Review

Pharmacotherapy of Dyslipidemias

Lucas Lentini Herling de Oliveira , Arthur Cicupira Rodrigues de Assis , Viviane Zorzanelli Rocha Giraldez , Thiago Luis Scudeler * , Paulo Rogério Soares

Posted Date: 15 December 2023

doi: 10.20944/preprints202312.1195.v1

Keywords: dyslipidemias; cholesterol; statins; gene therapy

Review

Pharmacotherapy of Dyslipidemias

Lucas Lentini Herling de Oliveira, Arthur Cicupira Rodrigues de Assis, Viviane Zorzanelli Rocha Giraldez, Paulo Rogério Soares and Thiago Luis Scudeler *

Instituto do Coração (InCor), Hospital das Clínicas da Faculdade de Medicina da Universidade de São Paulo, São Paulo, Brazil, Av Dr Enéas de Carvalho Aguiar, 44, Cerqueira César, São Paulo 05403-000, Brazil; lucas.lentini@hc.fm.usp.br (L.L.H.d.O.); arthurcicupira@usp.br (A.C.R.d.A.); vzrocha@hotmail.com (V.Z.R.G.); paulo.soares@hc.fm.usp.br (P.R.S.)

* Correspondence: thiago.scudeler@fm.usp.br; Tel.: +55-11-26615299

Abstract: Hypercholesterolemia plays a fundamental role in the development and progression of atherosclerosis. Current guidelines for treating dyslipidemia target low-density lipoprotein-cholesterol (LDL-C). Despite advances in the pharmacotherapy of atherosclerosis, the most successful agents used to treat this disease – statins – remain insufficient in the primary or secondary prevention of acute myocardial infarction. Advancing therapy for hypercholesterolemia with emerging new drugs, either as monotherapy or in combination, is expected to improve cardiovascular outcomes.

Keywords: dyslipidemias; cholesterol; statins; gene therapy

1. Introduction

Cholesterol is a hydrophobic molecule, insoluble in plasma and with several vital functions in our body, such as the production of hormones and the formation of cell membranes. Since the discovery of cholesterol at the end of the 18th century, when it was isolated from gallstones, to its relationship with atherosclerosis, vast knowledge has been built about the molecule, its metabolism and its role in atherosclerosis.

Due to its insoluble nature, cholesterol is transported in plasma through lipoproteins, which are generally spherical structures, internally made up of nonpolar lipids, such as cholesterol esters and triglycerides, and externally by polar lipids such as phospholipids, apolipoprotein and free cholesterol [1]. On its surface we observe the presence of apolipoproteins (apo), which are fundamental structures for signaling, transport and binding of lipoproteins to receptors. Due to their amphiphilic nature (membrane-forming molecules), they are crucial in the stability and function of lipoproteins.

Evidence from epidemiological and clinical studies supports a key role of circulating LDL-C and other apolipoprotein B (apoB) containing lipoproteins in atherogenesis. Although the benefits of lipid lowering are well established in high-risk individuals, a number of trials show that the benefits extend also to lower-risk individuals. Knowledge of cholesterol metabolism is essential for understanding dyslipidemia and the drugs used in its treatment.

The aim of this article was to review the currently available therapies and emerging therapeutic agents for the management of patients with dyslipidemia, in light of recent evidence and guideline recommendations.

2. Methods

This narrative review was performed by searching Pubmed, Scopus, and Web of Science the following terms: hydroxymethylglutaryl-CoA (HMG-CoA) reductase inhibitors, proprotein convertase subtilisin/kexin type 9 (PCSK9) inhibitors, ezetimibe, bempedoic acid, inclisiran, angiopoietin-like 3 (ANGPTL3) inhibitors, inhibitors of apoB, Lp(a)-targeted therapies, microsomal triglyceride transfer protein (MTP) inhibitors, inducible degrader of LDL receptors (IDOL), bile acid-

 doi:10.20944/preprints202312.1195.v1

binding resins, nicotinic acid, fibric acid derivatives, cholesteryl ester transfer protein (CETP) inhibitors, gene therapy, vaccines against PCSK9, and plasmapheresis (Figure 1).

3. HMG-CoA Reductase Inhibitors

HMG-CoA reductase inhibitors, or statins, are the mainstay of LDL-C lowering therapy, and are currently recommended as first-line therapy for secondary prevention of atherosclerotic disease and for primary prevention of patients at-risk [2,3].

These drugs achieve LDL-C lowering by reducing cholesterol synthesis in the liver, which culminates in augmentation of LDL receptors (LDLR) in the hepatocytes. Increasing expression of LDLR in the surface of hepatocytes leads to increased LDL-C removal from the circulation [2].

Statins have a relatively predictable effect on LDL-C. Low-intensity statins (simvastatin 10 mg, pravastatin 10-20 mg) reduce LDL-C by less than 30%, moderate-intensity statins (simvastatin 20-40 mg, atorvastatin 10-20 mg, rosuvastatin 5-10 mg, pravastatin 40-80 mg) reduce LDL-C by 30-50% and high-intensity statins (atorvastatin 40-80 mg or rosuvastatin 20-40 mg) reduce LDL-C by at least 50%. These values reflect populational averages and may not be entirely applicable to individual patients [2,3].

Beyond LDL-C effects, statins also produce modest reductions in triglycerides levels and may lead to discrete increases in HDL-C, usually with a neutral effect on lipoprotein(a) [Lp(a)]. The classical "pleiotropic effects" of statins traditionally refer to potential anti-inflammatory and antioxidant effects of the drug [2].

In terms of clinical applications for statins, this review is divided into primary prevention, secondary prevention and special groups, such as heart failure (HF) and chronic kidney disease (CKD) patients. Main trials in these categories are summarized in Tables 1, 2 and 3.

3.1. Primary Prevention

In the primary prevention setting, any intervention aimed at reducing difficult outcomes such as mortality or myocardial infarction (MI) must involve large and/or long trials with sufficient power to detect differences in the inherently low event rate when compared to secondary prevention trials. Furthermore, the highest degree of scrutiny and critical reasoning is necessary before indicating an intervention to asymptomatic individuals, since its benefits tend to occur in the long term, while unaccounted adverse effects may derive from any kind of intervention.

One of the first and most relevant trials that rose to this challenge was the West of Scotland Coronary Prevention Study Group (WOSCOPS) trial [4]. In this study, pravastatin 40 mg was tested against placebo in patients with elevated LDL-C levels and no documented coronary artery disease (CAD). Over the 4.9 years of follow-up, LDL-C levels were reduced by an average of 26% and patients in the pravastatin group had lower rates of MI and coronary heart disease-related death.

Subsequently, the Air Force/Texas Coronary Atherosclerosis Prevention Study (AFCAPS/TexCAPS) evaluated the effect of cholesterol lowering with lovastatin in patients with moderately elevated lipid levels without clinically evident atherosclerotic cardiovascular disease (ASCVD) [5]. Lovastatin reduced the risk of the primary outcome of MI, unstable angina (UA), or sudden cardiac death (SCD). However, due to the low cardiovascular (CV) risk of the enrolled patients, the absolute risk reduction (ARR) of 0.2% per year was much smaller than demonstrated in previous trials (Number Needed to Treat [NNT] of 86). Furthermore, the study was stopped early for efficacy. Statistical simulations, however, suggest that truncated studies overestimate the magnitude of benefit of the treatment being evaluated by up to 29% [6].

Two other important studies that showed the CV benefits of statins in patients without documented ASCVD were ASCOT-LLA and MEGA.

The Anglo-Scandinavian Cardiac Outcomes Trial - Lipid Lowering Arm (ASCOT-LLA) trial showed that among patients with hypertension and relatively low cholesterol, treatment with atorvastatin was associated with a reduction in the primary endpoint of MI or coronary death at 3-year follow-up [7].

 doi:10.20944/preprints202312.1195.v1

The Management of Elevated Cholesterol in the Primary Prevention Group of Adult Japanese (MEGA) trial showed that treatment with pravastatin in addition to diet modification was associated with a reduction in coronary heart disease events compared with diet modification alone at a mean 5.3-year follow-up [8].

Despite the evidence provided by the WOSCOPS study, concerns have arisen regarding patients with lower LDL-C levels but with an estimated risk of ASCVD. Thus, markers capable of detecting patients who may benefit from statin therapy for primary prevention have been investigated. The Justification for the Use of Statins in Prevention: an Intervention Trial Evaluating Rosuvastatin (JUPITER) trial compared rosuvastatin versus placebo in patients with LDL-C <130 mg/dL, no known ASCVD and high-sensitivity C-reactive protein (hsCRP) levels of 2.0 mg per liter or higher [9]. The study showed a reduction in the primary composite endpoint (MI, stroke, arterial revascularization, hospitalization for UA, or CV death) [HR 0.56, 95% CI 0.46 to 0.69, ARR of 0.59% per year for the primary endpoint, NNT 169]. For coronary events, including fatal or non-fatal myocardial infarction, 500 persons need to be treated for one year to prevent one event. In addition to the low effect estimate demonstrated by rosuvastatin in the JUPITER study, methodological concerns have limited the widespread application of the results of this trial. First, the trial was truncated, which is known to overestimate the benefits of the intervention [6]. A 5-year follow-up was planned, but a median of only 1.9 years of follow-up was achieved. Regarding the use of hsCRP, it was not possible to determine whether patients benefited from rosuvastatin because they had high levels of hsCRP, since all patients had similar levels, and the study was not designed to test the strategy of using hsCRP as a marker for statin initiation versus standard care nor multivariate models to evaluate the impact of hsCRP were reported. The benefit found may have derived, for example, from the reduction of LDL-C in patients with high CV risk related to uncontrolled hypertension (the upper quartile of blood pressure was 145/87 mmHg).

Subsequently, the Heart Outcomes Prevention Evaluation (HOPE)-3 trial was published, shedding a little more light into this scenario. Patients of intermediate risk (estimated annual rate of major adverse cardiovascular events [MACE] ~1%) were randomly assigned to either rosuvastatin 10 mg or placebo, in a 2 x 2 factorial design with blood pressure lowering agents (candesartan and hydrochlorothiazide) [10]. There was not a specific LDL-C threshold for eligibility. The rosuvastatin group had a reduction in the composite coprimary endpoint of CV death, MI or stroke (HR 0.76, 95% CI 0.64 to 0.91, NNT of 91).

