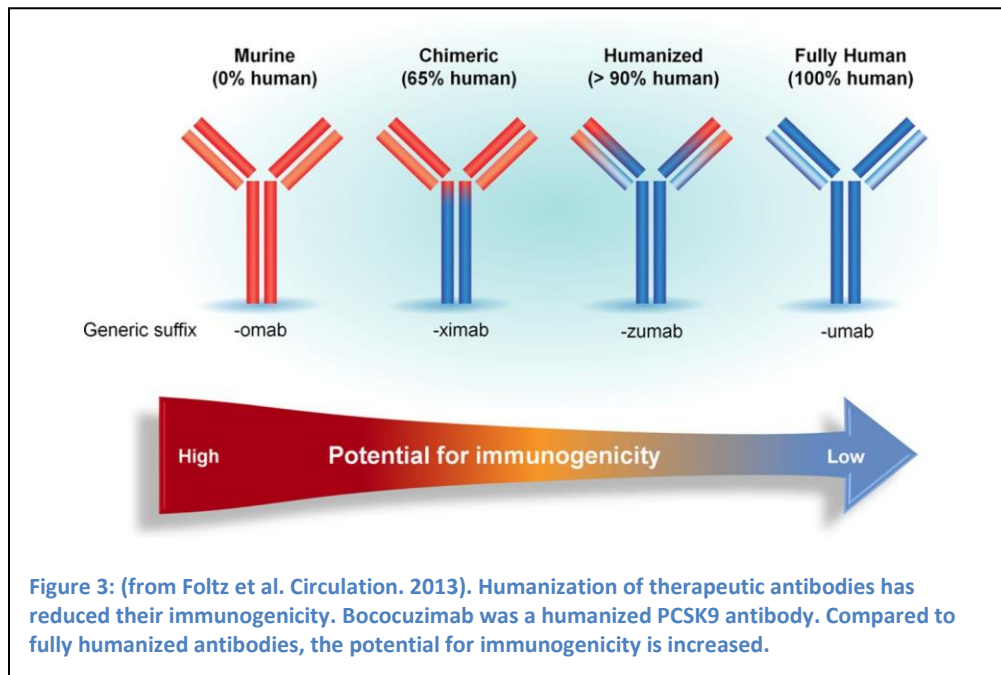
Evolocumab is approved in Europe. For patients moving or spending extended time in a European country, Evolocumab may be a better choice.

Alirocumab is free of latex. For patients with latex allergies, Alirocumab would be a better choice.

What other PCSK9 drugs are in development?

Several types of PCSK9-targeted agents are in development (based on my internet searches): monoclonal antibodies (Merck), adnectins specific inhibitors (Bristol-Myers Squibb/Adnexus), RNA interference (Alnylam/Novartis), locked nucleic acids (Santaris/Novartis), and small molecule inhibitors (Shifa Biomedical Corporation, Cadila

Healthcare, Serometrix). Most of these are in preclinical development. Two are worth discussing further: Pfizer's discontinued PCSK9 mAb and Alnylam's PCSK9 RNAi drug.



Pfizer discontinued the development of its PCSK9 mAb, bococuzimab. During the conduct of phase II and III trials, data became available from indicating that bococizumab was commonly associated with the development of high-titer antidrug antibodies that resulted in substantive attenuation of LDL-cholesterol lowering over time (Figure 3). In addition, the trials showed that bococizumab was associated with a wide variation in LDL-C, even among patients who were antibody negative. High antibody titers were also linked to higher rates of injection site reactions. On the basis of these data, Pfizer elected to discontinue further development of bococizumab on November 1, 2016.

Alnylam has developed a small interfering RNA targeting PCSK9 mRNA. The small interfering RNAs consist of two strands, guide and passenger (23). The guide strand carries the sequence information necessary for target-gene recognition, while the passenger strand serves as a prodrug that supports the geometry required for loading into the RNA-induced silencing complex (RISC). When siRNAs are introduced into the cells, the guide strand enters the RISC and reprograms the powerful natural mechanism RNA interference (RNAi), silencing genes on demand. The loaded RISC has a long half-life, and as few as 100 to 200 loaded RISC complexes per cell are sufficient to eliminate expression of the targeted gene.

