Abstract—Proprotein convertase subtilisin/kexin type 9 (PCSK9) plays an important role in the regulation of cholesterol homeostasis. By binding to hepatic low-density lipoprotein (LDL) receptors and promoting their lysosomal degradation, PCSK9 reduces LDL uptake, leading to an increase in LDL cholesterol concentrations. Gain-of-function mutations in PCSK9 associated with high LDL cholesterol and premature cardiovascular disease have been causally implicated in the pathophysiology of autosomal-dominant familial hypercholesterolemia. In contrast, the more commonly expressed loss-of-function mutations in PCSK9 are associated with reduced LDL cholesterol and cardiovascular disease risk. The development of therapeutic approaches that inhibit PCSK9 function has therefore attracted considerable attention from clinicians and the pharmaceutical industry for the management of hypercholesterolemia and its associated cardiovascular disease risk. This review summarizes the effects of PCSK9 on hepatic and intestinal lipid metabolism and the more recently explored functions of PCSK9 in extrahepatic tissues. Therapeutic approaches that prevent interaction of PCSK9 with hepatic LDL receptors (monoclonal antibodies, mimetic peptides), inhibit PCSK9 synthesis in the endoplasmic reticulum (antisense oligonucleotides, siRNAs), and interfere with PCSK9 function (small molecules) are also described. Finally, clinical trials testing the safety and efficacy of monoclonal antibodies to PCSK9 are reviewed. These have shown dose-dependent decreases in LDL cholesterol (44%–65%), apolipoprotein B (48%–59%), and lipoprotein(a) (27%–50%) without major adverse effects in various high-risk patient categories, including those with statin intolerance. Initial reports from 2 of these trials have indicated the expected reduction in cardiovascular events. Hence, inhibition of PCSK9 holds considerable promise as a therapeutic option for decreasing cardiovascular disease risk. (Circulation. 2015;132:1648-1666. DOI: 10.1161/CIRCULATIONAHA.115.016080.)

Key Words: antibodies, monoclonal ■ clinical trial ■ PCSK9 protein, human
Discovery of PCSK9

Gain-of-Function Mutations

Initially known as neural apoptosis-regulated convertase 1, PCSK9 was first identified and characterized in 2003. When levels of this protein were found to be elevated in cerebellar neurons during apoptosis, the clinical importance of PCSK9 became apparent when mutations in the PCSK9 gene were shown to cause dominant hypercholesterolemia. Mutations in PCSK9 were identified in 3 French families who had plasma cholesterol levels in the 90th percentile but lacked mutations in the genes encoding LDL-R and apolipoprotein (apo) B. PCSK9 overexpression studies in mice, which resulted in 2- to 5-fold increases in plasma total and non–HDL-cholesterol and a reduction in hepatic LDL-R levels, confirmed that the hypercholesterolemic phenotype observed in humans was caused by PCSK9 gain-of-function mutations. PCSK9 was thus identified as a third gene involved in the pathogenesis of autosomal-dominant familial hypercholesterolemia.

The initial gain-of-function mutations identified in the French families were near the protease autocatalytic processing site (S127R) and catalytic site (F216L) of PCSK9. S127R variants were found to have 5-fold greater binding affinity for LDL-R compared with wild type, whereas F216L mutants were reported to have a truncated form of secreted PCSK9 (≈53 kDa) resistant to digestion by furin. Notably, furin cleavage of PCSK9 yields a 55-kDa PCSK9 protein fragment considered less active than the intact protein in promoting LDL-R degradation. Numerous additional gain-of-function mutations associated with hypercholesterolemia and premature coronary heart disease (CHD) have since been identified and shown to distribute in all PCSK9 domains (reviewed elsewhere). One of the most potent mutations (D374Y), initially identified in a Utah family, is located in the catalytic domain of PCSK9 and has been shown to increase the affinity of PCSK9 for the LDL-R by as much as 25-fold and to reduce LDL-R levels by 23%.

Loss-of-Function Mutations

Two years after the identification of gain-of-function mutations, PCSK9 loss-of-function mutations were described and found to be associated with lower levels of LDL cholesterol (LDL-C) and reduced incidence of CVD. The 2 initially identified nonsense mutations, Y142X and C679X, were found to be more prevalent in individuals of African descent (≈2% compared with <0.1% in those of European ancestry) and were associated with age- and sex-matched LDL-C levels ranging from the 1st to the 50th percentile of the LDL-C distribution in African Americans. In a study comparing the incidence of CHD over 15 years among individuals taking part in the Atherosclerosis Risk in Communities (ARIC) study, nonsense mutations in PCSK9 were associated with a 28% reduction in LDL-C and an 88% reduction in CHD risk in blacks. Similarly, the R46L variant was associated with a 15% reduction in LDL-C and a 47% reduction in CHD risk among whites, finding consistent with those in subsequent reports. Numerous additional PCSK9 variants associated with a hypcholesterolemic phenotype have since been identified (reviewed elsewhere), namely G106R in a Norwegian family and R93C in a Japanese population. In a meta-analysis including 66698 individuals, carriers versus noncarriers of the 46L allele had a 12% reduction in LDL-C and a 28% decrease in risk of ischemic heart disease. This risk reduction substantially exceeded that predicted by the magnitude of LDL-C lowering, likely reflecting the cardiovascular benefit of genetically determined lifelong exposure to low levels of LDL-C.

Autocatalytic processing of pro-PCSK9, a necessary step for secretion of mature PCSK9 from the endoplasmic reticulum (ER), is prevented in those with a Q152H missense mutation at the site of PCSK9 autocatalytic cleavage and results in lower LDL-C levels. Notably, transfection of a mammalian cell line with a Q152H mutant construct was shown recently to interfere with both the cleavage and secretion of PCSK9, revealing a novel target for inhibition of PCSK9 function.

Concerns about possible deleterious effects associated with extreme reductions in plasma and LDL-C caused by low PCSK9 function are not supported by data from isolated cases such as a woman homozygous for the C679X loss-of-function mutation in PCSK9 whose LDL-C was 15 mg/dL, and another who was a compound heterozygote for the Y142X and AR97 loss-of-function mutations, which disrupt synthesis and processing/secretion of PCSK9, respectively, with no detectable levels of PCSK9 and an LDL-C level of 14 mg/dL. Although more extensive clinical experience is required to assess the generalizability of these anecdotal reports, as described below, there has been no excess of adverse events in patients in whom very low levels of LDL-C were induced in clinical trials of PCSK9 inhibitors.

PCSK9 Structure and Function in Regulating LDL Levels

PCSK9 is one of nine mammalian serine proteases aptly named due to their relation to the bacterial subtilisin and yeast kexin family. The first eight proteases, PCSK1-8, catalyze the proteolytic maturation of inactive secretory precursors to mature proteins such as hormones, enzymes and peptides (reviewed elsewhere). Although PCSK9 is synthesized and secreted primarily by the liver, it is expressed to a lesser extent in the small intestine, kidney, and central nervous system. In the hepatocyte, PCSK9 is synthesized as a 75-kDa zymogen consisting of a signal peptide, a prosegment, a catalytic domain, and a cysteine- and histidine-rich C-terminal domain that is required for trafficking of the PCSK9/LDL-R complex to the lysosome. It loses its N-terminal signal peptide and undergoes cotranslational autocatalytic cleavage in the ER to yield 2 products, the prosegment and a 62-kDa mature PCSK9 protein containing the catalytic domain and the C-terminal domain. The autocatalytic cleavage of PCSK9 is necessary for its maturation and secretion from the ER to the Golgi. Unlike other serine proteases, PCSK9 remains associated with the prosegment after cleavage. This prosegment facilitates folding of the protein but blocks access of potential substrates to the catalytic site of PCSK9, rendering the protease enzymatically inactive. The prosegment-PCSK9 complex then exits the ER and enters the secretory pathway for release of mature PCSK9. Despite its inactive state, PCSK9 binds to
LDL-Rs and promotes their lysosomal degradation\textsuperscript{43}, thereby preventing receptor recycling to the cell surface.\textsuperscript{22}

Although not enzymatically active,\textsuperscript{44} the catalytic domain of secreted PCSK9 associates with LDL-R at the hepatocyte plasma membrane by interacting with the epidermal growth factor precursor homology domain-A (EGF-A) of LDL-R.\textsuperscript{14} The PCSK9/LDL-R complex then enters the endosomal pathway and, in contrast to the LDL-R/LDL complex, does not dissociate at low pH in the endosome, likely because of the enhanced affinity of PCSK9 for EGF-A resulting from conformational changes in the LDL-R.\textsuperscript{45,46} Thus, the LDL-R does not recycle to the cell surface (Figure 1); rather, the PCSK9/LDL-R complex is directed to the lysosomal compartment for degradation of both PCSK9 and LDL-R.\textsuperscript{42} The interaction of PCSK9 with LDL-R can also occur within the hepatocyte, in which case the prosegment-PCSK9/LDL-R is directed from the Golgi to the lysosome for degradation before LDL-R transport to the cell surface.\textsuperscript{15} Although the molecular mechanism mediating PCSK9-induced LDL-R degradation is not completely understood, recent studies suggest that PCSK9 may mediate transport of LDL-R to the lysosome via interactions with amyloid precursor-like protein 2,\textsuperscript{47,48} a protein previously implicated in the transport of transmembrane proteins to lysosomes.\textsuperscript{49}