Despite the cardiovascular benefits suggested by the aforementioned trials, it is important not to let go of clinical reasoning and recognize the magnitude of the findings. Due to the inherent low incidence of events in the primary prevention population, it is important to acknowledge that the clinical benefit is marginal in the follow-up period of the trials, with high NNTs. Also, older patients comprise most of these study populations. The long-term benefit is probably greater than found in the aforementioned trials, and lifelong LDL-C lowering therapies might have an impact when considering younger patients with high LDL-C levels or higher than average CV risk factors.

Table 1. Primary prevention trials. Legend: CAD: coronary artery disease; CV: cardiovascular; hsCRP: high sensitivity C-reactive protein; MI: myocardial infarction; UA: unstable angina; TC: total cholesterol.

Study	Sample size	Characteristics of patients	Comparison groups	Follow-up	LDL effect	CV effects
WOSCOPS (1995) [4]	6,595	TC > 252 mg/dL	Pravastatin 40 mg vs. placebo	4.9 years	26%	MI or coronary death (HR 0.69, 95% CI 0.57 to 0.83, NNT 111)
AFCAPS / TexCAPS	6,605	LDL-C 130-180 mg/dL	Lovastatin 20-40 mg vs.	5.3 years	25%	major coronary events (HR 0.63, 95%

 doi:10.20944/preprints202312.1195.v1

(1998) [5]			placebo			CI 0.50 to 0.79, NNT 86)
ASCOT-LLA (2003) [7]	10,305	Hypertension and CV risk factors	Atorvastatin 10 mg vs. placebo	3.3 years	35%	MI or coronary death (HR 0.64, 95% CI 0.50 to 0.83, NNT 83)
MEGA (2006) [8]	7,832	TC 220-270 mg/dL	Pravastatin 10 mg vs. placebo	5.3 years	18%	CAD (HR 0.67, 95% CI 0.49 to 0.91, NNT 119)
JUPITER (2008) [9]	17,802	LDL-C <130 mg/dL + hsCRP ≥2 mg/L	Rosuvastatin 20 mg vs. placebo	1.9 years	50%	CV death, MI, stroke, arterial revascularization, or UA hospitalization (HR 0.56, 95% CI 0.46 to 0.69, NNT 169)
HOPE-3 (2016) [10]	12,705	Intermediate CV risk (CV event rate 1%/year)	Rosuvastatin 10 mg vs. placebo	5.6 years	26.5%	CV death, MI and stroke (HR 0.76, 95% CI 0.64 to 91, NNT 91)

3.2. Secondary Prevention

The consistent relative risk reduction (RRR) of MACE points towards a solid relationship between statin use and lower ratio of events. However, the clinical significance of such reduction will depend on the absolute rate of events, with more evident benefit seen in patients at the highest risk of MACE. This is found in secondary prevention clinical studies.

The Scandinavian Simvastatin Survival Study (4S) trial randomized 4,444 patients with coronary artery disease (CAD), defined by prior MI or angina, to simvastatin or placebo [11]. The trial was stopped early due to an ARR of 3.3% in all-cause mortality with simvastatin (11.5% vs. 8.2%; p=0.0003; NNT 30). In addition to being a truncated study, the low rate of aspirin use among the 4S trial population (~37%) draws attention. Possibly the magnitude of the benefit would be attenuated with widespread aspirin use. Also, the rate of patients with previous MI was high (79%), which configures a higher risk secondary prevention population, and benefits can be attenuated in a CAD population with no prior history of MI.

The Cholesterol and Recurrent Events (CARE) trial confirmed reduction of coronary events in patients with previous MI, even in a group with lower total cholesterol levels (<240 mg/dL, mean LDL-C of 139) [12] Similarly, the Long-term Intervention with Pravastatin in Ischaemic Disease (LIPID) trial showed a reduction in coronary death in patients with previous MI or hospitalization for UA (NNT of 53) [13]. The Heart Protection Study (HPS) showed a reduction in all-cause mortality, driven by vascular causes (7.6% vs. 9.1%, HR 0.83, 95% CI 0.75 to 0.91, NNT 67) in high-risk patients - 65% with previous CAD [13]. On the other hand, the FLuvastatin On RIsk Diminishing after Acute myocardial infarction (FLORIDA) trial showed that fluvastatin did not reduce coronary events in post-MI patients [15]. However, the trial was underpowered and a post hoc analysis revealed a trend towards a reduction in the primary endpoint in patients with pronounced ischaemia at trial onset [16].

So far, the relationship between intervention and outcome seems to be established, naturally with greater benefits seen in patients at higher baseline risk. However, new questions arose - What is the best statin regimen for reducing cardiovascular outcomes?

 doi:10.20944/preprints202312.1195.v1

The Pravastatin or Atorvastatin Evaluation and Infection Therapy-Thrombolysis in Myocardial Infarction 22 (PROVE IT-TIMI 22) trial tested different intensity regimens in patients in the acute phase of a MI [17]. Among those up to 10 days after an acute event, atorvastatin 80 mg reduced a composite of death from any cause, MI, documented UA requiring rehospitalization, revascularization after 30 days of randomization or stroke when compared to pravastatin 40 mg (22.4% vs 26.3%, HR 0.84, 95% CI 0.74 to 0.95). However, it is important to note that this endpoint is very broad and includes more fragile outcomes such as unstable angina and need for revascularization. Moreover, reductions in LDL-C levels in the pravastatin group were strikingly low (baseline LDL-C 106 mg/dL and LDL-C achieved at follow-up 95 mg/dL), which surely favors the atorvastatin group, which achieved LDL-C of 62 mg/dL on follow-up (41% reduction). This low LDL-C reduction in LDL-C is in part explained by the 25% of participants that were already on statin therapy and did not have their LDL-C levels changed by pravastatin use and had an additional 32% reduction with atorvastatin. In the 75% of patients who were not already on a statin, atorvastatin reduced LDL-C by 51% in 30 days and pravastatin reduced LDL-C by 22%.

The Incremental Decrease in Endpoints Through Aggressive Lipid Lowering (IDEAL) trial tested atorvastatin 80 mg versus simvastatin 20 mg and found no difference in the primary endpoint of coronary death, non-fatal MI or cardiac arrest with resuscitation (HR 0.89, 95% CI 0.78 to 1.01) [18]. One limitation of this trial was the smaller than expected reductions in LDL-C levels, which were around 34% with atorvastatin 80 mg and 17.7% in the simvastatin 20 mg group, possibly blunting an eventual difference in effects, but this is merely speculatory.

The Treating to New Targets (TNT) trial tested different regimens of atorvastatin (80 mg versus 10 mg, a high versus a moderate intensity regimen) in patients with stable CAD [19]. This trial found a reduction in coronary death, nonfatal non-procedural MI, resuscitation after cardiac arrest or stroke (8.7% vs 10.9%, NNT of 46). The primary endpoint was mainly driven by MI and stroke.

Finally, the Study of the Effectiveness of Additional Reductions in Cholesterol and Homocysteine (SEARCH) trial tested different simvastatin doses (20 mg versus 80 mg) and found no difference in efficacy outcomes, yet with a higher incidence of myopathy [20]. Thus, simvastatin 80 mg should not be used.

Based on findings from randomized clinical trials (RCTs), it appears that high intensity regimens lead to a modest benefit in patients with CAD, even with "normal" LDL-C levels, mainly driven by reduction in cardiac events but without clear mortality benefits.

Table 2. Secondary prevention trials. * Included both primary and secondary prevention patients. Legend: ACS: acute coronary syndrome; CA: cardiac arrest; CAD: coronary artery disease; HTN: hypertension; MI: myocardial infarction; TC: total cholesterol; UA: unstable angina.

Study	Sample size	Characteristics of patients	Comparison groups	Follow-up	LDL effect	CV effects
4S (1994) [11]	4,444	Angina or previous MI	Simvastatin 20-40 mg vs. placebo	5.4 years	35%	mortality (HR 0.70, 95% CI 0.58 to 0.85, NNT 30)
CARE (1996) [12]	4,159	Previous MI TC < 240 mg/dL LDL-C 115-174 mg/dL	Pravastatin 40 mg vs. placebo	5 years	28%	coronary death or MI (10.2% vs. 13.2%, NNT 34)
LIPID (1998) [13]	9,014	Previous MI or UA TC 155-271 mg/dL	Pravastatin 40 mg vs. placebo	6.1 years	25%	coronary death (6.4% vs. 8.3%, NNT 53)
FLORIDA	540	MI	Fluvastatin	1 year	21%	No significant

 doi:10.20944/preprints202312.1195.v1

(2000) [15]			80 mg vs. placebo			difference in the incidence of major coronary event
HPS* (2002) [14]	20,536	TC > 135 mg/dL + CAD or other arterial disease or diabetes or > 65y male w/ HTN	Simvastatin 40 mg vs. placebo	5 years	35%	all-cause mortality (12.9% vs. 14,7%, NNT 56)
PROVE-IT (2004) [17]	4,162	ACS < 10 days	Atorvastatin 80 mg vs. pravastatin 40 mg	24 months	31%	all-cause mortality, MI, UA hospitalization, revascularization in 30 days, or stroke (HR 0.84, 95% CI 0.74 to 0.95, NNT 53)
IDEAL (2005) [18]	8,888	Previous MI	Atorvastatin 80 mg vs. simvastatin 20 mg	4.8 years	20%	No significant difference in the incidence of major coronary event
TNT (2005) [19]	10,001	CAD	Atorvastatin 80 mg vs. atorvastatin 10 mg	4.9 years	24%	CV death, MI, CA, or stroke (HR 0.78, 95% CI 0.69 to 0.89, NNT 45)
SEARCH (2010) [20]	12,064	Previous MI LDL-C >135 mg/dL (statin use) or LDL-C >193 mg/dL (no statin)	Simvastatin 20 mg vs. simvastatin 80 mg	6.7 years	14%	No significant difference in the incidence of CV events

3.3. Special groups

Despite growing evidence solidifying statins as the cornerstone in LDL-C lowering therapy, the possibility has been raised that specific groups could have clinical benefits from statins.