In a randomized, double-blind, placebo-controlled, phase 2, multicenter trial, Alnylam tested their RNAi drug, Inclisiran (24). The drug was administered as a subcutaneous injection in patients at high risk for cardiovascular disease who had elevated LDL-C levels. A total of 501 patients were randomly assigned to receive a single dose of placebo or 200, 300, or 500 mg of inclisiran or two doses (at days 1 and 90) of placebo or 100, 200, or 300 mg of inclisiran. The

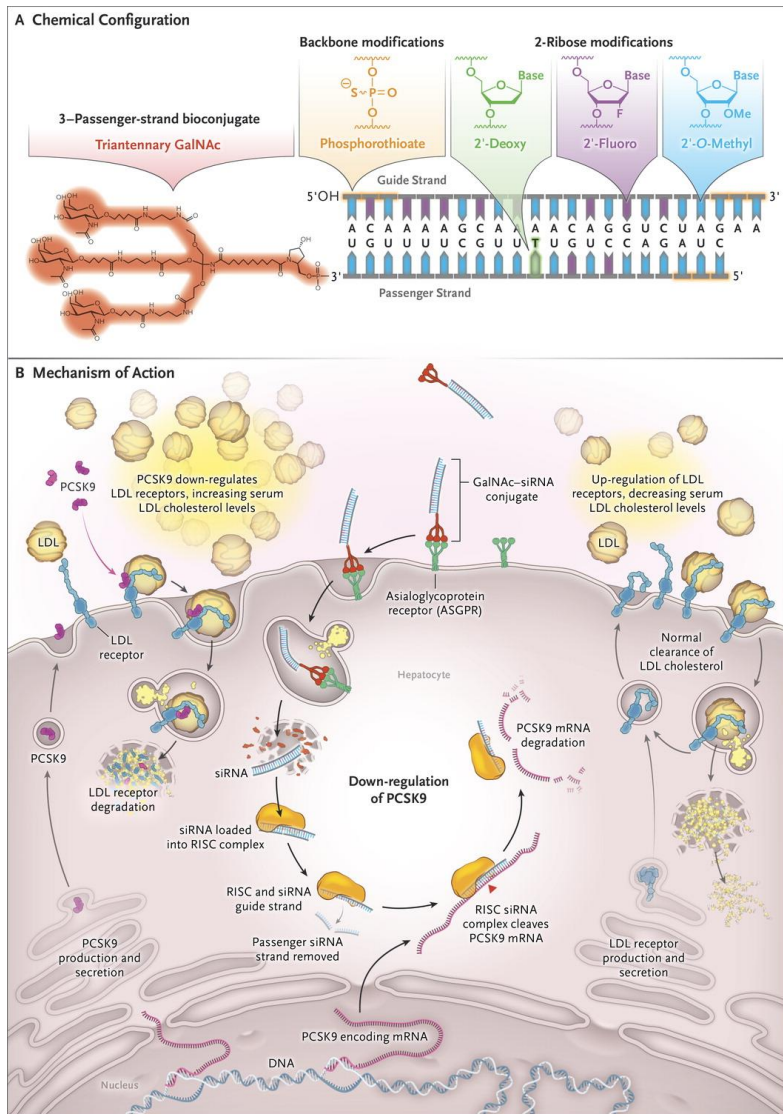


Figure 4: (from Khvorova A. *N Engl J Med* 2017;376:4-7). **Inclisiran Chemical Configuration and Mechanism of Action.** Inclisiran is a fully chemically modified siRNA conjugated to the triantennary GalNAc. Inclisiran is made from one 2' -deoxy, eleven 2' -fluoro, and thirty-two 2' -O-methyl modified nucleotides. Termini of the duplex are modified with phosphorothioates, and the 3' end of the passenger strand is functionalized with triantennary GalNAc (Panel A). When inclisiran is delivered to the hepatocytes through GalNAc interactions with the ASGPR receptor, the guide strand of the duplex enters the RISC complex, hybridizes to the PCSK9 mRNA and cleaves it (red triangle), preventing protein production. Down-regulation of PCSK9, a protein involved in low-density lipoprotein (LDL) receptor degradation, results in the up-regulation of LDL receptor levels on the surface of the hepatocytes, supporting more efficient clearance of LDL cholesterol from the bloodstream (Panel B).

primary endpoint was LDL-C levels at day 180. Based on the results, the optimal dose is 300 mg given at day 0 and day 90. With this dose, LDL-C reductions were 53% at day 180. In contrast, the single dose 300 mg had 38% LDL-C reduction at day 180. Adverse events were similar in patients who received inclisiran compared to placebo with a few notable exceptions: injection-site reactions occurred only in patients who received study drug; a few patients receiving study drug developed elevations in liver function tests and creatine kinase, and two

deaths occurred in patients who got the study drug. Of note, these two deaths occurred late in the study and were considered to be not related to study drug. Future plans for Alnylam include a phase II cardiovascular outcomes study in high risk primary and secondary prevention patients with average LDL-C ~ 130 mg/dL

How low is too low for LDL-C reduction?