The Role of PCSK9 in Intestinal Lipid Metabolism

As noted above, PCSK9 is also expressed in the small intestine\textsuperscript{19} and appears to play an important role in intestinal triglyceride-rich apoB lipoprotein production\textsuperscript{50} and postprandial lipemia.\textsuperscript{51} Human enterocytes (Caco-2 cells) treated with human PCSK9 showed a 40\% increase in the secretion of apoB\textsubscript{48} and a 55\% increase in secretion of apoB\textsubscript{100} compared with untreated cells.\textsuperscript{50} The stimulatory effect of PCSK9 on intestinal apoB secretion was accompanied by a 1.5-fold increase in apoB mRNA and a comparable increase in mRNA levels for genes involved in the biosynthesis of fatty acids (fatty acid synthase, stearoyl CoA desaturase) and triglycerides (diacylglycerol acyltransferase-2) and for the microsomal triglyceride transfer protein, indicating upregulation of processes that ultimately may protect apoB from degradation. Consistent with these findings, PCSK9 knockout (PCSK9\textsuperscript{−/−}) mice secreted less intestinal apoB, showed a 62\% reduction in their postprandial triglyceride response to an olive oil bolus, and had a 37\% faster chylomicron clearance compared with wild-type mice, likely as a result of a greater abundance of hepatic LDL-R.\textsuperscript{51} Although the functional role of intestinal LDL-R in lipoprotein metabolism remains to be established, these findings suggest that targeting PCSK9 holds promise for the amelioration of postprandial hypertriglyceridemia. A role for PCSK9 in regulating postprandial lipoprotein metabolism is further supported by data from a recent kinetic study in 17 obese men and women in whom fasting plasma PCSK9 concentrations were positively correlated with the total and incremental area under the curve for plasma apoB\textsubscript{48} and inversely correlated with the fractional catabolic rate of apoB\textsubscript{48} in triglyceride-rich lipoproteins.\textsuperscript{52} Whether PCSK9 inhibition affects the postprandial metabolism of triglyceride-rich lipoproteins in humans remains to be established.

PCSK9 has also been implicated in the modulation of transintestinal cholesterol excretion (TICE), a recently identified route for fecal cholesterol excretion.\textsuperscript{53} The ability of intestinal cells to acquire cholesterol is modulated in part by Niemann-Pick C1-Like 1 (NPC1L1)-mediated luminal cholesterol uptake and, on the basolateral side, by uptake of LDL particles, a process dependent on LDL-R expression. Notably, PCSK9\textsuperscript{−/−} mice were shown to have increased fecal cholesterol excretion, whereas intravenous injection of recombinant PCSK9 acutely in these mice reduced TICE by 35\%.\textsuperscript{54} Conversely, PCSK9 administration had no effect on TICE in LDL-R knockout mice. This observation, together with the finding that wild-type mice treated with lovastatin for 10 days experienced a 71\%
increase in TICE, provides evidence that TICE modulation by PCSK9 is dependent on LDL-R expression.54

Other PCSK9 Targets

Adipocytes

In addition to LDL-R, PCSK9 modulates the expression of other receptors in the LDL-R family, including very-low-density lipoprotein (VLDL) receptors (VLDL-Rs), which are highly expressed in adipose tissue, heart, muscle, and brain (reviewed previously55), a process under the transcriptional control of sterol regulatory element binding protein-2.56 VLDL-Rs bind apoE in triglyceride-rich lipoproteins and their remnants and thus function in the delivery of fatty acids to peripheral tissues. Recent studies have therefore explored the role for circulating PCSK9 in the regulation of adipogenesis. Consistent with the function of PCSK9 in promoting VLDL-R degradation, mice lacking PCSK9 experienced an increase in VLDL-R protein levels in perigonadal tissues and accumulated more visceral fat than their wild-type counterparts.57 In contrast, a recent study reported no association between plasma PCSK9 levels and visceral fat accumulation in abdominally obese men,58 suggesting that PCSK9 may not significantly affect body fat distribution in humans.

Brain

VLDL-R and apoE receptor 2 are Reelin signaling receptors that are expressed in the brain and have a role in neural development (reviewed by Reddy et al59). Like other members of the LDL-R family, VLDL-R and apoE receptor 2 undergo lysosomal degradation after binding of PCSK9 to the EGF-A domains of the receptors.60,61 Notably, PCSK9 has been implicated in central nervous system development62 and, as noted above, was shown to enhance neuronal apoptosis,19,63 an effect mediated by degradation of the apoE receptor 2.63 Conversely, inhibition of PCSK9 led to an increase in cell viability for up to 24 hours in the setting of neural apoptosis induced by potassium deprivation.61

There is conflicting evidence bearing on the possibility of a relationship between brain PCSK9 expression to the pathophysiology of Alzheimer disease. One study showed increased levels of β-site amyloid precursor protein cleaving enzyme 1 (the rate-limiting enzyme for generation of amyloid β-peptide) and amyloid β-peptide in PCSK9−/− mice,64 whereas another study reported no change in either β-site amyloid precursor protein cleaving enzyme 1 or amyloid β-peptide.65

As recently reviewed elsewhere,66 there is controversy concerning the potential for PCSK9-based therapies to cause neurocognitive impairment, an effect theoretically ascribed to the recognized ability of PCSK9 inhibitors to dramatically lower LDL-C, which has raised concern that this may affect central nervous system function. On the one hand, the brains of PCSK9−/− mice showed no gross structural differences compared with wild-type mice, suggesting no adverse effect of PCSK9 deficiency on brain development,67 although the applicability of this finding to humans is uncertain. On the other hand, it is argued that the potent lipid-lowering effect of PCSK9 inhibitors may improve cerebral arterial health in a manner that could diminish the risk of vascular dementia, as has been suggested for statins.68 Recent findings of slightly higher occurrences of neurocognitive events in the treatment versus standard of care patient groups from the Open-Label Study of Long-Term Evaluation Against LDL Cholesterol (OSLER)69 and Long-Term Safety and Tolerability of Alirocumab (SAR236553/REGN727) Versus Placebo on Top of Lipid-Modifying Therapy in High Cardiovascular Risk Patients With Hypercholesterolemia (ODYSSEY LONG TERM) trials,70 testing the safety and efficacy of evolocumab and alirocumab, respectively (described further below), suggest that further monitoring of cognitive safety with PCSK9 inhibition is required.

Pancreatic β-Cells

Excess LDL-R–mediated delivery of cholesterol to pancreatic β-cells has been associated with β-cell dysfunction manifested as reduced glucose-stimulated insulin secretion71 and β-cell apoptosis.72 The expression of PCSK9 in pancreatic islets19 has therefore raised concern about possible adverse effects of PCSK9 inhibition on β-cell function. As recently reviewed elsewhere,73 studies of β-cell function and glucose homeostasis in PCSK9−/− mice are inconsistent. Some showed increased β-cell apoptosis and glucose intolerance,74,75 whereas another suggests normal glucose-stimulated insulin secretion and glucose tolerance, possibly resulting from compensatory increases in islet cholesterol efflux.73 In humans, loss-of-function mutations in PCSK9 in the ARIC population were not associated with increased diabetes risk.30 Given the recognized association of statins with new-onset type 2 diabetes mellitus,12,13 it will be important to determine whether PCSK9-targeted therapies may adversely affect glucose homeostasis.

PCSK9 and Sepsis

In recent studies, PCSK9−/− mice were shown to have lower plasma concentrations of inflammatory cytokines and lower levels of endotoxins after lipopolysaccharide administration compared with wild-type mice, a finding consistent with faster lipopolysaccharide removal from the circulation.74 In addition, pharmacological inhibition of PCSK9 blunted lipopolysaccharide-induced sepsis in wild-type mice but not in LDL-R−/− mice, indicative of a role for LDL-R in lipopolysaccharide clearance. In agreement with these observations, genetic data from 2 cohorts treated for sepsis revealed that patients with at least 1 loss-of-function mutation in PCSK9 had increased survival, whereas those with gain-of-function mutations had decreased survival after sepsis.78 Notably, in a systematic review and meta-analysis of 7 randomized, controlled trials (n=1720 patients), statin therapy did not decrease in-hospital or 28-day mortality in patients with sepsis.77 Whether PCSK9 inhibition affects mortality outcomes in patients with sepsis requires clinical trial validation.

PCSK9 and Other Lipoprotein Markers of CHD Risk

Our current understanding of the effects PCSK9 on other lipoprotein markers of CHD risk is based on trials assessing these biomarkers after PCSK9 inhibition.
Small, Dense LDL

LDL particles comprise multiple distinct subclasses differing in size, density, and chemical composition. Of these, small, dense LDL is the most strongly related to CVD risk, independently of traditional risk factors, including LDL-C.86 In a phase II randomized, double-blind, placebo-controlled trial of alirocumab (150 mg every 2 weeks for 12 weeks) or placebo conducted in patients with LDL-C ≥100 mg/dL on stable atorvastatin therapy, alirocumab reduced concentrations of all LDL, intermediate-density lipoprotein, and VLDL fractions measured by ion mobility,81 with the smallest effects noted for very small LDL, likely owing to the lower LDL-R binding affinity of these particles compared with larger, more buoyant LDL.82

Lipoprotein(a)

Lipoprotein(a) [Lp(a)] is known to be independently and causally associated with cardiovascular morbidity and mortality,83 with limited treatment options to effectively lower its plasma concentration. Numerous clinical trials testing monoclonal antibodies (mAbs) to PCSK9 have shown potent reductions in Lp(a).84 In a pooled analysis of 3 phase II studies of alirocumab (150 mg every 2 weeks for 8–12 weeks), PCSK9 inhibition resulted in a 30% reduction in Lp(a) versus placebo.84 These findings are consistent with another pooled analysis of 4 phase II trials of evolocumab (140 and 420 mg every 2 and 4 weeks, respectively, for 12 weeks) that reported dose-dependent 25% to 29% reductions in Lp(a).85 Although the mechanisms governing Lp(a) levels are still poorly understood, the ability of anti-PCS9 therapies to markedly reduce Lp(a) suggests that LDL-R–mediated uptake is a viable candidate for mediating this process. This possibility is supported by recent studies in which gain-of-function mutants of PCSK9 were more effective than wild-type PCSK9 in inhibiting uptake of Lp(a) by HepG2 cells.86 Moreover, treatment of the cells with mAbs to LDL-R was associated with a marked reduction in Lp(a) uptake, with no further reduction in Lp(a) internalization on treatment of the cells with PCSK9.86 Finally, overexpression of LDL-R in HepG2 cells resulted in a dramatic increase in Lp(a) internalization.86 From these findings, the authors speculated that supraphysiological levels of LDL-R such as those achieved with PCSK9 inhibition may be required to unmask the function of LDL-R in Lp(a) clearance,86 a concept that requires validation in vivo.