The Collaborative Atorvastatin Diabetes Study (CARDS) trial tested atorvastatin 10 mg versus placebo for primary prevention in patients with diabetes and at least one additional risk factor (retinopathy, albuminuria, current smoking or hypertension), with LDL-C <160 mg/dL [21]. Although

 doi:10.20944/preprints202312.1195.v1

truncated (terminated 2 years earlier due to prespecified efficacy criteria), this trial showed a reduction in CV events (9.0% vs. 3.2%, NNT 32).

The Atorvastatin Study for Prevention of Coronary Heart Disease Endpoints in Non-Insulin-Dependent Diabetes Mellitus (ASPEN) trial randomized patients with diabetes in primary (79%) and secondary prevention (21%) to receive atorvastatin 10 mg or placebo and found no difference in the primary endpoint of CV death, MI, stroke, revascularization, worsening UA requiring hospitalization or resuscitated cardiac arrest [22]. These results can be explained by important steering disturbances during the trial. A widespread recognition of the importance of CV prevention in diabetes patients led to a recommendation to stop the study medications and allocate all secondary prevention patients and previously primary prevention patients that now met an endpoint to receive usual care. This led to a very low completion rate, and only 67% of the intervention and 58% of the placebo group were receiving the study medication at the end of the double-blind follow-up. This reduces the power to detect differences and hinders the evaluation of the results; care should be taken when using these findings.

Subanalyses of the HPS and ASCOT-LLA trials with diabetic patients showed that statin reduces CV events [23,24].

In patients with chronic kidney disease (CKD), several trials have evaluated the CV effects of statins. The Assessment of Lescol in Renal Transplantation (ALERT) trial showed that among renal transplant patients, treatment with fluvastatin was not associated with a reduction in the primary composite endpoint of cardiac death, MI, or coronary intervention procedure during 5.1 years of follow-up [25].

The Die Deutsche Diabetes Dialyse Study (4D) also showed no reduction in the composite primary endpoint of death from cardiac causes, MI or stroke with atorvastatin treatment in patients with type 2 diabetes undergoing routine hemodialysis [26]. Similarly, the Study to Evaluate the Use of Rosuvastatin in Subjects on Regular Hemodialysis: An Assessment of Survival and Cardiovascular Events (AURORA) trial found no benefit in 2,776 CKD patients on dialysis who were randomized to rosuvastatin 10 mg or placebo [27].

On the other hand, the Study of Heart and Renal Protection (SHARP) showed that ezetimibe/simvastatin compared to placebo reduces LDL-C and atherosclerotic and major vascular events in patients with CAD, but no overt CAD (11.3% versus 13.4%, HR 0.83, 95% CI 0.74 to 0.94) [28].

Thus, CKD patients not on dialysis might benefit from statins, with a modest impact, while CKD patients on dialysis do not seem to benefit from this therapy.

Patients with heart failure (HF) are another group of interest, and have been frequently excluded from statin trials. The Controlled Rosuvastatin Multinational Trial in Heart Failure (CORONA) trial showed that therapy with rosuvastatin was not associated with a reduction in the primary endpoint of CV death, MI, or stroke at a median follow-up of 32.8 months compared with placebo [29]. Additionally, the Gruppo Italiano per lo Studio della Sopravvivenza nell'Infarto Miocardico-Heart Failure (GISSI-HF) trial showed that rosuvastatin 10 mg daily is not beneficial in reducing cardiac outcomes among patients with chronic symptomatic HF [30].

The elderly population was exclusively studied in the Prospective Study of Pravastatin in the Elderly at Risk (PROSPER) trial [31]. This study showed a reduction in the composite primary outcome of coronary death, MI and stroke (14.1% versus 16.2%, HR 0.85, 95% CI 0.74 to 0.97, NNT 48) in the statin group.

Persons with HIV infection, a group with increased cardiovascular risk, were also analyzed in a recent phase 3 trial. In the Randomized Trial to Prevent Vascular Events in HIV (REPRIEVE) study, 7,769 participants with HIV infection were randomized to daily pitavastatin at a dose of 4 mg or placebo. After a median follow-up of 5.1 years, the study was interrupted due to efficacy. The rate of MACE was 4.81 and 7.32 per 1000 person-years in the pitavastatin and placebo groups, respectively (HR 0.65, 95% CI 0.48 to 0.90, p=0.002). Muscle-related symptoms and incident diabetes were more common in the pitavastatin group [32].

 doi:10.20944/preprints202312.1195.v1

Finally, a meta-analysis of data from 170,000 patients, evaluated statin versus placebo and different statin regimens and reported a 10% reduction in all-cause mortality per 38 mg/dL LDL-C reduction (HR 0.90, 95% CI 0.87 to 0.93) [33]. In an unweighted analysis of the 21 placebo-controlled trials included, any major vascular event occurred in 3.6% in the placebo groups versus 2.8% in the statin groups. translating into a 0.8% ARR and a 22% RRR. Regarding higher versus lower intensity regimens, higher intensity regimens led to reductions in major vascular events (RRR 15%, 95% CI 11 to 18), especially when weighted for LDL-C reductions (RRR per 1 mmol/L reduction in LDL-C), suggesting that greater reductions in LDL-C accompany greater reductions in MACE.

The evidence presented so far establishes the relationship between intervention and effect, and statins reduce CV events. The magnitude of benefits should however always permeate clinical reasoning, and NNTs ranging from 30 to a lot higher numbers have been found. The higher the baseline risk, the greater benefit that should be expected from statins. There is a logical chain binding CV risk factors. CV disease and death. Treating one will probably affect the other, but with progressively smaller magnitude. On the other hand, benefits in the above discussed trials seem to increase over time, which is expected since risk factors may be lifelong cumulative. Trials tend to follow patients over the course of a few years. and potential long-term benefits should be taken into account. The concepts proven with the studies above should be used as tools to individualized decision-making in clinical practice.

Table 3. Statins in special groups. Legend: ACS: acute coronary syndrome; CHD: coronary heart disease; CKD: chronic kidney disease; CV: cardiovascular; LVEF: left ventricular ejection fraction; MACE: major cardiovascular events; MI: myocardial infarction; NYHA: New York Heart Association; TG: triglycerides.

Study	Sample size	Characteristics of patients	Comparison groups	Follow-up	LDL effect	CV effects
CARDS (2004) [21]	2,838	Diabetes (40-75y) + LDL-C <160 mg/dL + TG <600 mg/dL + additional risk factor	Atorvastatin 10 mg vs. placebo	3.9 years	40%	ACS, additional revascularization, or stroke (HR 0.63, 95% CI 0.48 to 0.83, NNT 31)
ASPEN (2006) [22]	2,410	Diabetes (40-75y) + LDL <160 mg/dL or <140 mg/dL (prior MI or revascularization)	Atorvastatin 10 mg vs. placebo	4 years	29%	No significant difference in the incidence of CV events
ALERT (2003) [25]	2,102	Renal or combined renal and pancreas transplants >6 months	Fluvastatin 40 mg vs. placebo	5.1 years	25%	No significant difference in the incidence of CV events
4D (2005) [26]	1,255	Diabetes + CKD on dialysis	Atorvastatin 20 mg vs. placebo	4 years	42%	No significant difference in the incidence of CV events
AURORA	2,773	CKD on dialysis	Rosuvastatin	3.8 years	43%	No significant

 doi:10.20944/preprints202312.1195.v1

(2009) [27]			10 mg vs. placebo			difference in the incidence of CV events
SHARP (2011) [28]	9,270	CKD	Simvastatin 20 mg + ezetimibe 10 mg vs. placebo	4.9 years	31%	coronary death, MI, stroke, or revascularization (HR 0.83, 95% CI 0.74 to 0.94, NNT 53)
CORONA (2007) [29]	5,011	LVEF < 40% + NYHA II-IV	Rosuvastatin 10 mg vs. placebo	2.7 years	45%	No significant difference in the incidence of CV events
GISSI-HF (2008) [30]	6,975	Heart failure NYHA II-IV	Rosuvastatin 10 mg vs. placebo	3.9 years	16%	No significant difference in the incidence of CV events
PROSPER (2002) [31]	5,804	Elderly (70-82 years) + high CV risk	Pravastatin 40 mg vs. placebo	3.2 years	34%	coronary death, MI, or stroke (HR 0.85. 95% CI 0.74 to 0.97. NNT 48)
REPRIEVE (2023) [32]	7,769	HIV	Pitavastatin 4 mg vs. placebo	5.1 years	30%	MACE (HR 0.65, 95% CI 0.48 to 0.90)

4. PCSK9 inhibitors

PCSK9 inhibitors comprise a relatively new drug class that achieves LDL-C lowering and have been used on top of statins and ezetimibe in patients at very high CV risk (mainly secondary prevention and familial hypercholesterolemia with additional risk factors) who have not achieved prespecified target LDL-C levels [2].

PCSK9 is a protein that binds on LDLR in the hepatocytes and culminates in degradation of the receptor, which in turn leads to lesser LDL-C removal from the circulation. PCSK9 inhibitors bind to this protein and therefore augments LDLR levels and promotes LDL-C lowering [34]. Since its action depends on the existence of LDLR, receptor-deficient homozygous familial hypercholesterolemia patients do not respond well to this drug class [2]. These drugs are injectable monoclonal antibodies and usually have a long half-life, allowing for periodic administration. Available PCSK9 inhibitors are alirocumab and evolocumab, which may be administered every two weeks or even monthly.

These drugs greatly reduce LDL-C levels, on average by 60%, independently if used alone or on top of other drug classes. Similarly to statins, PCSK9 inhibitors may also reduce triglycerides and promote a slight raise in HDL-C levels [2].

However, an additional interesting response to PCSK9 inhibitors is the lowering of Lp(a), something not achieved by most drug classes. Both alirocumab and evolocumab reached about 30% reductions in Lp(a) levels [35].

