European Atherosclerosis Journal
www.eathj.org
EAJ 2024;2:51-53
https://doi.org/10.56095/eaj.v3i2.75

Novel approaches to lipid lowering

Maurizio Averna
Department of Health Promotion, Mother and Child Care, Internal Medicine and Medical Specialties (PROMISE), University of Palermo, Palermo, Italy.
Institute of Biophysics (IBF), National Research Council (CNR), Palermo, Italy.

Keywords
Lipid-lowering; oral PCSK9 inhibitors; antisense oligonucleotide; small interfering RNA; PCSK9 gene editing

ABSTRACT

Lowering cardiovascular risk by reducing apoB-containing lipoproteins (primarily low-density lipoproteins, LDL) is the key step in cardiovascular prevention. Current treatments such as high-intensity statins, ezetimibe and proprotein convertase subtilisin/kexin type 9 (PCSK9) inhibitors are effective, but some patients still experience cardiovascular events due to residual risks determined by factors beyond LDL-cholesterol (LDL-C) levels, including triglyceride levels and inflammation. New approaches are currently under investigation to further reduce cardiovascular risk. These include next-generation CETP inhibitors such as obicetrapib, which lowers LDL-C and increases HDL-C without the side effects of earlier drugs. Oral PCSK9 inhibitors (MK-0616 and AZD0780) show promise, potentially overcoming economic barriers. Efforts to reduce Lp(a) include antisense oligonucleotides, siRNAs and assembly inhibitors like muvalaplin, all showing significant Lp(a) reduction. PCSK9 gene editing using CRISPR-Cas9 technology has shown dramatic cholesterol-lowering effects in preclinical studies and thus offers potential for the future. These new approaches could significantly advance cardiovascular risk management.

Received 23 August 2024 accepted 26 August 2024

Introduction

This review focuses on novel and future approaches to reduce the cardiovascular (CV) residual risk by reducing the apoB-containing lipoprotein levels. We assume that in the clinical practice a combination therapy based on the use of high-intensity statins, ezetimibe, bempedoic acid, monoclonal antibodies (mAbs) targeting proprotein convertase subtilisin/kexin type 9 (PCSK9) or inclisiran may reduce most of the cardiovascular risk associated with the apoB containing lipoproteins and may contribute to achieve the LDL-cholesterol (LDL-C) goal in the majority of individuals at high and very high risk. However, we can assume that a proportion of well-treated patients will develop new cardiovascular events. Many factors such as the genetic background, the pre-existing burden of disease and the residual risk attributable to triglycerides, inflammation, coagulation and platelets may explain this recurrence of events. We discuss some novel options to reduce the residual risk due to the two main apoB-containing lipoproteins, LDL-C and Lp(a), that are (**Figure 1**):

- the newest-generation CETP (cholesteryl ester transfer protein) inhibitor, obicetrapib;
- the oral PCSK9 inhibitors;
- the novel approaches to reduce Lp(a) plasma levels (antisense oligonucleotides-ASO; small interfering RNA-siRNA; assembly inhibition);
- the PCSK9 gene editing.

Obicetrapib

CETP is a glycoprotein which regulates the two-way exchange of cholesteryl esters and triglycerides from high-density lipoprotein (HDL) particles to low-density and very low-density lipoproteins (LDL and VLDL) and also the transfer of triglycerides from LDL and VLDL to HDL particles. The human genetic model of CETP deficiency has shown that mutation carriers have no CETP activity and very high levels of HDL-cholesterol (HDL-C). Epidemiological data demonstrated that HDL-C is inversely correlated with the cardiovascular risk. It was obvious to design trials with drugs known to increase HDL-C such as fibrates and niacin to demonstrate a reduction of cardiovascular events due to the increase in HDL-C plasma levels [1].

The early CETP inhibitors, torcetrapib, dalcetrapib and evacetrapib were tested in clinical trials but, despite a significant increase