Previously, studies with statins or statins in combination with zetia achieved LDL-C as low as 40-50 mg/dL without any obvious increase in adverse events. Now, with PCSK9 inhibitors, LDL-C levels are being reduced to < 25 in a very large group of patients. Thus far, the adverse event rate has been similar for patients with LDL-C < 25 mg/dL compared to > 40 mg/dL. Of note, the long-term effects of extremely low LDL-C levels have not been established.

Aren't these drugs too expensive?

Evolocumab and Alirocumab cost \$14,000-\$15,000 per year. For a biologic agents, this cost is not excessive. However, for a lipid lowering drug, it far exceeds the next most expensive drug: \$2784/year for pitavastatin (the only non-generic statin).

More problematic is the large patient population that potentially qualifies for these agents. Even prior to being FDA approved, several payers had already nicknamed them “biologic budget busters.”

Not surprisingly, payer approval has proven difficult. Payers have required prior authorizations, STEP therapy, and prolonged appeal processes. Payer policy requirements vary, causing a great deal of confusion for providers and patients. Even when approved, some payers have high co-payments (\$300/month), making it financially difficult on middle- or low- income patients.

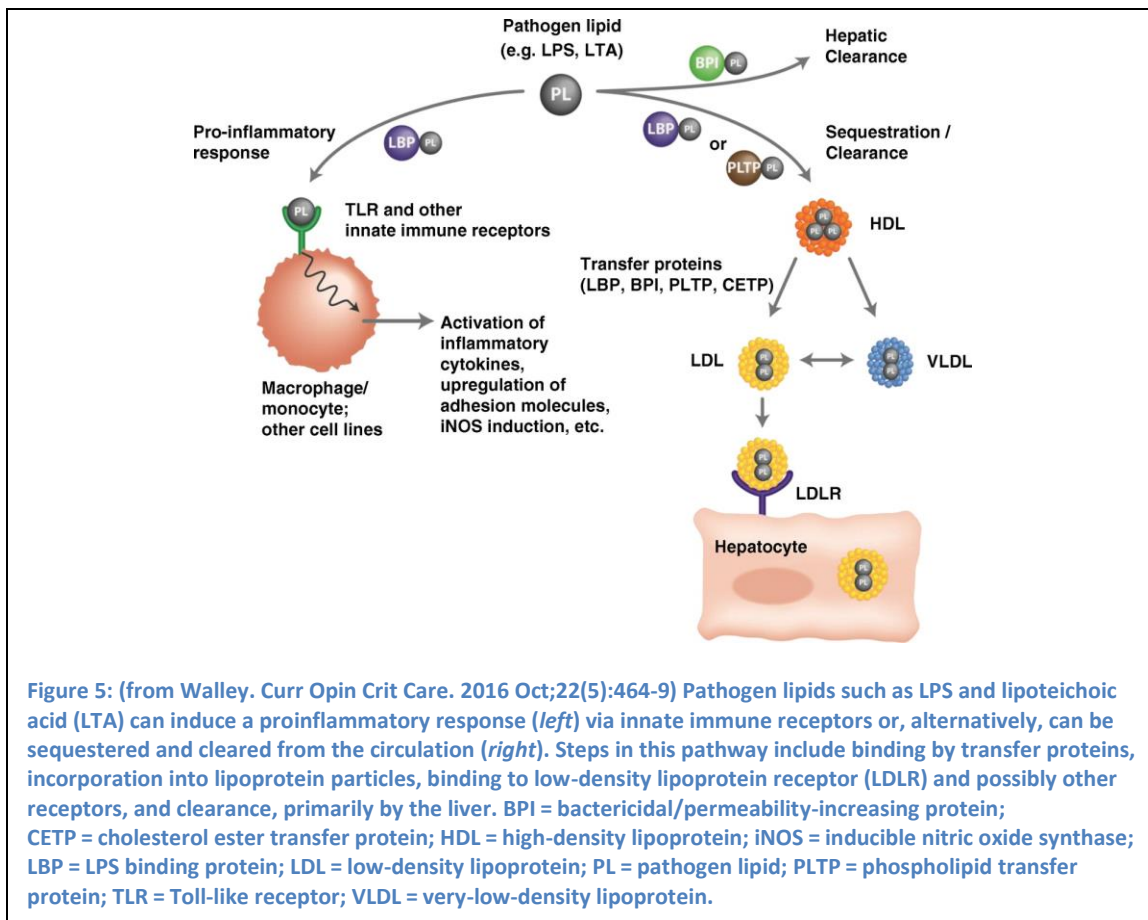
With regards to cost-effectiveness, the cost would need to be reduced to ~ \$6000/year for quality-adjusted life year of \$ 150,000. Of note, this estimate is based on estimates of ASCVD risk reduction; now that outcomes data is available, new estimates should be forthcoming.