In contrast to mAbs, siRNAs, which prevent PCSK9 synthesis, have no effect on Lp(a) levels,87 suggesting that processes implicated in Lp(a) reduction with PCSK9 inhibition occur outside the cell. On the other hand, recent studies showing a reduction in Lp(a) with apoB100-lowering medications (reviewed by Lamon-Fava et al88) suggest that apoB synthesis is an important regulator of Lp(a) production. Notably, hepatic apoB production is modulated by PCSK9, possibly via an interaction that prevents intracellular degradation of apoB,89 providing an alternative intracellular hypothesis whereby reduced apoB synthesis may be implicated in the Lp(a)-lowering effect of PCSK9 inhibition. Finally, the possibility must be considered that other receptors involved in Lp(a) catabolism, namely the megalin/glycoprotein 330 receptor90 and scavenger receptor class B1,91 highly expressed in kidney and liver, respectively, may mediate Lp(a) reduction with PCSK9 inhibition.

Current Therapeutic Approaches and Their Clinical Efficacy

Interest in PCSK9 inhibitors as potential targets for LDL-C lowering was first generated by the effects of loss- and gain-of-function mutations30,60 in human PCSK9, described above. In the past decade, a great deal of effort has been devoted to developing molecules that reduce extracellular or intracellular levels of PCSK9 (reviewed by others92–94). These include inhibition of the PCSK9 interaction with LDL-R at the cell surface with mAbs, mimetic peptides, and adnectins; inhibition of PCSK9 synthesis in the ER with gene-silencing agents such as antisense oligonucleotides (ASOs) and siRNA, which reduce protein synthesis by targeting PCSK9 mRNA; and the use of small molecules that interfere with the autocatalytic processing required for mature PCSK9 secretion from the ER (Figure 2). The potential advantages and disadvantages of each approach are summarized in Table 1.

Clinical Trials of mAbs to PCSK9

The development of mAbs that bind extracellular PCSK9 and prevent its interaction with LDL-R is the most advanced and tested approach to PCSK9 inhibition to date. Advantages of mAbs over small molecule inhibitors include high potency, high specificity, and reduced dosing frequency. Human mAbs are generally well tolerated and have significantly lower rates of immunogenic incidents compared with chimeric or murine mAbs.95,96 However, the generally high production cost and short shelf-life of mAbs, together with the requirement for intravenous or subcutaneous routes of administration, limit the use of mAbs to PCSK9 to patients at highest risk for CVD or to those who are intolerant of statins.

Thus far, as described below, phase III trials have been completed for alirocumab (Regeneron/Sanofi) and evolocumab (Amgen). Bococizumab (Pfizer) is currently being tested in 4 large phase III trials targeted for completion between November 2015 and August 2017. The efficacy of PCSK9 mAbs in reducing atherogenic lipoproteins and cardiovascular events in phase III clinical trials is reviewed in detail below.

The results from phase III clinical trials of alirocumab and evolocumab have been reported in a number of different populations, including patients who are unable to tolerate statin therapy, hypercholesterolemic patients on background statin therapy, patients with heterozygous and homozygous familial hypercholesterolemia, hypercholesterolemic patients not taking lipid-lowering medications, and patients on monotherapy compared with ezetimibe. For bococizumab, clinical efficacy and safety data have been reported in a phase II trial conducted in patients with hypercholesterolemia on a background of statin therapy. Other approaches interfering with the ability of PCSK9 to interact with LDL-Rs (eg, mimetic peptides and adnectins) or inhibiting PCSK9 synthesis (eg, siRNAs and ASOs) have not been studied extensively in clinical trials.

Alirocumab (Regeneron/Sanofi)

In the Efficacy and Safety of Alirocumab SAR236553 (REGN727) Versus Ezetimibe in Patients With Hypercholesterolemia (ODYSY MONO) study, alirocumab was evaluated in 103 patients with hypercholesterolemia who were not on lipid-lowering therapy but who had a 1%
The patients were randomized to ezetimibe 10 mg daily or alirocumab 75 mg every 2 weeks. The alirocumab dose was titrated up to 150 mg if week 8 LDL-C was ≥70 mg/dL. After 24 weeks, alirocumab reduced LDL-C by 47% compared with 16% with ezetimibe. With alirocumab, levels of apoB and non–HDL-C were reduced by 37% and 41%, respectively. There were no differences in treatment-related adverse events between alirocumab and ezetimibe.

In the Study of Alirocumab (REGN727/SAR236553) in Patients With Primary Hypercholesterolemia and Moderate, High, or Very High Cardiovascular (CV) Risk, Who Are Intolerant to Statins (ODYSSEY ALTERNATIVE) trial, alirocumab was evaluated in patients with statin intolerance who did not have muscle-related adverse events on screening with subcutaneous or oral placebo. Patients (n=314) were randomized to alirocumab 75 mg every 2 weeks, ezetimibe 10 mg daily, or atorvastatin 20 mg daily. At 24 weeks of treatment, alirocumab reduced LDL-C by 45% compared with 16% with ezetimibe. With alirocumab, levels of apoB and non–HDL-C were reduced by 37% and 41%, respectively. There were no differences in treatment-related adverse events between alirocumab and ezetimibe.

The Efficacy and Safety of Alirocumab (SAR236553/REGN727) Versus Placebo on Top of Lipid-Modifying Therapy in High Cardiovascular Risk Patients With Hypercholesterolemia (ODYSSEY LONG-TERM) trial evaluated the efficacy and safety of treatment with alirocumab for 78 weeks in patients with heterozygous familial hypercholesterolemia with established CHD or CHD equivalent with LDL-C ≥70 mg/dL on maximal tolerated statin dose (47% of patients were on high-dose statin) or other lipid-lowering therapy. Patients (n=2341) were randomized to alirocumab 150 mg or placebo every 2 weeks. Alirocumab decreased LDL-C by 61% from baseline at 24 weeks and by 52% at 78 weeks. Reductions in non–HDL-C of 52%, apoB of 54%, and Lp(a) of 26% were observed with alirocumab therapy. In addition to evaluating safety and efficacy, the trial collected data on prespecified adjudicated CVD outcomes. Notably, a post hoc analysis showed that treatment with alirocumab was associated with a lower rate of major adverse cardiovascular events (1.7%) compared with placebo (3.3%; hazard ratio, 0.52; 95% confidence interval, 0.31–0.90; P=0.02). It has recently been argued, however, that

To 5% 10-year risk of CV death as assessed by the European Systematic Coronary Risk Estimation.97 The patients were randomized to ezetimibe 10 mg daily or alirocumab 75 mg every 2 weeks. The alirocumab dose was titrated up to 150 mg if week 8 LDL-C was ≥70 mg/dL. After 24 weeks, alirocumab reduced LDL-C by 47% compared with 16% with ezetimibe. With alirocumab, levels of apoB and non–HDL-C were reduced by 37% and 41%, respectively. There were no differences in treatment-related adverse events between alirocumab and ezetimibe.

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the suboptimal use of high-dose statins in the high-risk patient population of the ODYSSEY LONG TERM trial failed to maximize LDL-C lowering in the placebo group and may thus have contributed to overestimating the benefit of PCSK9 inhibition.102

Evolocumab (Amgen)

The clinical efficacy and safety of evolocumab have been studied in a number of phase III trials. In the Monoclonal Antibody Against PCSK9 to Reduce Elevated LDL-C in Subjects Currently Not Receiving Drug Therapy for Easing Lipid Levels-2 (MENDEL-2) study, 614 patients with hypercholesterolemia, defined as LDL-C ≥100 and <190 mg/dL, were randomized to placebo, ezetimibe, evolocumab 140 mg biweekly, or evolocumab 420 mg monthly for 12 weeks. Evolocumab reduced LDL-C by 56% to 57% compared with placebo, with comparable reductions achieved with the monthly dose.103 These findings were confirmed in the long-term Durable Effect of PCSK9 Antibody Compared With Placebo Study (DESCARTES), in which 901 hypercholesterolemic patients with LDL-C ≥75 mg/dL on a background of atorvastatin 10 to 80 mg daily with or without ezetimibe were randomized to placebo, ezetimibe, evolocumab 140 mg biweekly, or evolocumab 420 mg monthly for 12 weeks. Evolocumab reduced LDL-C by 56% to 57% compared with placebo, with 18% to 19% with ezetimibe without differences in adverse events.103 These findings were confirmed in the long-term Durable Effect of PCSK9 Antibody Compared With Placebo Study (DESCARTES), in which 901 hypercholesterolemic patients with LDL-C ≥75 mg/dL on a background of atorvastatin 10 to 80 mg daily with or without ezetimibe were randomized to evolocumab 420 mg every 4 weeks or placebo.104 At 52 weeks, evolocumab reduced LDL-C by 57% compared with placebo, with percent reductions ranging from 49% to 62% depending on background lipid-lowering therapy (atorvastatin alone or in combination with ezetimibe). Eighty-two percent of the patients on evolocumab were able to achieve an LDL-C goal of <70 mg/dL. The frequency of occurrence of adverse events was similar across groups.