The Further Cardiovascular Outcomes Research with PCSK9 Inhibition in Subjects with Elevated Risk (FOURIER) trial found a 1.5% ARR in CV death, MI, stroke, hospitalization for UA or coronary revascularization with evolocumab (9.8% vs 11.3%, HR 0.85, 95% CI 0.79 to 0.92, NNT 67) [36].

 doi:10.20944/preprints202312.1195.v1

However, it is important to translate these findings to clinical practice in a critical manner - noteworthy, the reduction in risk itself is marginal (1.5%), considering it is a composite outcome.

The Evaluation of Cardiovascular Outcomes After an Acute Coronary Syndrome During Treatment With Alirocumab (ODYSSEY OUTCOMES) trial found a reduction in the composite outcome of coronary death, MI, stroke and hospitalization for UA with alirocumab (9.5% vs 11.1%, HR 0.85, 95% CI 0.78 to 0.93, NNT 63) [37]. The ODYSSEY OUTCOMES trial also reported a reduction in the secondary outcome of death from any cause (3.5% versus 4.1%) but without a significant reduction in CV death, which increases the odds that this finding was a result of chance. Like the FOURIER trial, the ODYSSEY OUTCOMES trial also presented a low effect estimate with PCSK9 inhibitors.

In terms of safety of PCSK9 inhibitors, both trials had similar findings, and some observations need to be made. The ODYSSEY OUTCOMES trial reported a high rate (~77%) of adverse events with no difference between treatment and placebo groups, with ~24% considered serious and only ~1.5% thought to be related to the study agent and leading to discontinuation of the treatment. Injection site reactions were more common in the evolocumab group (2.1% versus 1.6%), and were usually mild and led to discontinuation in only 0.1% of patients in each group.

Both trials point toward the same goal of furthering reducing LDL-C, for marginal reductions in events. Evidently, it is not expected that new treatments promote enormous absolute risk reductions on top of statins and ezetimibe, since the current scenario is of residual risk reduction. Nonetheless, it is important to maintain high standards when evaluating costs and possible measured and unmeasured adverse outcomes when prescribing a treatment that has such marginal clinical relevance. The expected effects of treatment should be discussed with patients for shared decision making, integrating the impact size of the treatment on outcomes in the discussion.

The short follow-up in these trials testing add-on therapies in patients already intensively treated could also contribute to those unimpressive results. Indeed, an open-label extension study of FOURIER called FOURIER-OLE shed some light on this matter. This extension enrolled a total of 6,635 patients from the FOURIER trial to receive evolocumab for an extension period, with 3,355 originally randomized to evolocumab and 3,280 originally randomized to placebo. The median follow-up in the FOURIER-OLE was 5 years, with the maximum exposure to evolocumab, considering FOURIER and FOURIER-OLE, of 8.4 years. At 12 weeks, the median LDL-C was 30 mg/dL. The incidence of serious adverse events, muscle events, new-onset diabetes, hemorrhagic stroke, and neurocognitive events did not exceed the incidence observed in the placebo group of the original study and did not increase over time. The risk of the composite cardiovascular outcome was 15% lower in patients originally randomized to evolocumab compared to patients originally randomized to placebo (OR 0.85, 95% CI 0.75 to 0.96, p=0.008), with 23% lower risk of cardiovascular death (HR 0.77, 95% CI 0.60 to 0.99, p=0.04), suggesting that long-term LDL-C reduction additionally decreases the risk of cardiovascular events [38].

Table 4. Main PCSK9 Inhibitors trials. Legend: ACS: acute coronary syndrome; ApoB: apolipoprotein B; CV: cardiovascular; MI: myocardial infarction; UA: unstable angina.

Study	Sample size	Characteristics of patients	Comparison groups	Follow-up	LDL effect	CV effects
FOURIER (2017) [36]	27,564	Documented atherosclerosis + LDL-C >70 mg/dL + on statin therapy	Evolocumab vs. placebo	2.2 years	59%	CV death, MI, stroke, UA hospitalization, or coronary revascularization (HR 0.85, 95% CI 0.79 to 0.92, NNT 67)
ODYSSEY	18,924	ACS <1-12	Alirocumab	2.8 years	54.7%	coronary death, MI,

 doi:10.20944/preprints202312.1195.v1

OUTCOMES (2018) [37]		months + LDL-C >70 mg/dL + on high-intensity statin or maximum tolerated dose	vs. placebo			stroke, or UA hospitalization (HR 0.85, 95% CI 0.78 to 0.93, NNT 63)

4.1. Tafolecimab

Tafolecimab is a fully human IgG2 PCSK9 monoclonal antibody. Phase 1 studies showed that Tafolecimab reduces LDL-C by more than 70% [39].

The phase 3 Clinical Research of Developing PCSK9 Inhibitor as Cholesterol-lowering Therapy in Chinese Patients with Dyslipidemia-1 (CREDIT-1) study showed that tafolecimab was safe and showed superior lipid-lowering efficacy versus placebo in non-familial hypercholesterolemia (HF) patients [40].

The Phase 3 Clinical Research of Developing PCSK9 Inhibitor as Cholesterol-lowering Therapy in Chinese Patients with Dyslipidemia-2 (CREDIT-2) study showed that tafolecimab yielded significant and persistent reductions in LDL-C levels and showed a favorable safety profile in chinese patients with heterozygous familial hypercholesterolemia (HeFH) [41].

4.2. Lerodalcibep

Lerodalcibep is a small binding protein that inhibits interaction between PCSK9 and LDL receptors - an alternative to monoclonal antibodies.

Phase 2 studies demonstrated that Lerodalcibep (LIB003) significantly reduced LDL-C [42,43]. The Long-term efficacy and safety of lerodalcibep in heterozygous familial hypercholesterolaemia (LIBerate-HeFH) trial randomized 319 patients with HeFH to monthly subcutaneous 1.2 mL injections of lerodalcibep 300 mg for 24 weeks and 159 patients to placebo [44]. Among patients who received monthly lerodalcibep injection within the steady window period (80% of the overall cohort) researchers reported a 24-week placebo-adjusted reduction in LDL-C of 63.6% ($p < 0.0001$) and a mean placebo-adjusted reduction during the average of weeks 22 and 24 of 70.2% ($p < 0.0001$) [45].

5. Ezetimibe

Ezetimibe binds to the Niemann–Pick C1-like 1 (NPC1L1) protein and selectively inhibits cholesterol absorption [46]. The decrease in cholesterol absorption leads to a reduction in the delivery of cholesterol to the liver, an increase in cholesterol clearance from the blood, and a reduction in hepatic cholesterol stores.

Dujovne et al. reported the efficacy of ezetimibe in 892 patients with primary hypercholesterolemia, observed in 12 weeks of treatment, by reducing LDL-C by an average of 17% when compared to placebo, as well as by reducing plasma levels of apolipoprotein B, triglycerides, and of the slight increase in HDL-C [47].

The Ezetimibe Add-On to Statin for Effectiveness (EASE) trial involved more than 3,000 patients randomized to combination therapy (statin and ezetimibe 10 mg) and statin alone for a period of six weeks. At the end of the study, the addition of ezetimibe to the statin represented an additional reduction of up to 23% in LDL-C, with 70% of these patients successfully achieving the objectives recommended by the National Cholesterol Education Program (NCEP) Adult Treatment Panel III (ATP III), compared to only 17% of those on standard therapy with statin alone [48].

In 2008, the publication of The Ezetimibe and Simvastatin in Hypercholesterolemia Enhances Atherosclerosis Regression (ENHANCE) trial raised doubts regarding the efficacy of ezetimibe. The trial randomized 720 patients with FH to receive simvastatin in combination with either ezetimibe or placebo to assess the effects of ezetimibe on carotid-artery intima-media thickness [49]. The results

 doi:10.20944/preprints202312.1195.v1

showed that ezetimibe in combination with simvastatin did not change the mean carotid-artery intima-media thickness significantly. However, a significant decrease in LDL-C, triglyceride and hsCRP levels were seen. Ezetimibe treatment was not associated with a significant increase in adverse events compared to placebo.

Subsequently, The IMProved Reduction of Outcomes: Vytorin Efficacy International Trial (IMPROVE-IT) showed that the association of ezetimibe 10 mg and simvastatin 40 mg reduced the incidence of CV events when compared to simvastatin 40 mg only in patients with high-risk acute coronary syndrome (ACS) and low LDL-C (<125 mg/dL) [50]. However, although the study had an NNT of 50, the RRR was only 6%. Furthermore, the confidence interval of this estimate, which is 1% to 11%, shows the inaccuracy of their estimates. Therefore, the IMPROVE-IT trial demonstrates that ezetimibe in patients using statins, with reasonably low cholesterol (mean LDL-C = 90 mg/dL), promotes a beneficial effect of minimal magnitude, yet proportional to its modest effect on LDL-C reduction.

The Heart Institute of Japan PRoper level of lipid lOwering with Pitavastatin and Ezetimibe in acute coronary syndrome (HIJ-PROPER) Trial, in turn, showed that the combination of pitavastatin + ezetimibe versus pitavastatin alone was not associated with a reduction in CV events (all-cause death, MI, stroke, UA or ischemia-guided coronary revascularization) in patients with non-ST elevation ACS (HR 0.89, 95% CI 0.76-1.04) [51].

The Ezetimibe Lipid-Lowering Trial on Prevention of Atherosclerotic Cardiovascular Disease in 75 or Older (EWTOPIA 75) trial showed that, compared with dietary counseling alone, the use of additional ezetimibe for primary prevention among elderly japanese patients with LDL-C ≥140 mg/dL and ≥1 high-risk feature reduced CV events, primarily cardiac events, with no difference in all-cause mortality. None of these patients were on statin therapy [52].

Finally, The Randomized Comparison of Efficacy and Safety of Lipid-lowering with Statin Monotherapy Versus Statin/ezetimibe Combination for High-risk Cardiovascular Disease (RACING) trial showed that among patients with ASCVD, moderate-intensity statin with ezetimibe combination therapy was non-inferior to high-intensity statin monotherapy for the 3-year composite outcomes (CV death, major CV events, or stroke) [53].

Table 5. Major clinical trials of ezetimibe. Legend: CAD: coronary artery disease; CABG: coronary artery bypass graft; CV: cardiovascular; DM: diabetes mellitus; ER-niacin: extended-release-niacin; HF: familial hypercholesterolemia; MI: myocardial infarction; NA: not evaluated; PAD: peripheral artery disease; PCI: percutaneous coronary intervention; SCD: sudden cardiac death; TC: total cholesterol; TG: triglycerides.