Corresponding Author
Maurizio Averna: maurizio.averna@people.unipa.it

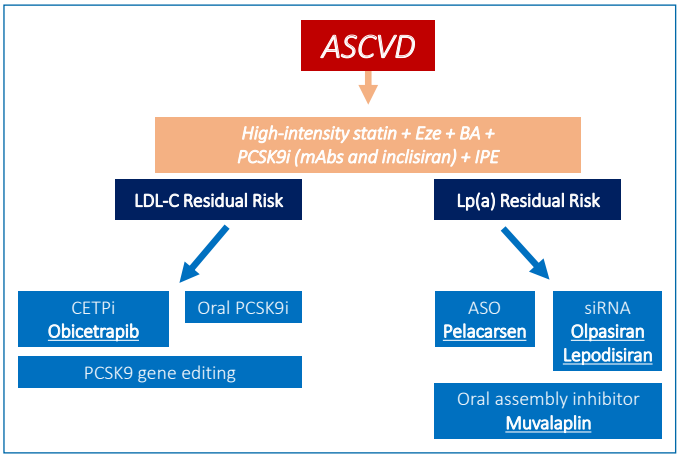

Figure 1 | Future approaches to apoB-containing lipoprotein-related residual risk. *ASCVD: atherosclerotic cardiovascular disease; Eze: ezetimibe; BA: bempedoic acid; PCSK9i: proprotein convertase subtilisin/kexin type 9 inhibitors; mAbs: monoclonal antibodies; IPE: icosapent ethyl; LDL-C: low-density lipoprotein cholesterol; Lp(a): lipoprotein (a); CETPi: cholesteryl ester transfer protein inhibitors; ASO: antisense oligonucleotide; siRNA: small interfering ribonucleic acid.*

in HDL-C (72%, 30% and 133%, respectively), torcetrapib increased cardiovascular events mainly for an off-target effect of blood pressure and dalcetrapib and evacetrapib trials were stopped for futility [2].

Anacetrapib, a CETP inhibitor, was tested in the REVEAL trial and showed a significant reduction in cardiovascular events attributable to the reduction in apoB-containing lipoproteins rather than the increase in HDL-C. The drug development was stopped because of the long-lasting accumulation of anacetrapib in the adipose tissue, but this trial paved the way for reversing the negative feelings regarding CETP inhibition as a cardiovascular prevention strategy [3]. In addition, recently the Mendelian randomization approach has shown that reducing LDL-C by CETP inhibition produces the same cardiovascular benefits as those achieved by statins, ezetimibe and PCSC9 inhibition, and there is also evidence of the ability of CETP inhibition to reduce the risk of new-onset diabetes since glucose tolerance and insulin sensitivity are improved [4]. Obicetrapib is the last-generation CETP inhibitor in the more advanced stage of development. The phase 1 and 2 studies have shown that obicetrapib significantly lowers all apoB-containing lipoproteins, including Lp(a) and the small LDL particles, and increases the levels of mature HDL as well as pre-beta HDL. In the phase 2 clinical trial ROSE2, obicetrapib in monotherapy or combination with ezetimibe reduced LDL-C by 43% and 63%, respectively, and increased HDL by 142%. Obicetrapib has been safe and well tolerated in thousands of patients enrolled in phase 1 to 3 clinical trials. None of the off-label or pharmacokinetic effects of torcetrapib and anacetrapib respectively have been seen in the thousands of patients enrolled in obicetrapib trials. The data from the outcomes study-PREVAIL will be known in 2026 and recently the results of the BROOKLIN trial have been released showing a significant reduction (41.5%) in LDL-C obtained in a difficult-to-treat population such as patients with heterozygous familial hypercholesterolemia (HeFH). In the ongoing TANDEM trial, the efficacy and safety of fixed-dose combination (FDC) of obicetrapib plus ezetimibe in adult patients with HeFH and/or atherosclerotic cardiovascular disease (ASCVD) or multiple risk factors for ASCVD are under evaluation [5]. Once approved for clinical use, obicetrapib will represent a novel LDL-receptor-independent approach to reduce LDL-C and cardiovascular risk.

Oral PCSK9 inhibition

The discovery of PCSK9 as a cause of familial hypercholesterolemia and the comprehension of its role in the LDL-receptor degradation pathway has opened a new era of LDL-C lowering pharmacology. Targeting PCSK9 by monoclonal antibodies, evolocumab and alirocumab, represents today a well-established therapeutic practice. Monoclonal antibodies to PCSK9 are safe, well tolerated and effectively reduce LDL-C up to 60%. The two outcomes trials, FOURIER and ODYSSEY Outcomes, have demonstrated significant reductions in cardiovascular events and mortality [6, 7]. In addition, the real-world data indicate a very high adherence and compliance. Recently the siRNA inclisiran, administered twice a year and targeting the PCSK9 gene expression in the liver, has been approved and entered into the clinical practice. However, some barriers are still limiting the use of PCSK9 mAbs, such as costs and prescription rules adopted in many countries. The RNA display screening technology led to the discovery of macrocyclic peptides that bind the PCSK9 with a monoclonal-like affinity. The two oral inhibitors in development are MK-0161 and AZD0780. MK-0161 has been evaluated in phase I and II trials. In the phase IIb trial, 381 adults with clinical ASCVD, intermediate/high ASCVD risk or borderline risk and LDL-C in a range between 70 mg/dL and 250 mg/dL according to the risk class were enrolled. The efficacy was dose-dependent: 41% LDL-C reduction with 6 mg daily, 56% LDL-C reduction with 12 mg daily, 59% LDL-C reduction with 18 mg daily, and 61% LDL-C reduction with 30 mg daily. The drug was safe and well tolerated. The results of the CORALreef Outcomes trial on cardiovascular benefits will be available at the end of 2029. AZD0780 is an oral small-molecule PCSK9 inhibitor; in the phase I trial the drug was administered on top of statin treatment and the efficacy results showed an LDL-C reduction of 52%. The development of oral, small molecules able to inhibit PCSK9 is promising and, if successful, would enrich our therapeutic armamentarium, potentially overcoming the economic issues and expanding the clinical settings of the PCSK9 targeting [8].