The Goal Achievement After Utilizing an Anti-PCSK9 Antibody in Statin-Intolerant Subjects 2 (GAUSS-2) trial evaluated evolocumab compared with ezetimibe in 307 patients with hypercholesterolemia who were unable to tolerate statin therapy as defined by muscle-related side effects on ≥2 statins.105 Patients were randomized to evolocumab 140 mg every 2 weeks, 420 mg monthly plus oral placebo, or subcutaneous placebo plus ezetimibe. After 12 weeks of therapy, evolocumab reduced LDL-C by 56% and 55% in the every-2-weeks and monthly doses, respectively. In addition, evolocumab reduced apoB by 46% and Lp(a) by 24% to 26%. Notably, in a population selected on the basis of statin-induced myopathy, the occurrence of muscle adverse events was lower in the evolocumab group (8%) compared with the ezetimibe group (18%).

The LDL-C Assessment with PCSK9 Monoclonal Antibody Inhibition Combined With Statin Therapy 2 (LAPLACE-2) study investigated evolocumab combined with moderate- and high-intensity statins in patients with primary hypercholesterolemia and mixed dyslipidemia.106 Patients (n=2067) were included if they had screening LDL-C ≥150 mg/dL without statin therapy, LDL-C ≥100 mg/dL on a low-intensity statin dose, or LDL-C ≥280 mg/dL on an intensive statin dose. On the basis of their statin dose, patients who tolerated a placebo injection were randomized to placebo, evolocumab 120 mg every 2 weeks, evolocumab 420 mg once a month, or ezetimibe 10 mg daily. At a mean of 10 and 12 weeks of therapy, evolocumab reduced LDL-C by 66% to 75% with the biweekly dose and by 63% to 75% with the monthly dose in the moderate- and high-intensity statin groups compared with placebo. Additionally, biweekly evolocumab reduced non–HDL-C by 58% to 65%, apoB by 51% to 59%, and Lp(a) by 21% to 36% compared with placebo, with comparable reductions achieved with the monthly dose. Evolocumab was also studied in patients with homozygous familial hypercholesterolemia in the Trial Evaluating PCSK9 Antibody in Subjects With LDL Receptor Abnormalities (TESLA) Part B study.107 Patients (n=50) who had homozygous familial hypercholesterolemia (mean LDL-C, 347 mg/dL) diagnosed by either genetic analysis or clinical criteria were randomized to evolocumab 420 mg or placebo every 4 weeks. Ninety-two percent of the patients had a documented

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**Table 1. Pharmaceutical Approaches Targeting PCSK9**

<table>
<thead>
<tr>
<th>Approach</th>
<th>Advantages</th>
<th>Disadvantages</th>
<th>Compound</th>
<th>Developmental stage</th>
<th>Company</th>
</tr>
</thead>
<tbody>
<tr>
<td>mAbs</td>
<td>Highly selective</td>
<td>Intravenous or subcutaneous administration</td>
<td>Alirocumab/REGN7272/ SAR236553</td>
<td>FDA approved on July 24, 2015</td>
<td>Sanofi/Regeneron</td>
</tr>
<tr>
<td></td>
<td>Less dosing frequencies</td>
<td>High cost</td>
<td>Evolocumab/AMG145</td>
<td>FDA approved on August 27, 2015</td>
<td>Amgen</td>
</tr>
<tr>
<td>Mimetic peptides</td>
<td>Highly selective</td>
<td>Injection administration</td>
<td>Bococizumab/RN-316/ PF04950615</td>
<td>Phase III ongoing</td>
<td>Pfizer/Genetech</td>
</tr>
<tr>
<td>Adnectin</td>
<td>Selective</td>
<td>Short half-life</td>
<td>LY3015014</td>
<td>Phase II completed</td>
<td>Eli Lilly</td>
</tr>
<tr>
<td></td>
<td>Easier and less costly production than mAbs</td>
<td>Preclinical</td>
<td>LGT209</td>
<td>Phase I completed</td>
<td>Novartis</td>
</tr>
<tr>
<td>siRNA</td>
<td>Highly selective</td>
<td>Intravenous or subcutaneous administration</td>
<td>EGF-A peptide</td>
<td>Preclinical completed</td>
<td>Merck &amp; Co</td>
</tr>
<tr>
<td></td>
<td>Less selective</td>
<td>Greater likelihood of side effects</td>
<td>SX-PCK9</td>
<td>Phase I completed</td>
<td>Serometrix</td>
</tr>
<tr>
<td></td>
<td>Oral administration</td>
<td></td>
<td>ALN-PCS (intravenous)</td>
<td>Phase I recruiting</td>
<td>Alnylam Pharmaceuticals</td>
</tr>
<tr>
<td></td>
<td>Low cost</td>
<td></td>
<td>ALN-PCSsc (subcutaneous)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Small molecules</td>
<td>Oral administration</td>
<td>Less selective</td>
<td>SBC-1 and SBC-2</td>
<td>Preclinical completed</td>
<td>Shifa Biomedical Corp</td>
</tr>
</tbody>
</table>

EGF-A indicates epidermal growth factor precursor homology domain-A; FDA, US Food and Drug Administration; and mAb, monoclonal antibody.
LDL-R mutation, with 45% being true homozygotes and 47% being compound heterozygotes. All patients were on a background of statin therapy, with 94% on high-intensity atorvastatin or rosuvastatin and 92% on combination therapy with ezetimibe. After 12 weeks, evolocumab reduced LDL-C by 23% and apoB by 19%. LDL-C responses to evolocumab varied according to LDL-R function. Patients with defective mutations in 1 or both LDL-R alleles had a better response to treatment (mean reduction, 30%) compared with patients with at least 1 negative or null mutation (mean reduction, 21%). As expected, 1 subject with null mutations in both LDL-R alleles had no significant response to evolocumab.

Additionally, evolocumab was studied in 2 open-label, randomized trials (OSLER 1 and OSLER 2) of 4465 patients who had completed prior phase 2 or 3 trials of evolocumab.69 The patients were randomized to standard therapy plus evolocumab 140 mg every 2 weeks or 420 mg monthly or to standard therapy alone. After 12 weeks, treatment with evolocumab reduced LDL-C by 61% to a median level of 48 mg/dL. In a post hoc analysis, treatment with evolocumab was associated with a significantly lower rate of cardiovascular events at 1 year compared with standard therapy (0.95% and 2.18%, respectively; hazard ratio, 0.47; 95% confidence interval, 0.28–0.78; P=0.003).

### Phase II Trial of Bococizumab (Pfizer)

The clinical efficacy and safety of bococizumab have been reported in a published phase II clinical trial conducted in 354 hypercholesterolemic patients (LDL-C ≥80 mg/dL) on statin therapy.108 These patients were randomized to placebo; 50, 100, or 150 mg bococizumab every 2 weeks; or a regimen of 200 or 300 mg bococizumab every 4 weeks. The dose of bococizumab was reduced if LDL-C fell to ≤25 mg/dL. After 12 weeks, the 150-mg dose administered every 2 weeks and the 300-mg dose administered every 4 weeks were shown to lower LDL-C the most (53 and 45 mg/dL, respectively). The LDL-C–lowering effect of bococizumab was less than that predicted by pharmacokinetic/pharmacodynamic models because up to 44% of patients required mAb dose reductions owing to LDL-C levels ≤25 mg/dL. The frequency of adverse events was similar in the placebo and bococizumab groups.

Additional data from phase I,87,109,110 phase II111–118 and phase III trials119–123 of PCSK9 inhibitors are summarized in Tables 2 through 7.

### Table 2. Phase I Clinical Trials Summary

<table>
<thead>
<tr>
<th>Study</th>
<th>n</th>
<th>Population</th>
<th>Treatment</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>sRNA</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ALN-PCs</td>
<td>Fitzgerald et al,87 2014</td>
<td>32 Healthy adults</td>
<td>Placebo vs ALN-PCS 0.015–0.4 mg/kg once daily</td>
<td>ALN-PCS was associated with a dose-dependent reduction in plasma PCSK9 protein levels and decrease in LDL-C levels of 14%–46% from baseline. No reported drug-related adverse events.</td>
</tr>
<tr>
<td>mAbs</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alirocumab</td>
<td>Stein et al,109 2012</td>
<td>72 Healthy adults</td>
<td>Alirocumab 0.3 to 12 mg/kg IV once daily vs alirocumab 50–250 mg SC once daily</td>
<td>Intravenous alirocumab reduced LDL-C by 28% to 65%. Subcutaneous alirocumab reduced LDL-C by 33%–46%; SAEs reported in 2 subjects. In the intravenous arm, 1 person had abdominal pain and rectal bleeding and another reported a transient serum creatinine kinase of 10 times the upper limit of normal, likely caused by strenuous physical activity. In the subcutaneous arm, there were 1 report of small bowel obstruction in a patient with history of appendectomy and several reports of mild injection-site reaction.</td>
</tr>
<tr>
<td></td>
<td>Stein et al,109 2012</td>
<td>51 Hypercholesterolemia; on statin therapy</td>
<td>Multidose alirocumab 50, 100, or 150 mg SC 3 times daily on days 1, 29, and 43 plus atorvastatin</td>
<td>LDL-C reduced by 38%–65%; HDL-C increased by up to 18%; apoA1 increased by up to 13%; Lp(a) reduced by up to 27%.</td>
</tr>
<tr>
<td></td>
<td>Stein et al,109 2012</td>
<td>10 Hypercholesterolemia; no statin therapy</td>
<td>Multidose alirocumab 50, 100, or 150 mg SC 3 times daily on days 1, 29, 43</td>
<td>LDL-C reduced by 57% from baseline.</td>
</tr>
<tr>
<td>Evolocumab</td>
<td>Dias et al,110 2012</td>
<td>56 Healthy adults; hypercholesterolemia on statins</td>
<td>Evolocumab 7, 21, 70, 210, or 420 mg SC 1 time daily; evolocumab 21 or 420 mg IV 1 time daily; placebo</td>
<td>In healthy adults, dose-dependent reduction in LDL-C by up to 64% and reduction in apoB by up to 55%. In hypercholesterolemic patients on statin therapy, LDL-C reduced up to 81%, apoB by up to 59%, and Lp(a) by 27%–50%. No effects on HDL-C or triglycerides. Incidence of adverse events similar to placebo for both healthy and hypercholesterolemic cohorts.</td>
</tr>
</tbody>
</table>