Other than LDL-C related effects, what physiological role does PCSK9 play?

PCSK9 is expressed at the highest levels in liver, kidney and brain. The role of PCSK9 other than LDL-C related effects has not entirely been determined.

Recently, PCSK9 has been implicated in pathogen lipid clearance (Figure 5). Specifically, pathogen lipids such as lipopolysaccharide and lipoteichoic acid – which induce a proinflammatory response via innate immune receptors - can be sequestered and cleared from the circulation after being incorporated into lipoprotein particles. Steps in this pathway include binding by transfer proteins, incorporation into lipoprotein particles, binding to LDLR, and clearance. Since PCSK9 affects LDLR, it exerts an effect on removing pathogen lipids.

Humans with both loss of function and gain of function PCSK9 variants have been investigated for their survival from septic shock. Patients with loss-of-function PCSK9 variants, and thus more LDLRs, have improved survival compared to wild-type individuals while patients with gain-of-function variants have worsened survival.

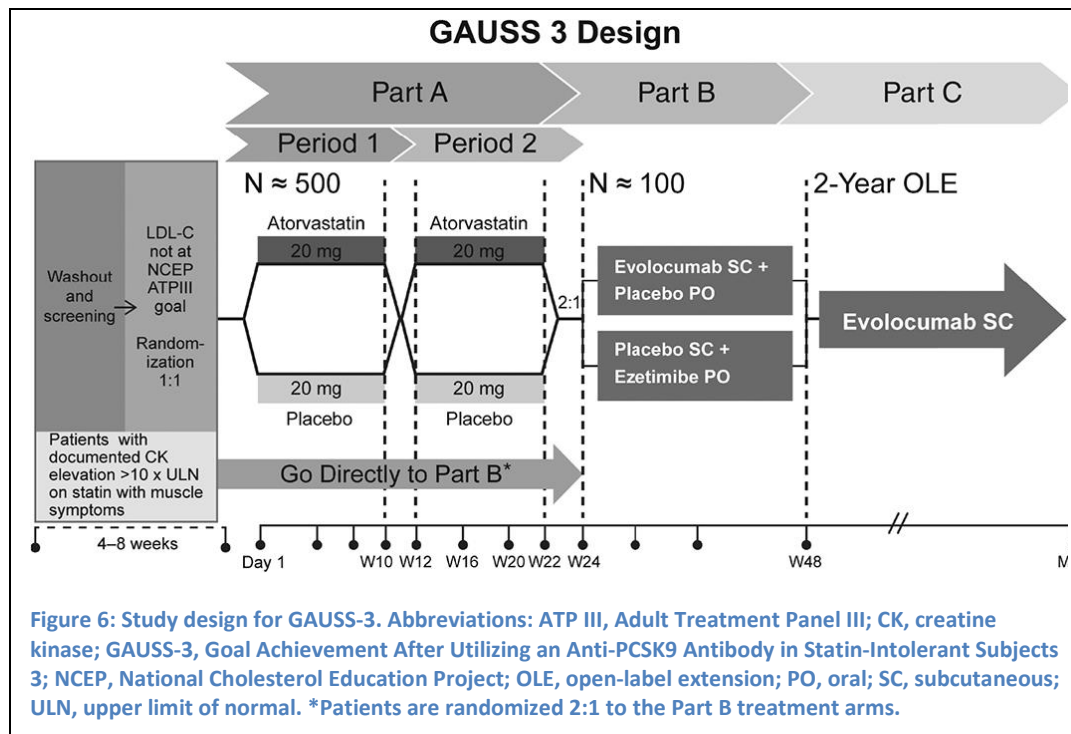


These findings led Novartis to license its PCSK9 antibody drug to a start-up, Cyon, for phase II studies in sepsis.

Does PCSK9 inhibition work in patients who are statin intolerant?

Four studies have been done in patients with statin intolerance. The best of these studies is GAUSS-3, a randomized clinical trial of evolocumab vs ezetimibe in patients with muscle-related statin intolerance (Figure 6) (25).

To be enrolled in the study, patients had to have previous intolerance to ≥ 3 statins (one of which at lowest daily dose), or intolerance to 2 statins, one of which must be atorvastatin ≤ 10 mg. The first phase of the study involved a double-blind, crossover (10 weeks per arm) challenge of atorvastatin 20 mg vs placebo. Only patients with muscle pain on atorvastatin but not on placebo enrolled in remainder of the study. A total of 491 patients entered into this phase, and 43% had muscle pain on atorvastatin but not while taking placebo. These patients entered phase B, which involved 2:1 randomization to subcutaneous evolocumab (420 mg monthly) or oral ezetimibe (10 mg daily). As expected, the evolocumab group reduced LDL-C by 53% while the ezetimibe group reduced LDL-C by 17%. Interestingly, muscle symptoms were reported by 29% of ezetimibe-treated patients and 21% of evolocumab-treated patients. These findings suggest that among patients with clinically reproducible statin intolerance, evolocumab can be tolerated by most.