Study	Sample size	Characteristics of patients	Comparison groups	Follow-up	LDL effect	CV effects
EASE (2005) [48]	3,030	LDL-C above the NCEP ATP guidelines + TG <350 mg/dL + on statins therapy	Ezetimbe vs. placebo	6 weeks	25.8%	NE
ENHANCE (2008) [49]	720	HF	Simvastatin vs. simvastatin + ezetimibe	2 years	55.6%	carotid-artery intima-media thickness
IMPROVE-IT (2015) [50]	18,144	ACS <10 days + LDL-C 50-125 mg/dL (50-100	Simvastatin vs. simvastatin +	6 years	25.7% vs. 43.4%	CV events (HR 0,93, 95% CI 0,89

 doi:10.20944/preprints202312.1195.v1

		mg/dL if prior lipid-lowering therapy)	ezetimibe			to 0,99, NNT 50)
HIJ-PROPER (2017) [51]	1,734	ACS + LDL-C >100 mg/dL + TG <400 mg/dL	Pitavastatin vs. pitavastatin + ezetimibe	3.8 years	37.6% vs. 51.7%	No significant difference in the incidence of CV events
EWTOPIA 75 (2019) [52]	3,411	≥75 years + LDL-C >140 mg/dL + ≥1 high risk factor	Ezetimibe vs. control	4.1 years	25.9%	SCD, MI, PCI or CABG, or stroke (HR 0,66 95% CI 0,50 to 0,86, NNT 38.5)
RACING (2023) [53]	3,780	Documented atherosclerotic disease	Rosuvastatin 10 mg + ezetimibe 10 mg vs. rosuvastatin 20 mg	3 years	58% vs. 72%	No significant difference in the incidence of CV events

6. Bempedoic acid

Bempedoic acid is an ATP citrate lyase (ACL) inhibitor that targets cholesterol synthesis upstream of 3-hydroxy-3-methylglutaryl coenzyme A reductase, the enzyme inhibited by statins [54]. ACL decreases the conversion of mitochondrial-derived citrate to cytosolic ATP citrate lyase, thus creating less substrate for cholesterol and fatty acid synthesis with the end result of decreasing LDL-C synthesis and upregulating hepatic LDL receptor expression.

The Cholesterol Lowering via Bempedoic Acid [ECT1002], an ACL-Inhibiting Regimen (CLEAR OUTCOMES) trial showed that bempedoic acid was associated with a lower risk of major adverse CV events (CV death, MI, stroke, or coronary revascularization) among statin-intolerant patients (bempedoic acid 11.7% vs. placebo 13.3%; HR 0.87, 95% CI 0.79 to 0.96; NNT 62.5) [55].

In the setting of high CV risk patients and elevated LDL-C even with maximally tolerated statin or in combination with other therapies, a fixed-dose combination of bempedoic acid and ezetimibe significantly reduced LDL-C versus placebo [56].

Furthermore, in a recent meta-analysis including 10 RCTs and 18,200 patients, bempedoic acid was associated with a lower risk of CV events (CV death, MI, or stroke) [OR 0.84; 95% CI 0.76 to 0.96; $p < 0.001$; $I2 = 0\%$) [57].

Interestingly, in an analysis by Ridker et al, bempedoic acid presented a similar relative efficacy compared to placebo across baseline hsCRP and LDL-C categories [56]. Additionally, the authors showed that inflammation assessed by hsCRP predicted risk for future CV events and death more strongly than LDL-C [58]. However, this analysis must not be interpreted as diminishing the critical role of lipid lowering beyond statins for patients with persistent or refractory hypercholesterolemia,

 doi:10.20944/preprints202312.1195.v1

but suggests that targeting LDL-C alone may not completely mitigate atherosclerotic risk, and anti-inflammatory pathways may provide incremental CV benefits.

7. Lp(a)-Targeted Therapies

Lipoprotein(a) [Lp(a)] was first described by Berg in 1963 as an antigenically distinct form of beta-lipoprotein, composed of an LDL-like particle to which apoliprotein B-100 is covalently linked by a single disulfide bond to apolipoprotein(a), the pathognomonic component of Lp(a) [59]. There is controversy regarding the use of Lp(a) in the prevention of ASCVD: more than 90% of circulating Lp(a) levels are genetically determined, without being greatly influenced by diet or behavioral measures and, as a result, its levels do not fluctuate as much throughout their lives, which makes it difficult to come up with a possible therapy to reduce their levels [60]. Several guidelines currently have recommended measuring Lp(a) once only in patients with early ASCVD, FH, family history of early ASCVD and/or elevated Lp(a), recurrent CV event despite statin use and for reclassification of individuals with borderline risk (however, below 15% 10-year risk of a CV event) [61-63].

It was believed that the use of statins would not influence the fluctuation of Lp(a) levels, as LDL receptors (LDLR), which are more expressed due to the inhibition of HMG-CoA reductase and consequent reduction in tissue LDL-C, do not appear to be play an important role in the clearance of Lp(a). However, in a study involving 3,896 patients, an average increase in Lp(a) of 11% was observed when dosed before and after the initiation of statins of different strengths [64]. The mechanisms for this observed increase are not clear, but it is conjectured that it may be one of the reasons why some patients who are using maximum doses of statins are not responders because the majority of their cholesterol is in Lp(a) instead of the LDL-C particles [65]. These results did not occur in the Justification for the Use of Statins in Prevention: an Intervention Trial Evaluating Rosuvastatin (JUPITER) trial [9], whose Lp(a) value was also measured before and after the use of statins (rosuvastatin) and the average variation was equal to zero. Although highest baseline Lp(a) values – above 50 nmol/L – were associated with a 64% increase in the risk of MI, stroke, hospitalization for UA, arterial revascularization and CV death when compared to the lowest values (≤10 nmol/L), the group that used rosuvastatin had a lower incidence of CV events regardless of Lp(a) levels.

Specific medications to reduce Lp(a) are not yet available, although niacin, PCSK9 inhibitors, mipomersen and estrogen have effects on reducing its levels. In the Atherothrombosis Intervention in Metabolic Syndrome with Low HDL/High Triglycerides: Impact on Global Health Outcomes (AIM-HIGH) trial, the use of niacin reduced mean Lp(a) compared to baseline by 19% (however, in absolute levels, there was a modest reduction from 13.5 mg/dL to 11 mg/dL) [66]. There were no clinical benefits in the subgroup of patients with the greatest reduction (fourth quartile with Lp(a) >50 mg/dL) despite a 39% reduction when compared to baseline levels. Mipomersen and PCSK9 inhibitors reduce Lp(a) between 20% and 30%, however, since they do not exclusively reduce Lp(a), it is not possible to conclude about the clinical benefits of Lp(a) reduction. In a post-hoc analysis of the Heart and Estrogen/progestin Replacement Study (HERS) trial, patients who received estrogen and progestin showed an average reduction of 5.8 mg/dL in Lp(a) versus 0.34 mg/dL in the placebo group after one year of follow-up [67]. Patients who had a greater reduction in Lp(a) (> -8.8 mg/dL) had 38% fewer events (MI and CV death) than patients with a smaller reduction in Lp(a). However, due to the increased incidence of thromboembolic events in this study, hormonal therapy in menopausal women is not a common alternative.

Mendelian randomization studies estimate that each 60 to 100 mg/dL reduction in Lp(a) could translate into a reduction in the rate of cardiovascular events of around 20%, corresponding to a reduction in LDL-C of around 40 mg/dL with the use of statins. Possibly the benefit will be greater, the higher the level of Lp(a) and the greater the reduction of its levels. Therefore, therapies specifically focused on intensive Lp(a) reduction have been developed and have been currently tested in high-risk individuals with high Lp(a) concentrations. Pelacarsen and olpasiran are both therapies directed against apo(a) mRNA and able to reduce Lp(a) levels in approximately 80% and 100%, respectively. Phase 3 outcomes studies are ongoing [68,69].

 doi:10.20944/preprints202312.1195.v1

8. Bile Acid–Binding Resins

Currently, three bile-acid sequestrants (BAS) are available: (1) cholestyramine (developed in the 1950s), (2) colestipol (developed in the 1970s), and (3) colesevelam (approved in 2000).

The bile acid-binding resins are highly positively charged molecules that bind to the negatively charged bile acids in the intestine, preventing their reabsorption in the terminal ileum and consequently increasing fecal excretion of bile acids. Reduction of the body's bile acid pool stimulates the conversion of cholesterol to bile acids in the liver. Increased bile acid synthesis reduces hepatic cholesterol levels, leading to increased expression of enzymes that act in the uptake of cholesterol from the bloodstream to the liver through greater expression of LDLR [70].

The first large double-blind study with adequate sample size and quality to evaluate the effect of LDL-C-lowering drugs on the occurrence of CV events was the Lipid Research Clinics Coronary Primary Prevention Trial (LRC-CPPT) [71]. The trial included 3,806 asymptomatic men between 35 and 59 years of age with a diagnosis of primary hypercholesterolemia (defined as LDL-C ≥190 mg/dL) and who had never had a history of heart attack, angina or heart failure, being followed for an average time of 7.4 years. The mean baseline LDL-C was 216 mg/dL. The trial showed that cholestyramine reduced LDL-C in 20.3% (12.6% greater reduction than that obtained in the placebo group). There was 19% less occurrence of primary events (CV death or MI) in the cholestyramine group. The main side effects were gastrointestinal symptoms, more present in the cholestyramine group (68% versus 43% in the placebo group). An important limitation of most studies that evaluate the impact of plasma lipid measurements on CV risk is the long period without measurement between one dose and another, leading to the uncertain thought that the behavior of LDL-C over the unmeasured period occurs linearly.

Hypothetically, if between the measurement of LDL-C in the first year and the measurement of LDL-C in the seventh year, a patient in the cholestyramine group did not adhere to treatment for five years, but returned to using it one year earlier, it is not possible have a faithful analysis of the behavior of LDL-C in the five years of poor adherence, but only the record of the shortest period of adherence. Obviously, this is an extreme example, but it serves to illustrate the caution we must take with studies that evaluate associations with widely spaced LDL-C measurements.