ApoA1 indicates apolipoprotein A1; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; Lp(a), lipoprotein(a); mAb, monoclonal antibody; PCSK9, proprotein convertase subtilisin/kexin type 9; and SAEs, serious adverse events.
Ongoing Phase III trials

In addition to the already completed phase III safety and efficacy trials, there are 4 large ongoing outcomes trials evaluating the impact of PCSK9 inhibition on CVD end points (Table 8). The ODYSSEY Outcomes: Evaluation of Cardiovascular Outcomes After an Acute Coronary Syndrome During Treatment With Alirocumab SAR236553 (REGN727; ODYSSEY OUTCOMES) trial (http://www.clinicaltrials.gov; NCT01663402) will randomize to alirocumab every 2 weeks or placebo 18000 patients who have had recent hospitalization for acute myocardial infarction or unstable angina and are on a background of statin therapy. The primary outcome is time to first occurrence of CHD death, acute myocardial infarction, hospitalization for unstable angina, or ischemic stroke. The estimated study completion date is January 2018. The Further Cardiovascular Outcomes Research With PCSK9 Inhibition in Subjects With Elevated Risk (FOURIER) study (http://www.clinicaltrials.gov; CT01764633) will randomize an estimated 19000 patients with LDL-C ≥100 mg/dL or non–HDL-C ≥100 mg/dL to evolocumab or placebo every 2 or 4 weeks with atorvastatin with or without ezetimibe. The primary outcome is time to first occurrence of CVD death, nonfatal myocardial infarction, or hospitalization for unstable angina, stroke, or coronary revascularization. The estimated study completion date is February 2018. The Evaluation of Bococizumab (PF-04950615; RN316) in Reducing the Occurrence of Major Cardiovascular Events in High Risk Subjects (SPIRE)-1 (http://www.clinicaltrials.gov; NCT01975376) and SPIRE-2 (http://www.clinicaltrials.gov; NCT01975389) are evaluating the effect of bococizumab on CV outcomes in high-risk patients. SPIRE-1 will recruit an estimated 17000 patients with LDL-C of 70 to 100 mg/dL or non–HDL-C of 100 to 130 mg/dL on a background of lipid-lowering therapy, whereas SPIRE-2 will recruit an estimated 9000 patients with LDL-C ≥100 mg/dL or non–HDL-C ≥130 mg/dL. Patients in SPIRE 1 and SPIRE-2 will be randomized to bococizumab every 2 weeks or placebo. The primary outcome will be time to first occurrence of CVD death, nonfatal myocardial infarction, stroke, or

Table 3. Alirocumab Phase II Clinical Trials Summary

<table>
<thead>
<tr>
<th>Study</th>
<th>n</th>
<th>Population</th>
<th>Treatment</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Roth et al,111</td>
<td>92</td>
<td>Hypercholesterolemia; LDL-C ≥100 mg/dL with atorvastatin 10 mg/d</td>
<td>Alirocumab 150 mg every 2 wk plus atorvastatin 10 mg/d; alirocumab 150 mg every 2 wk plus atorvastatin 80 mg/d</td>
<td>LDL-C reduced by 73% with alirocumab plus atorvastatin 80 mg/d; all patients with alirocumab achieved LDL-C &lt;100 mg/dL; Lp(a) reduced by 35%</td>
</tr>
<tr>
<td>Mckenney et al,112</td>
<td>183</td>
<td>Hypercholesterolemia; LDL-C ≥100 mg/dL with atorvastatin 10–40 mg/d</td>
<td>Alirocumab 50, 100, or 150 mg every 2 wk; alirocumab 200 or 300 mg every 4 wk</td>
<td>LDL-C reduced by 40%–72% with alirocumab every 2 wk and 43%–48% with alirocumab every 4 wk; 89%–100% of patients reached LDL-C &lt;100 mg/dL; Lp(a) reduced up to 29%</td>
</tr>
<tr>
<td>Stein et al,113</td>
<td>77</td>
<td>Heterozygous familial hypercholesterolemia; LDL-C ≥100 mg/dL on statin with or without ezetimibe</td>
<td>Alirocumab 150, 200, or 300 mg every 4 wk; alirocumab 150 mg every 2 wk; placebo</td>
<td>LDL-C reduced by 29%–43% with alirocumab every 4 wk; LDL-C reduced by 68% with alirocumab every 2 wk; 81% patients treated with alirocumab every 2 wk reached LDL-C &lt;70 mg/dL</td>
</tr>
</tbody>
</table>

LDL-C indicates low-density lipoprotein cholesterol; and Lp(a), lipoprotein(a).

Table 4. Evolocumab Phase II Clinical Trials Summary

<table>
<thead>
<tr>
<th>Study</th>
<th>n</th>
<th>Population</th>
<th>Treatment</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>LAPLACE-TIMI 57114</td>
<td>631</td>
<td>Hypercholesterolemia; LDL-C ≥85 mg/dL with statin therapy with or without ezetimibe</td>
<td>Evolocumab 70, 105, or 140 mg every 2 wk; evolocumab 280, 350, or 420 mg every 4 wk; placebo</td>
<td>LDL-C reduced 42%–50% with dose every 2 wk; 94% of patients achieved LDL-C goal &lt;70 mg/dL with 140 mg every 2 wk; Lp(a) reduced 18%–32%</td>
</tr>
<tr>
<td>MENDEL115</td>
<td>406</td>
<td>Hypercholesterolemia; not on lipid therapy; LDL-C ≥100 mg/dL</td>
<td>Evolocumab 70, 105, or 140 mg every 2 wk; evolocumab 280, 350, or 420 mg every 4 wk; ezetimibe 10 mg/d; placebo</td>
<td>LDL-C reduced 41%–51% with dose every 2 wk and 39%–48% with dose every 4 wk; Lp(a) reduced 9%–7%; no difference in adverse events compared with placebo</td>
</tr>
<tr>
<td>RUTHERFORD116</td>
<td>168</td>
<td>Heterozygous familial hypercholesterolemia; LDL-C ≥100 mg/dL on lipid-lowering therapy</td>
<td>Evolocumab 350 or 420 mg every 4 wk; placebo</td>
<td>LDL-C reduced 43% with 350-mg dose and 55% with 420-mg dose; 89% patients achieved LDL-C goal &lt;100 mg/dL; no serious treatment-related adverse events</td>
</tr>
<tr>
<td>GAUSS117</td>
<td>236</td>
<td>Statin intolerance with muscle-related adverse events with statins</td>
<td>Evolocumab 280, 350, or 420 mg every 4 wk; evolocumab 430 mg every 4 wk plus ezetimibe 10 mg/d; placebo plus ezetimibe 10 mg/d</td>
<td>LDL-C reduced 41%–51% with evolocumab and 63% with evolocumab plus placebo; no difference in adverse events</td>
</tr>
</tbody>
</table>

GAUSS indicates Goal Achievement After Utilizing an Anti-PCSK9 Antibody in Statin-Intolerant Subjects; LAPLACE-TIMI 57, LDL-C Assessment with PCSK9 Monoclonal Antibody Inhibition Combined With Statin Therapy—Thrombolysis in Myocardial Infarction; LDL-C, low-density lipoprotein cholesterol; Lp(a), lipoprotein(a); MENDEL, Monoclonal Antibody Against PCSK9 to Reduce Elevated LDL-C in Subjects Currently Not Receiving Drug Therapy for Easing Lipid Levels; and RUTHERFORD, Reduction of LDL-C With PCSK9 Inhibition in Heterozygous Familial Hypercholesterolemia Disorder.
Table 5. Bococizumab Phase II Clinical Trials Summary

<table>
<thead>
<tr>
<th>Study</th>
<th>n</th>
<th>Population</th>
<th>Treatment</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gumbiner et al,114, 118 2012</td>
<td>135</td>
<td>Hypercholesterolemia; maximal doses of statin therapy</td>
<td>Bococizumab dose ranging from 0.25–6 mg/kg every 4 wk; placebo</td>
<td>LDL-C reduced 12%–58% with bococizumab; no adverse events related with bococizumab</td>
</tr>
<tr>
<td>Ballantyne et al,119, 120 2015</td>
<td>354</td>
<td>Hypercholesterolemia; LDL-C ≥80 on statin therapy</td>
<td>Bococizumab 50, 100, or 150 mg every 2 wk; bococizumab 200 or 300 mg every 4 wk; placebo</td>
<td>LDL-C reduced 53 mg/d with 150-mg dose every 2 wk; reported rates of adverse events were similar in bococizumab groups and placebo</td>
</tr>
</tbody>
</table>

LDL-C indicates low-density lipoprotein cholesterol.