Novel approaches to reduce Lp(a)

Lp(a) was discovered 61 years ago by Kare Berg, but only in the last two decades a body of epidemiological and genetic studies have confirmed the role of Lp(a) as a cause of ASCVD and calcific aortic valve disease (CAVD). Lp(a) is the main carrier of oxidized phospholipids (oxPL) and plays a pivotal role in the residual CV risk; to date, lipid-lowering drugs are ineffective o poorly effective in reducing Lp(a) plasma levels. An ASO, a siRNA and a small molecule that interferes with the LDL-apo(a) binding are under development [9].

Antisense oligonucleotide-ASO

Pelacarsen is a N-acetylgalactosamine-conjugated oligonucleotide that was demonstrated in phase 1 and 2 studies to reduce Lp(a) plasma levels up to 80%, with up to 98% of patients achieving Lp(a) plasma levels <50 mg/dL [10]. The outcome trial, Lp(a) Horizon, was designed to measure the cardiovascular endpoints reduction in patients with myocardial infarction (MI) or ischemic stroke and clinically significant symptomatic peripheral artery disease and with entry-level of Lp(a) of 70 mg/dL. The endpoints-time to CV death, nonfatal MI, nonfatal stroke, and urgent coronary revascularization requiring hos-

pitalization - will be evaluated in patients with baseline Lp(a) ≥70 mg/dL and ≥90 mg/dL. The results are expected in 2025 [11].

Small interfering RNA-siRNA

Olpasiran and lepodisiran are two siRNAs that potently lower Lp(a), according to the results of the phase 1 trials. Olpasiran has reduced Lp(a) levels up to 98% in individuals with entry levels >75mg/dL. The drug was well tolerated and safe. Lepodisiran reduced Lp(a) levels up to 97% in subjects with entry levels >30 mg/dL and was also safe and well tolerated. The outcome trials to establish the clinical effectiveness of reducing Lp(a) are ongoing [9]. For olpasiran the results of the OCEAN study are expected in 2026; OCEAN has enrolled 7,000 patients with very high CV risk with an entry-level of Lp(a) ≥90 mg/dL and the endpoint is a 3-point coronary heart disease MACE (cardiovascular death, myocardial infarction, and coronary revascularization) [12]. Lepodisiran outcome trial, ACCLAIM-Lp(a), will enrol 12,500 patients with ASCVD or high-risk patients including HeFH patients and an Lp(a) entry level of 80 mg/dL. The main expected outcome is the reduction of CV mortality and the results are estimated to be delivered in 2029 [13].

Assembly inhibition

Lp(a) is assembled following a noncovalent interaction between apo(a) kringle 7 and 8 domains and lysine residues of apoB100 in the hepatocyte. Muvalaplin is a small molecule that inhibits the formation of Lp(a) by blocking the formation of the covalent disulfide bond [14]. In the phase 1 trial muvalaplin reduced Lp(a) plasma levels by 65%, with up to 93% of enrolled individuals achieving Lp(a) levels <50 mg/dL. The drug was safe and well tolerated. Muvalaplin could represent a valid alternative to the other more expensive Lp(a)-lowering drugs.