Colestipol can reduce LDL-C levels by 15 to 30% [72], but there are no published clinical trials that have evaluated the benefit of colestipol in reducing cardiovascular events. Currently, its use is restricted to some cases of patients with primary hypercholesterolemia.

Colesevelam was developed with the expectation that it would be better tolerated from a gastrointestinal point of view and have fewer drug interactions. In fact, due to its greater affinity for bile acids (it has a mechanism of action very similar to other BAS, but has a different biochemical structure), colesevelam is better tolerated and has a better interaction profile when compared to other medications.

Colesevelam monotherapy was studied in two clinical trials of patients with moderate primary hypercholesterolemia. Maximum LDL-C reduction of 19% compared to baseline was achieved after 2 weeks of treatment [73,74].

Meta-analysis using Mendelian randomization evaluated the effect of cholestyramine and colesevelam by quantifying the effect on CV risk reduction of rs4299376 (ABCG5/ABCG8), which affects the intestinal absorption pathway of BAS target cholesterol [75]. Nineteen RCTs with a total of 7,021 study participants were included. Cholestyramine was associated with a reduction in LDL-C of 23.5 mg/dL and a trend towards reduced risk of CAD (OR 0.81, 95% CI 0.70 to 1.02; p=0.07) while colesevelam was associated with a reduction in LDL-C of 22.7 mg/dL (95% CI -28.3 to -17.2). There is no description of baseline LDL-C in these studies. Based on the findings that rs4299376 was associated with a 2.75 mg/dL decrease in LDL-C and a 5% decrease in risk of CAD outcomes, cholestyramine was associated with an OR for CAD of 0.63 (95% CI 0.52 to 0.77; $p=6.3x10^{-6}$) and colesevelam with an OR of 0.64 (95% CI 0.52 to 0.79, $p=4.3x10^{-5}$).

Due to the modest reduction in LDL-C and undesirable gastrointestinal effects, these medications remain in the background in the treatment of dyslipidemia, in addition to being used for

 doi:10.20944/preprints202312.1195.v1

the treatment of type 2 diabetes mellitus, as it has been shown to lower the glycosylated hemoglobin (A1C) by 0.5% on average in patients [75].

9. Nicotinic Acid

Niacin or nicotinic acid is a soluble vitamin with lipid-lowering properties. Niacin appears to reduce the mobilization of free fatty acids from adipocytes, acting on specific receptors, reducing the formation of lipoproteins rich in triglycerides in the liver. There are two forms of niacin, one with rapid absorption (crystalline), more commonly associated with flushing, and another with extended release, with better tolerability.

Niacin is capable of increasing HDL-C levels and reducing apoB-containing lipoproteins concentrations [VLDL, IDL, LDL and lipoprotein(a)]. The medication was evaluated in several clinical trials as shown in Table 6.

Table 6. Major clinical trials of niacin. Legend: ACS: acute coronary syndrome; CAD: coronary artery disease; CABG: coronary artery bypass graft; CV: cardiovascular; DM: diabetes mellitus; ER-niacin: extended-release-niacin; MI: myocardial infarction; PAD: peripheral artery disease; TC: total cholesterol; TG: triglycerides; TIA: transient ischemic attack.

Study	Sample size	Characteristics of patients	Comparison groups	Follow-up	Lipids effect	CV effects
CDP (1975) [76]	8,341	Men with previous MI	Niacin or clofibrate vs. placebo	6 years	TC 9.9%	nonfatal MI 27% cerebrovascular events 24%
Stockholm trial (1977) [77]	558	ACS + aged <70 years	Clofibrate + niacin vs. placebo	5 years	TC 26%	CV events (HR 0.59, 95% CI 0.41 to 0.84)
CLAS (1987) [78]	162	Men aged 40-59 years + previous CABG	Niacin + colestipol vs. placebo	2 years	LDL-C 43% HDL-C 37%	atherosclerotic regression (16.2% vs. 2.4%, p=0.002)
HATS (2001) [79]	160	CAD + LDL-C <140 mg/dL + TG <400 mg/dL	Simvastatin + niacin vs. antioxidant vs. simvastatin + niacin + antioxidant vs. placebo	3 years	LDL-C 42% HDL-C 26%	death, MI, stroke, or revascularization
ARBITER-2 (2004) [80]	149	CAD + on statin therapy	ER-niacin vs. placebo	1 year	HDL-C 21%	progression of carotid intima-media thickness
AFREGS (2005) [82]	143	CAD + LDL-C <160 mg/dL + HDL-C <40 mg/dL	Niacin + gemfibrozil + cholestyramine vs. placebo	2.5 years	LDL-C 24% HDL-C 38%	angiographic progression (34.72% vs. 36.02%, p=0.0002)
ARBITER-6 (2009) [81]	208	CAD or CAD risk equivalent on statin	ER-niacin vs. ezetimibe + pre-existing	1.2 years	LDL-C (20% vs.12%)	progression of carotid intima-media thickness

 doi:10.20944/preprints202312.1195.v1

		therapy	statin therapy		HDL-C (+18% vs. -7%)	
AIM-HIGH (2011) [66]	3,414	Aged >45 years + CV disease	ER-niacin vs. placebo	3 years	LDL-C 14% HDL-C 25%	No significant difference in the incidence of CV events
HPS2-THRIVE (2014) [83]	25,673	Aged >50 years + CV disease	ER-niacin/laropiprant vs. placebo	3.9 years	LDL-C 10% HDL-C 6%	No significant difference in the incidence of CV events

Meta-analysis of 17 studies with niacin that provided data on cardiovascular disease (CVD) outcomes showed that niacin might have some utility in lipid control for secondary prevention as monotherapy, although the evidence is from older studies in a population potentially not representative of current patients [84].

However, Ronsein et al. recently showed that the addition of niacin to statin therapy resulted in elevated levels of multiple HDL proteins linked to increased atherosclerotic risk, which might have compromised the cardioprotective effects associated with higher HDL-C levels and lower levels of LDL-C and triglycerides [85].

10. Fibric Acid Derivatives

Fibrates have multiple pharmacological actions, mainly as a synthetic ligand for the peroxisome proliferator-activated receptors (PPARs), especially PPARα. Clinically, fibrates reduce plasma triglycerides (TG) or TG-rich lipoproteins (TRLs) and increase HDL-C levels.

Fibrates were evaluated in several clinical trials as shown in Table 7.

Table 7. Major clinical trials and epidemiological studies of fibrates. Legend: CAD: coronary artery disease; CV: cardiovascular; MI: myocardial infarction; NE: not evaluated; PAD: peripheral artery disease; TG: triglycerides; T2DM: type 2 diabetes mellitus.

Study	Sample size	Characteristics of patients	Comparison groups	Follow-up	Lipids effect	CV effects
HHS (1987) [86]	4,081	Men with non-HDL-C >200 mg/dL	Gemfibrozil 1200 mg/d vs. placebo	5 years	HDL-C 11% LDL-C 11% TG 35%	incidence of CAD
VA-HIT (1999) [87]	2,531	Documented CAD + LDL <140 mg/dL	Gemfibrozil 1200 mg/d vs. placebo	5.1 years	TG 31% HDL-C 6%	MI or coronary death
BIP (2000) [88]	3,090	Previous MI or stable angina + LDL-C <180 mg/dL	Bezafibrate 400 mg/d vs. placebo	6.2 years	TG 21% HDL-C 18%	No significant difference in the incidence of CV events
LEADER (2002) [89]	1,568	Men with lower PAD	Bezafibrate 400 mg/d vs. placebo	4.6 years	NE	No significant difference in the incidence of CAD and

 doi:10.20944/preprints202312.1195.v1

						stroke
FIELD (2005) [90]	9,795	T2DM	Fenofibrate 200 mg/d vs. placebo	5 years	TG 29% HDL-C 5% LDL-C 12%	No significant difference in the incidence of CAD death or non-fatal MI
ACCORD-Lipid (2010) [91]	5,518	T2DM + CV risk factor or documented CV disease	Fenofibrate 160 mg/d vs. placebo	4.7 years	HDL-C 9% TG 23%	No significant difference in the incidence of CV events
ECLIPSE-REAL (2019) [92]	10,705	Metabolic syndrome	Statin vs. statin + fenofibrate	6 years	TG with statin + fenofibrate	CAD, stroke or CV death with statin + fenofibrate (HR 0.74, 95% CI 0.58 to 0.93)
PROMINENT (2022) [95]	10,497	T2DM + TG 200-499 mg/dL + HDL <40 mg/dL	Pemafibrate 0.4 mg/d vs. placebo	3.4 years	TG 31.1%	No significant difference in the incidence of CV events

Meta-analysis of six trials including 16,135 individuals evaluated the clinical benefits of fibrates for primary prevention of cardiovascular disease and showed that fibrates lower the risk for CV and coronary events in primary prevention, although the absolute treatment effects in the primary prevention setting were modest (absolute risk reductions <1%) [93].

In the context of secondary prevention, meta-analysis of 13 RCTs including 16,112 patients showed evidence of a protective effect of the fibrates compared with placebo in terms of the primary composite outcome stroke, MI, and CV death (HR 0.88, 95% CI 0.83 to 0.94) [94]. Nevertheless, the latest studies, particularly FIELD and ACCORD, did not provide further scientific support for the use of fibrates in the context of cardiovascular prevention. On the other hand, both studies did not specifically have baseline triglyceride levels as an inclusion criterion, and a subsequent subgroup analysis showed a benefit in reducing cardiovascular risk in those with hypertriglyceridemia compared to individuals with lower triglyceride values. The Pemafibrate to Reduce Cardiovascular OutcoMes by Reducing Triglycerides IN patiENts with diabetes (PROMINENT) was conducted to evaluate the role of fibrate additive therapy to reduce the residual risk of atherosclerotic cardiovascular disease in patients with high triglycerides [95]. Pemafibrate is a synthetic ligand to PPAR-alpha with a potency greater than 2,500 times that of fenofibrate and with greater selectivity for PPAR-alpha in relation to PPAR-gamma. In 2022, the study was interrupted due to futility. These results suggest that the fibrates do not present a sufficient level of evidence for their use in cardiovascular prevention in patients with mild to moderate hypertriglyceridemia (triglycerides between 150 and 500 mg/dL).