In summary, data from phase I, II, and III clinical trials published to date have consistently demonstrated the ability of PCSK9 mAbs, alone and in combination with statin therapy, to lower

Table 6. Alirocumab Phase III Clinical Trials Summary

<table>
<thead>
<tr>
<th>Study</th>
<th>n</th>
<th>Population</th>
<th>Treatment</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>ODYSSEY MONO47</td>
<td>103</td>
<td>Hypercholesterolemia; not on lipid-lowering therapy; 10-y risk of CVD death of 1%–5% by European Systematic Coronary Risk Estimate</td>
<td>Alirocumab 75 mg every 2 wk titrated to 150 mg if LDL-C &gt;70 mg/dl; ezetimibe 10 mg/d</td>
<td>LDL-C reduced 47% vs 16% with ezetimibe; non–HDLC reduced 41%; no significant difference in Lp(a) compared with ezetimibe</td>
</tr>
<tr>
<td>ODYSSEY ALTERNATIVE28</td>
<td>314</td>
<td>Statin-intolerant patients who reported no muscle-related adverse events with trial of subcutaneous or oral placebo</td>
<td>Alirocumab 75 mg every 2 wk; ezetimibe 10 mg/d; atorvastatin 20 mg/d</td>
<td>LDL-C reduced 45% vs 15% with ezetimibe; lower rate of muscle-related events reported with alirocumab compared to atorvastatin</td>
</tr>
<tr>
<td>ODYSSEY OPTIONS I and II114,120</td>
<td>650</td>
<td>Hypercholesterolemia; high CVD risk; LDL-C ≥70 mg/dl with CVD history or DM with target-organ damage; LDL-C ≥100 mg/dl 10-y CVD risk SCORE ≥5%, moderate CKD, or DM without target-organ damage</td>
<td>Atorvastatin 20–40 mg/d (OPTIONS I) or rosuvastatin 10–20 mg/d (OPTIONS II) plus alirocumab 75 mg every 2 wk; doubling statin dose; switching atorvastatin 40 mg/d to rosuvastatin 40 mg/d</td>
<td>LDL-C reduced 44% and 54% in patients on atorvastatin 20 and 40 mg/d with addition of alirocumab; LDL-C reduced 51% and 36% in patients on rosuvastatin 10 and 20 mg/d with addition of alirocumab</td>
</tr>
<tr>
<td>ODYSSEY COMBO I and II100,101</td>
<td>1036</td>
<td>Hypercholesterolemia; maximally tolerated statin therapy with LDL-C ≥70 mg/dl with CVD or LDL-C ≥100 mg/dl with CV risk factors</td>
<td>Alirocumab 75 mg every 2 wk titrated to 150 mg if LDL-C &gt;70 mg/dl or placebo (COMBO I); alirocumab 75 mg every 2 wk titrated to 150 mg if LDL-C &gt;70 mg/dl or ezetimibe 10 mg/d (COMBO II)</td>
<td>LDL-C reduced 48% with evolocumab (COMBO I) and by 51% with evolocumab vs 21% with ezetimibe (COMBO II); no significant differences in treatment-related adverse events compared with placebo or ezetimibe</td>
</tr>
<tr>
<td>ODYSSEY FH (FH I, FH II, HIGH FH)101,122</td>
<td>841</td>
<td>Heterozygous FH; LDL-C ≥70 mg/dl with CVD or LDL-C ≥100 mg/dl without CVD (FH I and II) or LDL-C ≥160 mg/dl (HIGH FH)</td>
<td>Alirocumab 75 mg every 2 wk increased to 150 mg if &gt;70 mg/dl or placebo (FH I and II); alirocumab 150 mg every 2 wk or placebo</td>
<td>LDL-C reduced 49% (FH I and II) and 46% (HIGH FH); 41% of patients reaching LDL-C goal of ≤100 mg/dl for patients at high CV risk or ≤70 mg/dl for patients at very high CV risk; similar adverse events with alirocumab and placebo</td>
</tr>
<tr>
<td>CHOICE I and II123</td>
<td>1036</td>
<td>Hypercholesterolemia; elevated CVD risk on maximally tolerated statin dose, with muscle-related statin intolerance, or not on statin therapy (CHOICE I); elevated CV risk not on statin therapy with or without muscle-related statin intolerance (CHOICE II)</td>
<td>Alirocumab 300 mg every 4 wk, alirocumab 75 mg every 2 wk, or placebo (CHOICE I); alirocumab 150 mg every 4 wk, 75 mg every 2 wk, or placebo (CHOICE II); alirocumab was uptitrated if LDL-C ≥70 mg/dl or 100 mg/dl, depending on CV risk</td>
<td>LDL-C reduced 52% with alirocumab 300 mg every 4 wk in patients not on statin therapy and by 59% on statin therapy (CHOICE I); LDL-C reduced 56% with alirocumab 150 mg every 4 wk</td>
</tr>
<tr>
<td>ODYSSEY LONG TERM40</td>
<td>2341</td>
<td>Heterozygous FH with CHD or CHD equivalent with LDL-C ≥70 mg/dl on lipid therapy</td>
<td>Alirocumab 150 mg every 2 wk or placebo</td>
<td>LDL-C reduced with alirocumab by 52%; no difference in adverse events compared with placebo; post hoc analysis showed 48% relative risk reduction in CVD events</td>
</tr>
</tbody>
</table>

CHD indicates coronary heart disease; CKD, chronic kidney disease; CV, cardiovascular; CVD: cardiovascular disease; DM, diabetes mellitus; FH, familial hypercholesterolemia; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol; Lp(a), lipoprotein(a); ODYSSEY ALTERNATIVE, Study of Alirocumab (REGN727/SAR236553) in Patients With Primary Hypercholesterolemia and Moderate, High, or Very High Cardiovascular (CV) Risk, Who Are Intolerant to Statins; ODYSSEY COMBO, Efficacy and Safety of Alirocumab (SAR236553/REGN727) Versus Ezetimibe on Top of Statin in High Cardiovascular Risk Patients With Hypercholesterolemia; ODYSSEY LONG TERM, Long-Term Safety and Tolerability of Alirocumab (SAR236553/REGN727) Versus Placebo on Top of Lipid-Modifying Therapy in High Cardiovascular Risk Patients With Hypercholesterolemia; ODYSSEY MONO, Efficacy and Safety of Alirocumab SAR236553 (REGN727) Versus Ezetimibe in Patients With Hypercholesterolemia; and SCORE, systematic coronary risk evaluation.
LDL-C in a number of different populations. Treatment with PCSK9 mAbs has demonstrated an additional 50% to 60% reduction in LDL-C when used in combination with statins. Whereas post hoc analyses suggest the potential benefit of PCSK9 mAbs in reducing clinical events, outcome data from ongoing large, randomized trials are needed to definitively determine the role of PCSK9 mAbs in cholesterol management and CVD treatment.

Clinical Trial of siRNA to PCSK9

ALN-PCS (Alnylam Pharmaceuticals)

Another therapeutic approach has been the use of siRNAs to promote target gene mRNA degradation by RNA-induced silencing complexes. ALN-PCS is an siRNA that has been shown to inhibit PCSK9 synthesis after intravenous administration.124

Table 7. Evolocumab Phase III Clinical Trials Summary

<table>
<thead>
<tr>
<th>Study</th>
<th>n</th>
<th>Population</th>
<th>Treatment</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td>MENDEL-2</td>
<td>614</td>
<td>Hypercholesterolemia; LDL-C ≥100 mg/dL and &lt;190 mg/dL</td>
<td>Evolocumab 140 mg every 2 wk; evolocumab 420 mg monthly; ezetimibe 10 mg/d; placebo</td>
<td>LDL-C reduced 56% to 57% with evolocumab compared with 18%–19% with ezetimibe; no difference in reported adverse events</td>
</tr>
<tr>
<td>DESCARTES</td>
<td>901</td>
<td>Hypercholesterolemia; LDL-C ≥75 mg/dL on background of atorvastatin 10–80 mg/d with or without ezetimibe</td>
<td>Evolocumab 420 mg every 4 wk; placebo</td>
<td>LDL-C reduced 57% with evolocumab; 82% of patients were able to achieve LDL-C goal of &lt;70 mg/dL; no differences in adverse events</td>
</tr>
<tr>
<td>GAUSS</td>
<td>307</td>
<td>Hypercholesterolemia with statin intolerance to ≥2 statins owing to muscle-related side effects</td>
<td>Evolocumab 140 mg every 2 wk; 420 mg monthly; ezetimibe 10 mg/d; placebo</td>
<td>LDL-C reduced 55%–56% with evolocumab; apoB reduced 46% and Lp(a) reduced by 24%–26% with evolocumab; similar rate of adverse events</td>
</tr>
<tr>
<td>LAPLACE-2</td>
<td>2067</td>
<td>Hypercholesterolemia; LDL-C ≥150 mg/dL without statin therapy, LDL-C ≥100 mg/dL on nonintensive statin dose, or LDL-C ≥80 mg/dL on intensive statin dose</td>
<td>Evolocumab 120 mg every 2 wk; evolocumab 420 mg every month; ezetimibe 10 mg/d</td>
<td>Evolocumab reduced LDL-C 63%–75%, non–HDL-C 58%–65%, apoB 51%–59%, and Lp(a) 21%–36%; similar rates of adverse events</td>
</tr>
<tr>
<td>TESLA B</td>
<td>50</td>
<td>Homozygous FH on background of statin therapy; 92% with documented LDL-R mutation</td>
<td>Evolocumab 420 mg or placebo every 4 wk</td>
<td>Evolocumab reduced LDL-C 23%; LDL-C response to evolocumab varied according to LDL-R function</td>
</tr>
<tr>
<td>OSLER-1 and OSLER-2</td>
<td>4465</td>
<td>Subjects who completed phase 2 or 3 evolocumab studies</td>
<td>Standard therapy plus evolocumab (420 mg monthly or 140 mg every 2 wk) or standard therapy alone</td>
<td>Evolocumab reduced LDL-C by 61%; 53% reduction in cardiovascular events in post hoc analysis</td>
</tr>
</tbody>
</table>

apoB indicates apolipoprotein B; DESCARTES, Durable Effect of PCSK9 Antibody Compared With Placebo Study; GAUSS, Goal Achievement After Utilizing an Anti-PCSK9 Antibody in Statin-Intolerant Subjects; FH, familial hypercholesterolemia; LAPLACE, LDL-C Assessment with PCSK9 Monoclonal Antibody Inhibition Combined With Statin Therapy; LDL-C, low-density lipoprotein cholesterol; LDL-R, low-density lipoprotein receptor; Lp(a), lipoprotein(a); MENDEL, Monoclonal Antibody Against PCSK9 to Reduce Elevated LDL-C in Subjects Currently Not Receiving Drug Therapy for Easing Lipid Levels; non–HDL-C, non–high-density lipoprotein cholesterol; OSLER, Open-Label Study of Long-Term Evaluation Against LDL Cholesterol; and TESLA B, Trial Investigating PCSK9 Antibody in Subjects With LDL Receptor Abnormalities Part B.