Based on GAUSS-3 and other PCSK9 inhibitions studies of statin intolerant patients, we can state the following: PCSK9 inhibition causes less muscle symptoms than ezetimibe.

Outcomes

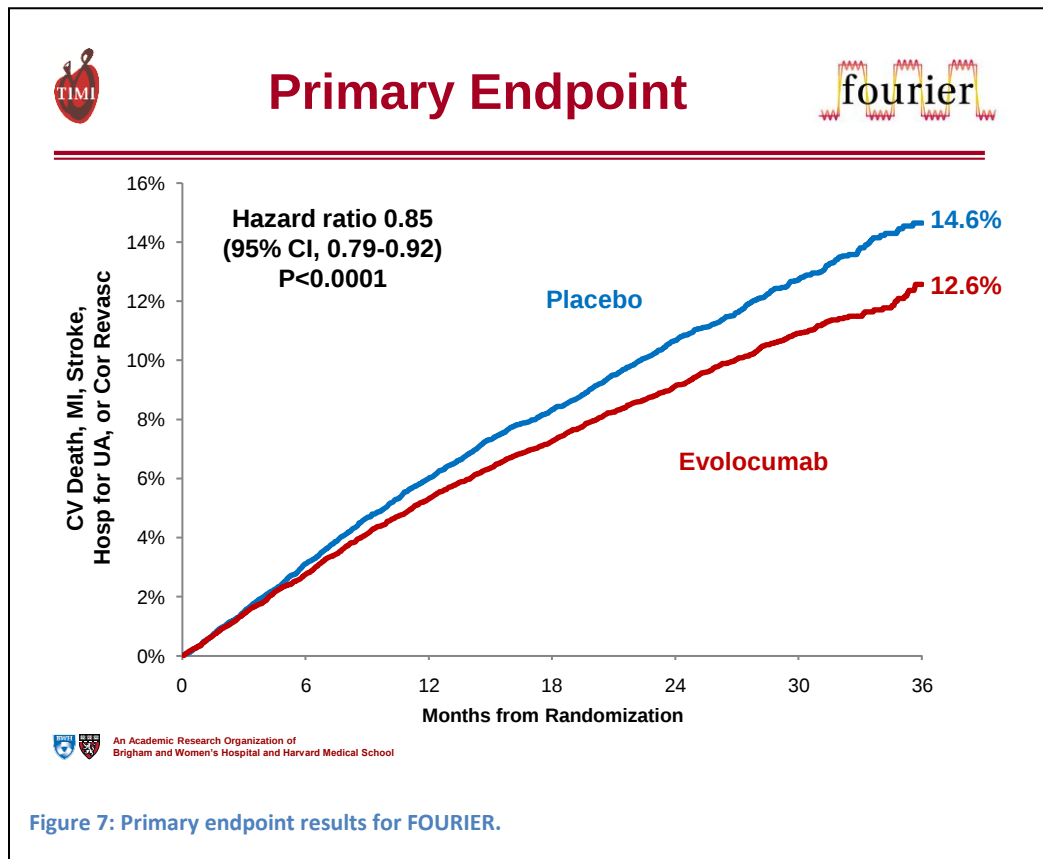
The FOURIER study showed that Evolocumab reduces cardiovascular events

The first cardiovascular outcomes trial was published just two weeks prior to this presentation. Amgen presented data from the Further cardiovascular Outcomes Research with PCSK9 Inhibition in subjects with Elevated Risk (FOURIER) trial at the 2017 ACC meeting in Washington, DC. The study enrolled 27, 564 patients with established cardiovascular disease. Patients first went into a stabilization phase that included the use of high intensity statin therapy (with or without ezetimibe). Then, they were randomized, in a double-blinded manner, in 1:1 ratio to get evolocumab (either 140 mg every 2 weeks or 420 mg every month, according to patient preference) or matching placebo. The primary endpoint was major cardiovascular events, defined as the composite of cardiovascular death, myocardial infarction, stroke, hospitalization for unstable angina, or coronary revascularization.

In FOURIER, evolocumab reduced LDL-C from a baseline median of 92 mg/dL (IQR 80-109) to a median of 30 mg/dL (IQR 19-46).