11. Omega-3 fatty acids

Omega-3 fatty acids (OM3FAs) are unsaturated fatty acids with at least one double bond located between the third and fourth omega end carbon. Currently, the three most clinically relevant omega-3 polyunsaturated fatty acids (PUFAs) are α-linolenic acid (ALA), eicosapentaenoic acid (EPA), and docosahexaenoic acid (DHA). OM3FAs suppress lipogenic gene expression by decreasing the

 doi:10.20944/preprints202312.1195.v1

expression of sterol regulatory element-binding protein 1c, inhibiting phosphatidic acid phosphatase, and acyl-CoA:1,2-diacylglycerol acyltransferase (NGAT). Sterol regulatory element-binding proteins (SREBP's) are membrane-bound enzymes that, when cleaved, travel to the nucleus to transcribe enzymes involved in cholesterol, LDL, and fatty acid synthesis. When a diet is high in omega-3 fatty acids, the SREBPs (particularly 1c) are not activated because of negative feedback inhibition and lowers SREBP synthesis and the cholesterol synthesizing enzymes that it regulates; FPP synthase (farnesyl diphosphate synthase) and HMG-CoA reductase (3-hydroxy-3-methylglutaryl-CoA reductase) [96].

Two initial studies, the Japan EPA Lipid Intervention Study (JELIS) [97] and OMEGA [98], showed controversial results on the benefits of omega-3 in patients without and with established cardiovascular disease, respectively.

The Vitamin D and Omega-3 Trial (VITAL) trial showed that n-3 fatty acids did not result in a lower incidence of major CV events than placebo after a median follow-up of 5.3 years (HR 0.92; 95% CI 0.80 to 1.06; p=0.24) [99].

The multinational Reduction of Cardiovascular Events with Icosapent Ethyl – Intervention Trial of icosapent ethyl (REDUCE-IT) trial showed a 25% RRR in CV events (17.2% vs. 22.0%, HR 0.75, 95% CI 0.68 to 0.83, NNT 21) [100].

Recently, a post-hoc exploratory analysis of patients from the REDUCE-IT study assessed whether the magnitude of the reduction in CV outcomes observed was not caused by possible harm from mineral oil, used as placebo in the comparator group [101]. In this group, levels of biomarkers associated with atherosclerosis increased over time. In other words, in the placebo group, at 12 months of follow-up, significant increases in homocysteine, Lp(a), interleukin-6, lipoprotein-associated phospholipase A2 (Lp-PLA2), C-reactive protein (CRP), and beta-1 were observed. These changes remained similar at the end of 24 months. However, it should be noted that the REDUCE-IT study was designed for hard outcomes, and that post-hoc analysis with biomarkers (exploratory outcomes) may raise hypotheses, but do not prove that mineral oil made a difference in the study. Finally, these data may call the results of the REDUCE-IT study into question, and caution is needed until definitive evidence is obtained.

The Residual Risk with Epanova in High Cardiovascular Risk Patients with Hypertriglyceridemia (STRENGTH) trial included 13,078 individuals with established ASCVD and showed that there was no significant difference in the primary endpoint (CV death, MI, stroke, coronary revascularization or UA requiring hospitalization) between the treatment (4g daily of omega-3 carboxylic acid (EPA plus DHA)) and placebo arms (12.0% vs. 12.2%; HR 0.99, 95% CI 0.90 to 1.09, p=0.84) [102].

The Omega-3 Fatty acids in Elderly with Myocardial Infarction (OMEMI) trial assessed the efficacy of 1.8 g daily of n-3 PUFA (930 mg EPA and 660 mg DHA) compared with placebo among older adults (mean age 75 years) after acute MI and also showed that there was no significant difference between the n-3 FA treatment group compared with the placebo control (21.4% vs. 20.0%; HR 1.08, 95% CI 0.82 to 1.41, p=0.06) [103].

The reasons for the contrasting results in CVD outcomes of REDUCE-IT compared with STRENGTH and OMEMI are likely multifactorial, which include patient selection, difference in endpoints, dosing, and choice of placebo.

Meta-analysis including 149,051 participants showed that omega-3 fatty acids were associated with reduced cardiovascular events (CV death, MI, CAD events, and coronary revascularization) [104].

 doi:10.20944/preprints202312.1195.v1

Table 8. Characteristics of the main published trials involving omega-3 fatty acids. Legend: ACS: acute coronary syndrome; CV: cardiovascular; MI: myocardial infarction; NA: not evaluated; SCD: sudden cardiac death; TC: total cholesterol.

Study	Sample size	Characteristics of patients	Comparison groups	Follow-up	LDL effect	CV effects
JELIS (2007) [97]	18,645	TC >250 mg/dL	Omega-3 1.8 g/d + statin vs. only statin	4.6 years	LDL-C TC (no differences between groups)	CV events (HR 0.81, 95% CI 0.69 to 0.95, NNT 143)
OMEGA (2010) [98]	3,818	ACS	Omega-3 1 g/d vs. placebo	1 year	NE	No difference in SCD
VITAL (2019) [99]	25,871	No known CV disease	Omega-3 1 g/d vs. placebo	5.3 years	NE	No significant difference in the incidence of CV events
REDUCE-IT (2019) [100]	8,179	Age >45 years + CV disease or age >50 years + diabetes and ≥1 risk factor + TG 150-499 mg/dL + LDL-C 41-100 mg/dL	Omega-3 4 g/d vs. placebo	4.9 years	LDL-C 3.1% TG 18.3%	CV death, MI, stroke, coronary revascularization, or UA (HR 0.75, 95% CI 0.68 to 0.83, NNT 21)
STRENGTH (2020) [102]	13,078	High CV risk	Omega-3 4 g/d vs. placebo	42 months	NE	No significant difference in the incidence of CV events
OMEMI (2021) [103]	1,027	Aged 70-82 years + recent MI (2-8 weeks)	Omega-3 1.8 g/d vs. placebo	2 years	NE	No significant difference in the incidence of CV events

12. Cholesteryl ester transfer protein (CETP) inhibitors

Cholesteryl ester transfer protein (CETP) is a plasma glycoprotein that mediates the transfer of cholesteryl esters from HDL to the apoB-containing lipoproteins, with a balanced transfer of triglycerides [105]. Inhibition of CETP results in an accumulation of cholesterol esters in HDL, thus resulting in increased HDL-C.

The first large study that evaluated the CV effects of CETP inhibitors was the Investigation of Lipid Level Management to Understand its Impact in Atherosclerotic Events (ILLUMINATE) [106]. This study showed an increase in cardiovascular events with the medication, probably due to increased blood pressure. Subsequently, other medications in the same class had negative results in large phase 3 trials. The dal-OUTCOMES study did not show a reduction in CV events, despite

 doi:10.20944/preprints202312.1195.v1

improving the lipid profile, with dalcetrapib [107]. Similar findings were found with evacetrapib in the Assessment of Clinical Effects of Cholesteryl Ester Transfer Protein Inhibition with Evacetrapib in Patients at a High Risk for Vascular Outcomes (ACCELERATE) trial [108].

On the other hand, the phase 3 Randomized Evaluation of the Effects of Anacetrapib through Lipid Modification (REVEAL) trial showed a reduction in the primary composite outcome with anacetrapib [109], although with a RRR of only 9% and a modest NNT of 100, demonstrating the beneficial effect of minimal magnitude. Despite the trial's positive results, a few weeks after its publication, Merck announced that it would not attempt to approve the medication at the food and drug administration (FDA).

Obicetrapib, although belonging to the class of CETP inhibitors, has attracted interest due to the potent LDL-C reduction and favorable safety profile. In the Randomized Study of Obicetrapib as an Adjunct to Statin Therapy (ROSE) study, treatment with obicetrapib for 8 weeks when compared with placebo in 120 individuals with dyslipidemia on statin treatment, demonstrated a reduction in LDL-C, apoB and non-HDL-C by 51%, 30% and 44% compared to placebo, respectively [110]. Obicetrapib has also been evaluated in combination with ezetimibe in individuals treated with statins in a phase 2 study, showing a 63% reduction in LDL-C. The Cardiovascular Outcomes Study to Evaluate the Effect of Obicetrapib in Patients with Cardiovascular Disease (PREVAIL), which aims to evaluate obicetrapib in about 9,000 patients with atherosclerotic cardiovascular disease, is ongoing (NCT05202509) [111].

Table 9. Characteristics of the main published trials involving CETP inhibitors. Legend: ACS: acute coronary syndrome; CAD: coronary artery disease; CV: cardiovascular; HeFH: heterozygous familial hypercholesterolemia; MI: myocardial infarction; NE: not evaluated.

Study	Sample size	Characteristics of patients	Comparison groups	Follow-up	LDL effect	CV effects
ILLUMINATE (2007) [106]	15,067	CV disease	Torcetrapib 60 mg/d vs. placebo	1 year	HDL-C 72.1% LDL-C 24.9%	CV events (HR 1.25, 95% CI 1.09 to 1.44)
dal-OUTCOMES (2012) [107]	15,871	ACS	Dalcetrapib 600 mg/d vs. placebo	31 months	HDL-C 30%	No significant difference in the incidence of CV events
ACCELERATE (2017) [108]	12,092	CV disease	Evacetrapib 130 mg/d vs. placebo	26 months	HDL-C 133.2% LDL-C 31.1%	No significant difference in the incidence of CV events
REVEAL (2017) [109]	30,449	Atherosclerotic disease + on atorvastatin therapy + mean LDL-C 61 mg/dL	Anacetrapib 100 mg/d vs. placebo	4.1 years	non-HDL-C 18%	coronary death, MI, or coronary revascularization (HR 0.91, 95% CI 0.85 to 0.97, NNT 100)
ROSE (2022) [110]	120	Dyslipidemia on statin treatment	Obicetrapib 10 mg vs. placebo	8 weeks	LDL-C 51%	NE

 doi:10.20944/preprints202312.1195.v1

13. Gene Therapy

Gene therapy can be carried out by replacing a missing gene, overexpressing a certain gene, interfering with the expression of a gene or by promoting the repair of a gene carrying a mutation. Several strategies can be used to repair a gene, but crucial to the success of this form of therapy is the form of transport to the target cell and its nucleus. Viral vectors have been widely used as carriers, but this method has the limitations of preventing large-scale production and being contaminated by other viruses, among others.