Table 8. Ongoing Phase III Outcomes Trials

<table>
<thead>
<tr>
<th>Study</th>
<th>n</th>
<th>Population</th>
<th>Treatment</th>
<th>Primary Outcome</th>
<th>Expected Completion</th>
</tr>
</thead>
<tbody>
<tr>
<td>ODYSSEY OUTCOMES (NCT01663402)</td>
<td>18000</td>
<td>Hospitalization for acute MI or UA on statin therapy</td>
<td>Alirocumab every 2 wk; placebo</td>
<td>CHD death, acute MI, hospitalization for UA, or ischemic stroke; study completion</td>
<td>January 2018</td>
</tr>
<tr>
<td>FOURIER (NCT01764633)</td>
<td>27500</td>
<td>CVD with LDL-C ≥70 mg/dL or non–HDL-C ≥100 mg/dL</td>
<td>Evolocumab or placebo every 2 or 4 wk with atorvastatin with or without ezetimibe</td>
<td>CV-related death, nonfatal MI, or hospitalization for UA, stroke, or coronary revascularization</td>
<td>February 2018</td>
</tr>
<tr>
<td>SPIRE-1 and -2 (NCT01975376 and NCT01975389)</td>
<td>26000</td>
<td>High-risk patients; LDL-C 70–100 mg/dL or non–HDL-C 100–130 mg/dL on a background of lipid-lowering therapy (SPIRE-1); LDL-C ≥100 mg/dL or non–HDL-C ≥130 mg/dL (SPIRE-2)</td>
<td>Bococizumab every 2 wk or placebo</td>
<td>CV death, nonfatal MI, stroke, or hospitalization for UA needing urgent revascularization</td>
<td>March 2018 (SPIRE-1), June 2018 (SPIRE-2)</td>
</tr>
</tbody>
</table>

CHD indicates coronary heart disease; CV, cardiovascular; CVD, cardiovascular disease; FOURIER, Further Cardiovascular Outcomes Research With PCSK9 Inhibition in Subjects With Elevated Risk; LDL-C, low-density lipoprotein cholesterol; MI, myocardial infarction; non–HDL-C, non–high-density lipoprotein cholesterol; ODYSSEY OUTCOMES, ODYSSEY Outcomes: Evaluation of Cardiovascular Outcomes After an Acute Coronary Syndrome During Treatment With Alirocumab SAR236553 (REGN727); SPIRE, The Evaluation of Bococizumab (PF-04950615,RN316) in Reducing the Occurrence of Major Cardiovascular Events in High Risk Subjects; and UA, unstable angina.
and a subcutaneously administered form has been tested in a small phase I clinical trial.87 In this dose-escalation study in 32 healthy participants with LDL-C >3 mmol/L (116 mg/dL), a single dose of ALN-PCS (0.015–0.400 mg/kg) administered intravenously resulted in dose-dependent reductions in plasma PCSK9 and LDL-C levels, with the highest dose conferring 70% and 40% reductions in PCSK9 and LDL-C levels, respectively, an effect sustained for 2 to 3 weeks after administration. Overall, ALN-PCS was well tolerated. The incidence of treatment-related adverse events (including transient rash, hiccups, headache, and cold symptoms) was similar across active drug and placebo groups (ALN-PCS, 79% versus placebo, 88%). The maximum tolerable dose of ALN-PCS and its efficacy in high-risk and statin-resistant patient populations remain to be established, but this small study holds promise for therapeutic approaches that inhibit hepatic PCSK9 synthesis. Alnylam has recently initiated another phase I clinical trial testing subcutaneously administered ALN-PCS (http://www.clinicaltrials.gov; NCT02314442).

**Other Therapeutic Approaches to PCSK9 Inhibition: Preclinical Data**

**Approaches That Inhibit Interaction of PCSK9 With LDL-Rs**

**Mimetic Peptides**

Mimetic peptides are short sequences of amino acids that are designed to mimic the structure of a target protein in a manner that affects its biological activity. This approach has been used to modify the ability of PCSK9 to interact with LDL-Rs. As described above, the catalytic domain of PCSK9 binds to the EGF-A of LDL-R, directing the complex toward lysosomal degradation rather than recycling of LDL-R. Several small peptides inhibiting this interaction have been developed, including mimetic peptides to EGF-A, the catalytic domain of PCSK9, the prodomain of PCSK9, and the C-terminal domain of PCSK9. As therapeutic tools, small peptides are highly selective, relatively easy to synthesize and modify, and less costly to produce than antibodies. Similar to mAbs, however, mimetic peptides often require systemic delivery because of limited oral bioavailability.

**Mimetic Peptides to EGF-A.** Small peptides engineered to mimic the EGF-A domain have been reported to bind to the catalytic domain of PCSK9 and, through competitive inhibition, to limit the interaction between PCSK9 and LDL-R.14,127 Thus, a synthetic EGF-A mimetic peptide was shown to inhibit PCSK9-induced degradation of LDL-R in HepG2 cells in a dose-dependent manner.61 Another synthetic peptide mimicking the H306Y gain-of-function mutation in the EGF-A domain was shown to inhibit binding of PCSK9 to LDL-R and to successfully recover LDL-R expression in vitro.126 Pep2-8 is the smallest identified peptide that mimics the secondary structure of the EGF-A domain.126 It covers the catalytic domain of PCSK9, thereby hindering the interaction between PCSK9 and LDL-R. Through competitive inhibition, Pep2-8 effectively restored LDL-R recycling in HepG2 cells.14

**Other Mimetic Peptides Affecting PCSK9 Function.** Mimetic peptides targeting gain-of-function mutations in the catalytic domain of PCSK9 have recently been tested and found to restore LDL-R recycling in vitro.127 Three mimetic peptides derived from the prosegment of PCSK9 were shown to increase LDL-R levels in vitro, but their effects on LDL in vivo have not been investigated. Small mimetic peptides to annexin-A2, an endogenous natural antagonist that binds to the C-terminal domain of PCSK9, have also been proposed as a potential approach for PCSK9 inhibition. Finally, a small peptide that impedes normal PCSK9 folding (SX-PCK9, Serometrix), thus hindering its binding to LDL-R, is currently being studied.

There are no clinical trials testing the use of small peptides to inhibit PCSK9 at this time. However, the identification of peptides that interfere with PCSK9 function has advanced our understanding of LDL-R regulation and can provide the foundation for developing other small peptide or small molecule inhibitors in the future.

**Adnectins**

Adnectins, also known as monobodies, are a newly developed class of therapeutic proteins derived from the 10th extracellular type III domain of human fibronectin.133 Because they are smaller than mAbs, adnectins can easily be modified to bind to their target protein (eg, PCSK9) with high specificity and affinity by exchanging amino acids in β-sheet loops while preserving structural stability.134 BMS-962476 (Bristol-Myers Squibb/Adnexus), a PCSK9-binding adnectin, was designed to adhere to the LDL-R binding site of PCSK9 and to prevent PCSK9-induced degradation of LDL-R. In preclinical trials, a single intravenous dose of BMS-962476 (5 mg/kg) in cynomolgus monkeys reduced plasma PCSK9 levels to almost zero within 10 minutes, resulting in reductions in LDL-C (~51%) within 48 hours that persisted for nearly 3 weeks.134

**Approaches That Inhibit PCSK9 Synthesis**

**Antisense Oligonucleotides**

ASOs are short, single-stranded complementary sequences of nucleotides commonly used as gene-silencing agents. They inhibit protein synthesis by binding to the target mRNA directly, thereby preventing protein translation and reducing protein levels with high specificity both intracellularly and extracellularly. ASOs with hepatic targets have been studied extensively because of their effective delivery to the hepatocyte nucleus.135 The efficacy of ASOs for reducing LDL has previously been demonstrated with the US Food and Drug Administration–approved agent mipomersen, which inhibits the synthesis of apoB100.136 ASOs offer high specificity, but like mAbs, this therapeutic class is costly to produce and requires intravenous or subcutaneous routes of administration.

Two PCSK9 ASOs, BMS84421/ISIS394814 and SPC5001, were initially explored in preclinical trials and showed promising lipid-lowering effects with minimal safety concerns. Intraperitoneal injections of BMS84421/ISIS394814 twice weekly for 6 weeks in hyperlipidemic mice resulted in a 90% decrease in PCSK9 mRNA levels, a 2-fold increase in hepatic LDL-R levels, and a concomitant 53% and 38% reduction in total and LDL-C concentrations, respectively.137 A phase
I trial testing BMS-844421 (http://www.clinicaltrials.gov; NCT01082562) was terminated because of safety concerns,\textsuperscript{135} and focus was shifted to new-generation locked nucleic acid (LNA) ASOs.