Although the absolute risk reduction was just 1.5%, the relative risk reduction was 15% for the primary endpoint (Figure 7). Based on analyses of secondary outcomes, the major benefit was in reduction of MI or stroke. No improvement of cardiovascular death was noted, which may be for several reasons: the study may not be long enough in duration, or the study may lack power

to compare evolocumab against less intensive therapy (rather than placebo) for mortality outcomes.



Atheroma regression likely is the mechanism linking LDL-C reduction to the relative risk reduction for MI and stroke seen with Evolocumab. In the Global Assessment of Plaque Regression with a PCSK9 Antibody as Measured by Intravascular Ultrasound (GLAGOV) trial, the addition of evolocumab to statin therapy produced a significant atheroma regression and continuous benefit with LDL-C levels right down to 20 mg/dL. The GLAGOV trial enrolled 968 patients with angiographic coronary disease (mean age 59.8 years, 27.8% women, mean LDL-C 92.5 mg/dL) from 197 hospitals in North America, Europe, South America, Asia, Australia, and South Africa. All patients were taking statins at enrollment (60% high intensity, 40% moderate intensity). The study showed an average reduction of approximately 1% in atheroma volume after 18 months of treatment with evolocumab added to statin therapy. About two-thirds of patients showed plaque regression.

In the safety data, patients on Evolocumab experienced more injection-site reactions (2.1% vs 1.6%, $p < 0.001$). Otherwise no other adverse events occurred in the evolocumab group more frequently than the placebo group. The most common adverse event was allergic reaction (3.1% vs 2.9%).

Despite the trials being halted prematurely, Bococizumab also showed a reduction in the risk of major cardiovascular events.

As part of the Studies of PCSK9 Inhibition and the Reduction of Vascular Events (SPIRE) program, two large-scale trials to evaluate cardiovascular outcomes (designated SPIRE-1 and SPIRE-2) were initiated in October 2013 with the intent of evaluating the clinical efficacy and safety of bococizumab administered at a dose of 150 mg subcutaneously every 2 weeks among patients who had evidence of cardiovascular disease or who were at high risk for a first vascular event. However, during the conduct of these two outcomes trials, data became available from the all Bococizumab lipid-lowering trials indicating that bococizumab was commonly associated with the development of high-titer antidrug antibodies that resulted in substantive attenuation of LDL-cholesterol lowering over time. In addition, the trials showed that bococizumab was associated with a wide variation in LDL-C, even among patients who were antibody negative. On the basis of these data, the sponsor elected to discontinue further development of bococizumab on November 1, 2016. As a consequence of that decision, the sponsor also elected to prematurely stop the ongoing SPIRE-1 and SPIRE-2 outcome trials.

In SPIRE-1, the lower-risk, shorter-duration trial (in which the patients had a baseline LDL cholesterol level of ≥ 70 mg per deciliter and the median follow-up was 7 months), no reduction in major cardiovascular events occurred.

In SPIRE-2, the higher-risk, longer-duration trial (in which the patients had a baseline LDL cholesterol level of ≥ 100 mg per deciliter and the median follow-up was 12 months), bococizumab reduced the risk of major cardiovascular events (hazard ratio, 0.79; 95% CI, 0.65 to 0.97; $P=0.02$).

Injection site reactions led to discontinuation of study drug more commonly than placebo (incidence difference 8.33, $p < 0.001$). Glucose was slightly higher in the study drug groups than in the placebo group (incidence difference 1.7 mg/dL, $p = 0.004$).

Conclusions

For patients with ASCVD, FH, and statin intolerance, a need exists for additional lipid lowering drugs to adequately reduce their LDL-C and risk of ASCVD events. PCSK9 inhibitors can reduce LDL-C by 50-60% with a favorable safety profile. Two such drugs were approved in 2015 by the FDA for use in patients with ASCVD or FH. Recent outcome studies of one of these drugs (evolocumab) found a 15% risk reduction for major cardiovascular events. Overall, PCSK9 inhibitors provide a new and efficacious method of lowering LDL-C. If costs can be reduced, a large group of patients will benefit from these drugs.