PCSK9 gene editing

The study of the mechanisms of DNA repair including the discovery of the enzymes involved opened the way to the concept of gene editing. A crucial step has been the discovery of the CRISPR-CAS9 as a gene-editing tool. As a DNA-editing tool, CRISPR-Cas9 can abolish or increase the function of a given gene. The potential of this methodology will lead to the cure for many genetic diseases. Since the adenovirus cannot be used to deliver the CRISPR-CAS9 apparatus because it is too large for the vector capacity, lipid nanoparticles with N-acetylgalactosamine (GalNAc) are currently used. GalNAc is a high-affinity ligand for the asialoglycoprotein receptor which is located only on the hepatocytes and this allows selective delivery to the liver. PCSK9 gene is a good candidate for a gene editing approach: i- carriers of loss-of-function mutations in the PCSK9 gene have very low levels of LDL-C and this marked reduction from birth translates into a dramatic reduction in cardiovascular diseases; ii- the clinical use of mAbs anti-PCSK9 resulted in an effective reduction in CV events. In the mouse model targeting the PCSK9 gene by CRISPR-CAS9 gene editing produced a ~95% decrease in plasma PCSK9 and a ~40% total cholesterol levels decrease [15]. In non-human primates, the improvement of gene editing technology produced a ~90% decrease in plasma PCSK9 levels and a parallel 60% reduction in LDL-C levels [16]. The preclinical results opened the way to clinical studies and the efficacy of the PCSK9 gene editing is currently tested in heterozygous familial hypercholesterolemia patients (trial NCT05398029 by VERVE Therapeutics). The preliminary results of the VERVE-101 trial on HeFH patients with severe ASCVD are promising, as a stable reduction in LDL-C up to 55% was observed after 6 months. However, many issues remain unanswered including the ethical aspects, the long-term safety and the right target disease-population.

References

[1] Nurmohamed NS, Ditmarsch M, Kastelein JJP. Cholesteryl ester transfer protein inhibitors: from high-density lipoprotein cholesterol to low-density lipoprotein cholesterol lowering agents? Cardiovasc Res 2022; 118:2919-31.
[2] Rehman WU, Yarkoni M, Ilyas MA, et al. Cholesteryl Ester Transfer Protein Inhibitors and Cardiovascular Outcomes: A Systematic Review and Meta-Analysis. J Cardiovasc Dev Dis 2024; 11:152.
[3] Kuhnast S, van der Tuin SJ, van der Hoorn JW, et al. Anacetrapib reduces progression of atherosclerosis, mainly by reducing non-HDL-cholesterol, improves lesion stability and adds to the beneficial effects of atorvastatin. Eur Heart J 2015; 36:39-48.
[4] Voight BF, Peloso GM, Orho-Melander M, et al. Plasma HDL cholesterol and risk of myocardial infarction: a mendelian randomisation study. Lancet 2012; 380:572-80.
[5] Kastelein JJP, Hsieh A, Dicklin MR, et al. Obicetrapib: Reversing the Tide of CETP Inhibitor Disappointments. Curr Atheroscler Rep 2024; 26:35-44.
[6] Sabatine MS, Giugliano RP, Keech AC, et al. Evolocumab and clinical outcomes in patients with cardiovascular disease. N Engl J Med 2017; 376:1713-22.
[7] Schwartz GG, Steg PG, Szarek M, et al. Alirocumab and cardiovascular outcomes after acute coronary syndrome. N Engl J Med 2018; 379:2097-107.
[8] Schonck WAM, Stroes ESG, Hovingh GK, Reeskamp LF. Long-Term Efficacy and Tolerability of PCSK9 Targeted Therapy: A Review of the Literature. Drugs 2024; 84:165-78.
[9] Tsimikas S. Lipoprotein(a) in the Year 2024: A Look Back and a Look Ahead. Arterioscler Thromb Vasc Biol 2024; 44:1485-90.
[10] Tsimikas S, Karwatowska-Prokopczuk E, Gouni-Berthold I, et al. Lipoprotein(a) Reduction in Persons with Cardiovascular Disease. N Engl J Med 2020; 382:244-55.
[11] Malick WA, Goonewardena SN, Koenig W, Rosenson RS. Clinical Trial Design for Lipoprotein(a)-Lowering Therapies: JACC Focus Seminar 2/3. J Am Coll Cardiol 2023; 81:1633-45.
[12] O'Donoghue ML, JA GL, Knusel B, et al. Study design and rationale for the Olpasiran trials of Cardiovascular Events And lipoproteiN(a) reduction-DOSE finding study (OCEAN(a)-DOSE). Am Heart J 2022; 251:61-9.
[13] Nissen SE, Linnebjerg H, Shen X, et al. Lepodisiran, an Extended-Duration Short Interfering RNA Targeting Lipoprotein(a): A Randomized Dose-Ascending Clinical Trial. JAMA 2023; 330:2075-83.
[14] Agarwala A, Asim R, Ballantyne CM. Oral PCSK9 Inhibitors. Curr Atheroscler Rep 2024; 26:147-52.
[15] Hoekstra M, Van Eck M. Gene Editing for the Treatment of Hypercholesterolemia. Curr Atheroscler Rep 2024; 26:139-46.
[16] Musunuru K, Chadwick AC, Mizoguchi T, et al. In vivo CRISPR base editing of PCSK9 durably lowers cholesterol in primates. Nature 2021; 593:429-34.