LNAs are nucleic acid analogs that contain at least 1 monomer in locked conformation, resulting in a more stable conformation.\textsuperscript{138} The benefits of LNA-based therapeutics include higher binding affinity and specificity to the target mRNA and thus higher potency compared with longer oligonucleotides. LNA ASOs targeting PCSK9 were shown to be effective in binding mRNA in mice and nonhuman primates.\textsuperscript{139–141} In a study in mice, a single intravenous injection of the LNA ASO resulted in a dose-dependent reduction in PCSK9 mRNA and hepatic LDL-R protein levels with no evidence of liver injury compared with control mice injected with saline. Investigation of the duration of action of the LNA ASO showed that a 20-mg/kg IV dose resulted in a reduction in PCSK9 mRNA that remained significant for up to 16 days, returning to baseline levels by day 32. Increased hepatic LDL-R protein occurred within 24 hours and lasted >8 days.\textsuperscript{140} In a study in nonhuman primates, 4 weekly intravenous maintenance doses of LNA ASO resulted in an 85% reduction in mRNA and serum concentrations of PCSK9 and a 50% reduction in LDL-C.\textsuperscript{141}

SPC5001 (Santaris Pharma) is an ASO with locked RNA nucleotides on both ends of the DNA. The first phase I clinical trial testing the efficacy of this ASO (http://www.clinicaltrials.gov; NCT01350960), administered subcutaneously, was terminated for undisclosed reasons, although serious renal side effects were reported in several healthy study participants.\textsuperscript{142}

**Splice-Switching Oligonucleotides**

These agents act by splicing premRNA into inactive splice variants. Recently, the use of such a reagent directed at PCSK9 was shown to increase LDL-R levels in vitro.\textsuperscript{143}

**Small Molecules That Inhibit Autocatalytic Processing of PCSK9**

Because autocatalytic processing of PCSK9 is a necessary step for the secretion of the mature protein from the ER,\textsuperscript{42} intracellular inhibition of this process with small molecule inhibitors holds promise as a therapeutic strategy. Because of their small size, small molecule inhibitors are generally not as selective and have a greater potential for side effects than other therapeutic approaches, but they have the advantage of significantly lower production cost and an oral route of administration. However, the development of PCSK9-specific small molecule inhibitors remains a challenge because the site of interaction between the catalytic subunit of PCSK9 and the EGF-A domain of LDL-R is fairly flat and lacks pockets necessary for small molecules to bind.\textsuperscript{144} The development of fusion proteins that interact with the prosegment or the catalytic domain of the PCSK9/prosegment complex has been proposed as a viable approach for interference with PCSK9 processing and maturation. In a recent study, a recombinant fusion protein derived from the Fc portion of human IgG and containing the prosegment of PCSK9 was shown to bind directly to human PCSK9.\textsuperscript{145} When HEK293 human embryonic kidney epithelial cells and an HepG2 cell line that does not express PCSK9 were cotransfected with human PCSK9 and increasing ratios of the fusion protein, the PCSK9-mediated degradation of LDL-R was largely reversed by the fusion protein. In a separate series of experiments, in vitro coinubation of HepG2 cells with the fusion protein and extracellular PCSK9 significantly attenuated PCSK9-mediated LDL-R degradation, providing evidence that this fusion protein also interferes with the effect of PCSK9 on LDL-R at the extracellular level.\textsuperscript{145}

**Safety of PCSK9 Inhibitors**

Phase II and III clinical trials have shown no difference in serious adverse events between treatment with evolocumab, alirocumab, or bococizumab and placebo. In LAPLACE–Thrombolysis in Myocardial Infarction (LAPLACE-TIMI-57), a greater number of adverse events were reported with evolocumab (58%) than placebo (46%), with the most common events being nasopharyngitis, cough, and nausea.\textsuperscript{114} In the ODYSSEY ALTERNATIVE trial with statin-intolerant patients, treatment with alirocumab was associated with rates of skeletal muscle–related events similar to that for the statin group.\textsuperscript{99} Similarly, in GAUSS-2 with statin-intolerant patients, myalgia occurred in 8% of the evolocumab-treated patients and 18% in the ezetimibe-treated patients.\textsuperscript{105} In ODYSSEY LONG TERM, treatment with alirocumab for 78 weeks was associated with similar rates of adverse events with alirocumab (81%) and placebo (83%), but alirocumab had higher rates of injection-site reactions (5.9% versus 4.2%), myalgia (5.4% versus 2.9%), neurocognitive events (1.2% versus 0.5%), and ophthalmologic events (2.9% versus 1.9%).\textsuperscript{70} Similar to ODYSSEY LONG TERM, OSLER-1 and OSLER-2 reported a higher number of neurocognitive adverse events with evolocumab (0.9%) than placebo (0.3%).\textsuperscript{69} Of note, neurocognitive events in these trials were self-reported and not confirmed with a formal neurological evaluation. In ODYSSEY LONG TERM, 37% of patients had LDL-C <25 mg/dL at 2 consecutive measurements, and the rates of adverse events among these patients were similar to those in the overall alirocumab group.\textsuperscript{70} Although the trial data to date have demonstrated the relative tolerability of PCSK9 inhibitors, outcome data from the large, ongoing phase III trials are needed to evaluate their long-term safety. The need for long-term safety data was also highlighted in editorial comments by Goemann et al,\textsuperscript{102} who raised the concern that the similar rates of adverse events among evolocumab and standard therapy groups in the OSLER-1 and -2 trials may have been the result of patient selection bias.

**Regulatory Status**

mAbs are currently the most advanced and clinically documented approach to PCSK9 inhibition. Very recently, US Food and Drug Administration approval has been obtained for 2 PCSK9 mAbs for the treatment of severe hypercholesterolemia. Praluent (alirocumab), the first to be approved in this drug class in the United States, is “approved for use in addition to diet and maximally tolerated statin therapy in adult patients with heterozygous familial hypercholesterolemia or patients with clinical atherosclerotic cardiovascular disease such as heart attacks or strokes, who
require additional lowering of LDL cholesterol.”146 The recommended starting dose of Praluent is 75 mg subcutaneously administered every 2 weeks, with the possibility of increasing the dose to a maximum of 150 mg to achieve optimal LDL-C lowering. No dose adjustments are needed for patients with mild or moderate renal or hepatic impairment. Repatha (evolocumab) was first approved for use by the European Commission. Under European Commission regulations, it is indicated for the following:

in adults with primary hypercholesterolemia (heterozygous familial and nonfamilial) or mixed dyslipidemia, as adjunct to diet: i) in combination with a statin, or statin with other lipid-lowering therapies in patients unable to reach LDL-C goals with the maximum tolerated dose of a statin or ii) alone or in combination with other lipid-lowering therapies in patients who are statin-intolerant, or for whom a statin is contraindicated.147

For these indications, the recommended dose of Repatha, administered subcutaneously, is either 140 mg every 2 weeks or 420 mg once per month. Lastly, a 420-mg dose administered monthly is approved for use in combination with other lipid-lowering therapies in adults and adolescents ≥12 years of age with homozygous familial hypercholesterolemia and can be titrated to a maximum of 420 mg every 2 weeks for additional LDL-C lowering. In the United States, Repatha received US Food and Drug Administration approval in August 2015 for use “in addition to diet and maximally-tolerated statin therapy in adult patients with heterozygous familial hypercholesterolemia, homozygous familial hypercholesterolemia, or clinical atherosclerotic cardiovascular disease, such as heart attacks or strokes, who require additional lowering of LDL cholesterol.”148 The recommended dose of Repatha is 140 mg every 2 weeks or 420 mg once a month.

Summary and Future Directions

PCSK9 inhibitors have been demonstrated to result in substantial lowering of LDL levels, and preliminary data from trials of anti-PCSK9 mAbs in high-risk patients indicate that they yield the expected reduction in CVD events without major adverse effects. Results from ongoing trials will establish the efficacy and safety of PCSK9 inhibition in a variety of patient categories, including those at lower CVD risk and those with statin intolerance. In this regard, consideration may be given to evaluating genetic and other factors that may contribute to variation in clinical response to PCSK9 inhibitors. For example, a genome-wide association study recently identified a common single-nucleotide polymorphism in the WD repeat domain 52 gene (WDR52, rs1306441) that was associated with statin-induced PCSK9 levels and LDL-C response to statins,149 and the minor allele of this single-nucleotide polymorphism was subsequently reported to be associated with reduced LDL-C response to statins.150 Hence, carriers of this allele might be predicted to achieve greater benefit from the addition of an anti-PCSK9 drug to statin therapy. Moreover, the minor allele of rs688, a common synonymous-coding single-nucleotide polymorphism in the LDLR gene in the vicinity of the β-propeller region where PCSK9 binds, was recently shown to alter LDL-R intracellular distribution in a hepatoma cell line, presumably by altering LDL-R protein conformation, and this effect appeared to inhibit PCSK9-mediated reduction in LDL uptake as manifest by a reduced response to a PCSK9 mAb.151 Further studies of genetic influences on response to PCSK9 inhibition may identify new targets for therapeutic intervention on the basis of regulation and function of this key pathway of cholesterol metabolism.

Disclosures

Dr Krauss reports research grants from Sanofi/Regeneron and Quest Diagnostics and is a consultant to Merck and Quest Diagnostics. The other authors report no conflicts.

References


Proprotein Convertase Subtilisin/Kexin Type 9 Inhibition: A New Therapeutic Mechanism for Reducing Cardiovascular Disease Risk
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