References (maximum 25)

1. **Ohsfeldt RL, Gandhi SK, Smolen LJ, Jensen MM, Fox KM, Gold A, Hsia J** Cost effectiveness of rosuvastatin in patients at risk of cardiovascular disease based on findings from the JUPITER trial. *J Med Econ* 13:428-437
2. **Cannon CP, Blazing MA, Giugliano RP, McCagg A, White JA, Theroux P, Darius H, Lewis BS, Ophuis TO, Jukema JW, De Ferrari GM, Ruzyllo W, De Lucca P, Im K, Bohula EA, Reist C, Wiviott**

- SD, Tershakovec AM, Musliner TA, Braunwald E, Califf RM** 2015 Ezetimibe Added to Statin Therapy after Acute Coronary Syndromes. *New England Journal of Medicine* 372:2387-2397
3. **Jones PH, Nair R, Thakker KM** 2012 Prevalence of dyslipidemia and lipid goal attainment in statin-treated subjects from 3 data sources: a retrospective analysis. *J Am Heart Assoc* 1:e001800
4. **Huijgen R, Kindt I, Verhoeven SB, Sijbrands EJ, Vissers MN, Kastelein JJ, Hutten BA** 2010 Two years after molecular diagnosis of familial hypercholesterolemia: majority on cholesterol-lowering treatment but a minority reaches treatment goal. *PLoS One* 5:e9220
5. **Thomas ME, Harris KP, Ramaswamy C, Hattersley JM, Wheeler DC, Varghese Z, Williams JD, Walls J, Moorhead JF** 1993 Simvastatin therapy for hypercholesterolemic patients with nephrotic syndrome or significant proteinuria. *Kidney Int* 44:1124-1129
6. **Hopkins PN, Toth PP, Ballantyne CM, Rader DJ, National Lipid Association Expert Panel on Familial H** 2011 Familial hypercholesterolemias: prevalence, genetics, diagnosis and screening recommendations from the National Lipid Association Expert Panel on Familial Hypercholesterolemia. *Journal of clinical lipidology* 5:S9-17
7. **Benn M, Watts GF, Tybjaerg-Hansen A, Nordestgaard BG** 2012 Familial hypercholesterolemia in the danish general population: prevalence, coronary artery disease, and cholesterol-lowering medication. *The Journal of clinical endocrinology and metabolism* 97:3956-3964
8. **Nordestgaard BG, Chapman MJ, Humphries SE, Ginsberg HN, Masana L, Descamps OS, Wiklund O, Hegele RA, Raal FJ, Defesche JC, Wiegman A, Santos RD, Watts GF, Parhofer KG, Hovingh GK, Kovanen PT, Boileau C, Aversa M, Boren J, Bruckert E, Catapano AL, Kuivenhoven JA, Pajukanta P, Ray K, Stalenhoef AF, Stroes E, Taskinen MR, Tybjaerg-Hansen A, European Atherosclerosis Society Consensus P** 2013 Familial hypercholesterolaemia is underdiagnosed and undertreated in the general population: guidance for clinicians to prevent coronary heart disease: consensus statement of the European Atherosclerosis Society. *European heart journal* 34:3478-3490a
9. **Do R, Stitzel NO, Won HH, Jorgensen AB, Duga S, Angelica Merlini P, Kiezun A, Farrall M, Goel A, Zuk O, Guella I, Asselta R, Lange LA, Peloso GM, Auer PL, Project NES, Girelli D, Martinelli N, Farlow DN, DePristo MA, Roberts R, Stewart AF, Saleheen D, Danesh J, Epstein SE, Sivapalaratnam S, Hovingh GK, Kastelein JJ, Samani NJ, Schunkert H, Erdmann J, Shah SH, Kraus WE, Davies R, Nikpay M, Johansen CT, Wang J, Hegele RA, Hechter E, Marz W, Kleber ME, Huang J, Johnson AD, Li M, Burke GL, Gross M, Liu Y, Assimes TL, Heiss G, Lange EM, Folsom AR, Taylor HA, Olivieri O, Hamsten A, Clarke R, Reilly DF, Yin W, Rivas MA, Donnelly P, Rossouw JE, Psaty BM, Herrington DM, Wilson JG, Rich SS, Bamshad MJ, Tracy RP, Cupples LA, Rader DJ, Reilly MP, Spertus JA, Cresci S, Hartiala J, Tang WH, Hazen SL, Allayee H, Reiner AP, Carlson CS, Kooperberg C, Jackson RD, Boerwinkle E, Lander ES, Schwartz SM, Siscovick DS, McPherson R, Tybjaerg-Hansen A, Abecasis GR, Watkins H, Nickerson DA, Ardissino D, Sunyaev SR, O'Donnell CJ, Altshuler D, Gabriel S, Kathiresan S** 2015 Exome sequencing identifies rare LDLR and APOA5 alleles conferring risk for myocardial infarction. *Nature* 518:102-106
10. **Sjouke B, Kusters DM, Kindt I, Besseling J, Defesche JC, Sijbrands EJ, Roeters van Lennep JE, Stalenhoef AF, Wiegman A, de Graaf J, Fouchier SW, Kastelein JJ, Hovingh GK** 2014 Homozygous autosomal dominant hypercholesterolaemia in the Netherlands: prevalence, genotype-phenotype relationship, and clinical outcome. *European heart journal*
11. **Goldstein JL HH, Brown MS.** 2000 Familial hypercholesterolemia: new insights in pathogenesis and treatment. In: Scriver CR BA, Valle D, Sly WS ed. *The Metabolic and Molecular Bases of Inherited Disease*. New York: McGraw-Hill

12. **Versmissen J, Oosterveer DM, Yazdanpanah M, Defesche JC, Basart DC, Liem AH, Heeringa J, Witteman JC, Lansberg PJ, Kastelein JJ, Sijbrands EJ** 2008 Efficacy of statins in familial hypercholesterolaemia: a long term cohort study. *BMJ* 337:a2423
13. **Hadfield SG, Horara S, Starr BJ, Yazdgerdi S, Bhatnagar D, Cramb R, Egan S, Everdell R, Ferns G, Jones A, Marenah CB, Marples J, Prinsloo P, Sneyd A, Stewart MF, Sandle L, Wang T, Watson MS, Humphries SE** 2008 Are patients with familial hypercholesterolaemia well managed in lipid clinics? An audit of eleven clinics from the Department of Health Familial Hypercholesterolaemia Cascade Testing project. *Ann Clin Biochem* 45:199-205
14. **Leren TP, Finborud TH, Manshaus TE, Ose L, Berge KE** 2008 Diagnosis of familial hypercholesterolemia in general practice using clinical diagnostic criteria or genetic testing as part of cascade genetic screening. *Community Genet* 11:26-35
15. **Vandenberg BF, Robinson J** 2010 Management of the patient with statin intolerance. *Curr Atheroscler Rep* 12:48-57
16. **Stein EA** 2009 Other therapies for reducing low-density lipoprotein cholesterol: medications in development. *Endocrinol Metab Clin North Am* 38:99-119
17. **Abifadel M, Varret M, Rabes JP, Allard D, Ouguerram K, Devillers M, Cruaud C, Benjannet S, Wickham L, Erlich D, Derre A, Villegier L, Farnier M, Beucier I, Bruckert E, Chambaz J, Chanu B, Lecerf JM, Luc G, Moulin P, Weissenbach J, Prat A, Krempf M, Junien C, Seidah NG, Boileau C** 2003 Mutations in PCSK9 cause autosomal dominant hypercholesterolemia. *Nat Genet* 34:154-156
18. **Cohen JC, Boerwinkle E, Mosley TH, Jr., Hobbs HH** 2006 Sequence variations in PCSK9, low LDL, and protection against coronary heart disease. *N Engl J Med* 354:1264-1272
19. **Seidah NG, Benjannet S, Wickham L, Marcinkiewicz J, Jasmin SB, Stifani S, Basak A, Prat A, Chretien M** 2003 The secretory proprotein convertase neural apoptosis-regulated convertase 1 (NARC-1): liver regeneration and neuronal differentiation. *Proc Natl Acad Sci U S A* 100:928-933
20. **Horton JD, Cohen JC, Hobbs HH** 2009 PCSK9: a convertase that coordinates LDL catabolism. *J Lipid Res* 50 Suppl:S172-177
21. **Zhang DW, Garuti R, Tang WJ, Cohen JC, Hobbs HH** 2008 Structural requirements for PCSK9-mediated degradation of the low-density lipoprotein receptor. *Proc Natl Acad Sci U S A* 105:13045-13050
22. **Stefanutti C, Morozzi C, Di Giacomo S** 2012 New clinical perspectives of hypolipidemic drug therapy in severe hypercholesterolemia. *Curr Med Chem* 19:4861-4868
23. **Khvorova A** 2017 Oligonucleotide Therapeutics — A New Class of Cholesterol-Lowering Drugs. *New England Journal of Medicine* 376:4-7
24. **Ray KK, Landmesser U, Leiter LA, Kallend D, Dufour R, Karakas M, Hall T, Troquay RPT, Turner T, Visseren FLJ, Wijngaard P, Wright RS, Kastelein JJP** Inclisiran in Patients at High Cardiovascular Risk with Elevated LDL Cholesterol. *New England Journal of Medicine* 0:null
25. **Nissen SE, Stroes E, Dent-Acosta RE, Rosenson RS, Lehman SJ, Sattar N, Preiss D, Bruckert E, Ceska R, Lepor N, Ballantyne CM, Gouni-Berthold I, Elliott M, Brennan DM, Wasserman SM, Somaratne R, Scott R, Stein EA, Investigators G-** 2016 Efficacy and Tolerability of Evolocumab vs Ezetimibe in Patients With Muscle-Related Statin Intolerance: The GAUSS-3 Randomized Clinical Trial. *JAMA* 315:1580-1590