The emergence of new technologies, such as small interfering ribonucleic acid (siRNA), antisense oligonucleotides (ASO), clustered regularly interspaced short tandem repeats (CRISPR), and new transport methods, such as nanomaterials and lipid carriers, has provided a major advance in applicability clinic of this form of treatment.

The primary defect in 85% of FH cases is mutation or deletion of the LDLR-encoding gene responsible for removing LDL-C via endocytosis and intracellular degradation. However, different targets have been used to reduce LDL-C levels, such as LDL receptors, PCSK9, ANGPTL3, APOC3, and Lp(a).

The first gene therapy applied to hypercholesterolemia showed a 15% reduction in serum LDL-C concentration in only three patients [112]. However, the low efficiency of genetic reconstitution (5-10% of cultured cells incorporated the LDLR cDNA) using the retroviral vector was considered the major hurdle that led to this relatively small effect.

Since then, different strategies have been applied to increase the therapeutic efficacy or reduce the immunogenicity of gene therapy, including the following mechanisms: (1) Virus Vector-Mediated Therapy; (2) Exosome-mediated therapy; (3) siRNA; (4) ASO; (5) Minicircle DNA Vectors; (6) MicroRNAs; (7) Long Non-Coding RNAs (lncRNAs); (8) CRISPR/Cas9 System [113].

13.1. siRNA

Small interfering RNAs (siRNAs) are short regulatory RNA molecules that suppress genes that are overexpressed in a given disease through post-transcriptional silencing.

Effective pharmacological use of siRNA requires 'carriers' that can deliver the siRNA to its intended site of action, such as viral and non-viral vectors (polymeric or lipid nanoparticles, polyplexes, lipoplexes, liposomes, or multifunctional nanocarriers).

In the cytoplasm, siRNA is processed from double-stranded RNA, which comes from endogenous transcription of DNA or from exogenous sources such as a virus. This double-stranded RNA is then cleaved by the ATP-dependent RNA endonuclease, Dicer, into fragments 21-23 nucleotides long with two nucleotide overhangs at both ends. This siRNA is then loaded into another protein, Argonaute. Argonaute has four different domains – N-terminal, PAZ, Mid, and PIWI. Its PIWI domain has RNase activity that allows Argonaute to cleave target mRNA. The Argonaute-siRNA complex then binds with a helicase and other proteins to form the RNA-induced silencing complex (RISC). In RISC, the sense strand is separated from the antisense, or guide strand, which is thought to be catalyzed by the helicase. The sense strand is degraded in the cytoplasm, and the guide strand directs the RISC to a complementary target mRNA.

The fate of the target mRNA is determined by whether the guide mRNA exhibits optimal or sub-optimal base pairing with the target mRNA. If the guide strand shows ideal base pairing with the target mRNA, the target mRNA is cleaved by Argonaute. Then the RISC complex is reused again to target another mRNA. In contrast, if the guide strand exhibits suboptimal base pairing with the target mRNA strand, Argonaute will not cleave the mRNA. Instead, it will lead to translation arrest, as the RISC complex will obstruct ribosome binding and translocation. These mRNAs are then guided to processing bodies (P-bodies) where they are gradually degraded. In the nucleus, siRNA can silence transposable DNA elements and thus prevent their unwanted and dangerous random insertions into the genome.

There are, however, some challenges to the administration of siRNAs [114], as shown in Table 9.

 doi:10.20944/preprints202312.1195.v1

Table 9. Challenges for administering siRNAs.

Physicochemical characteristics of the siRNA molecule	Hydrophilic macromolecule
	High molecular weight (~13 kDa)
	Negative charge
	Low stability in biological fluids
Barriers inherent to the route of administration	Tissue penetration
	Interstitial nucleases
	Internalization by target cells
	Activation of the immune response

13.1.1. Lepodisiran

Lepodisiran is a short interfering RNA directed at hepatic production of apolipoprotein(a). In a phase 1 study of 48 participants with elevated Lp(a) levels and without cardiovascular disease, lepodisiran reduced Lp(a) levels by 94% at 48 weeks [115].

13.1.2. Inclisiran

Since the discovery of the interaction of PCSK9 with LDL-C metabolism, several studies have sought alternative therapies for LDL-C reduction, targeting PCSK9. The initial results of monoclonal antibodies (alirocumab and evolocumab) were highlighted in the scientific world due to the significant decrease in LDL-C levels and reduction in cardiovascular death, MI, stroke and hospitalization for angina in an average follow-up of 2.5 years [116,117]. However, there are still doubts about potential adverse immunological effects of these medications in the long-term, as the studies are recent. Furthermore, the current dosage (biweekly application) of these medications can make it difficult for patients to adhere to them. Due to these concerns, an alternative to monoclonal antibody treatment was sought, but still involving PCSK9 as a target.

Inclisiran is a small interfering RNA (siRNA), small synthetic molecule that binds to PCSK9 mRNA, promoting its degradation and consequent inhibition of PCSK9 synthesis [118]. These molecules called siRNA are about 25 nucleotides of RNA, which have recently become known as important regulators of the expression and function of the human genome. Long double-stranded RNA, like that present in some types of viruses, induces an immunological response via interferon, which, in turn, causes a generalized interruption of protein synthesis. Due to this characteristic, long double-stranded RNA cannot be used to silence specific genes. On the other hand, siRNAs can escape the response radar via interferon and, therefore, promote the effective silencing of specific genes through complementary nucleotide sequences that affect post-transcriptional mRNA degradation, thus preventing the translation process [119]. Inclisiran is composed of these long-acting synthetic siRNA molecules, which bind intracellularly to the RISC, cleaving mRNA that would specifically participate in the translation of PCSK9 and consequently its synthesis in the liver, allowing the reduction of circulating levels of PCSK9 by up to 70% in phase 1 study [120].

The characteristics of the main trials published on inclisiran are available in Table X. The first phase 2 study was published in 2017 [121]. The ORION-1 trial included patients with ASCVD and elevated LDL-C (>70 mg/dL) despite maximum tolerated statin therapy and patients without ASCVD but with high cardiovascular risk conditions such as diabetes and FH in whom LDL-C was >100 mg/dL despite maximally tolerated statin therapy. Patients using PCSK9 inhibitors were excluded. Patients were randomized into eight treatment groups: a single dose regimen of 200, 300 or 500 mg inclisiran, or placebo; or a two-dose regimen of 100, 200 or 300 mg inclisiran, or placebo. The primary endpoint of LDL-C reduction at 180 days was reduced by 27.9-41.9% with one subcutaneous injection and by 35.5-52.6% with two injections ($p < 0.001$). At 240 days, the reductions in PCSK9 and LDL-C remained significantly lower than baseline with all the studied doses of inclisiran. Two injections of

 doi:10.20944/preprints202312.1195.v1

the 300 mg dose of inclisiran produced the greatest reduction in LDL-C, with 48% of patients receiving this dose achieving an LDL-C level <50 mg/dL.

The ORION-3 trial, an open-label extension of the phase II ORION-1 trial, assessed whether LDL-C level reduction was sustained over a four-year follow-up [122]. Patients treated with inclisiran achieved an average 47.5% reduction in LDL-C from baseline (day 1 of ORION-1) to day 210 (95% Cl -50.7 to -44.3) and a time-averaged reduction in LDL-C of 44.2% over the 4 years through twice-yearly dosing. Another result of this study is the evaluation of the efficacy and safety of switching from a monoclonal antibody (evolocumab) to inclisiran 24 days after the last dose of evolocumab (staggered change) or on the same day of the last application (concurrent change). The researchers allocated patients into two groups: "inclisiran-only", which was the extension of the duration of use of those patients who were originally already receiving inclisiran (in the ORION-1 study) and a second group called "switching arm", formed by patients who had received placebo in the ORION-1 trial and started receiving evolocumab for one year and a pre-established switch to inclisiran for the next 3 years. There was a greater mean percentage reduction in LDL-C in patients using evolocumab - 61.0% (95% CI 64.5 to 57.4) than after switching to inclisiran - time-averaged 3-year LDL-C reduction of 45.3% (95% CI 49.7 to 40.9). However, it was not a study designed to compare the effectiveness of inclisiran versus evolocumab, but rather, the ease, safety and effectiveness of switching from one medication to the other at different times and in a single arm – formally, to compare the effectiveness of the two medications, ideally randomization should take place into three arms: inclisiran, evolocumab and a placebo group. Interestingly, the placebo group showed a very small average variation in LDL-C levels (less than 10% compared to baseline levels), even with more than 70% of the population using statins. Absolute LDL-C levels in both groups ("inclisiran-only" and "switching arm") were similar at the time of switching from one medication to the other. There was no placebo group in this study: 92 patients out of the 127 originally belonging to the placebo group of the ORION-1 trial started receiving evolocumab before the scheduled switch to inclisiran (on day 336 or 360, as mentioned above) while 290 of 370 patients were allocated to drug continued into the inclisiran-only arm.

On the other hand, the phase 3 ORION-5 trial showed that inclisiran treatment did not reduce LDL-C levels in patients with homozygous familial hypercholesterolemia (HoFH) despite substantial lowering of PCSK9 levels [123]

Subsequently, the ORION-9 trial evaluated the safety and efficacy of inclisiran in lowering LDL-C among patients with HeFH [124]. There was a 50% observed LDL-C lowering at Day 510 with a 45% time-adjusted LDL-C lowering between days 90 and 540 indicating that inclisiran is superior to placebo in reducing LDL-C among patients with HeFH who are already on statins and ezetimibe. Almost all patients in both groups used statins (90%), however, 76.4% of the group that received inclisiran used high-potency statins versus 71.2% of the placebo group and 55.8% of the inclisiran group used ezetimibe versus 50.0% of the placebo group. These differences in lipid-lowering treatment between the groups do not seem to have considerably affected the result, but the increase of 8.2% in LDL-C levels on day 510 when compared to baseline levels in the placebo group is noteworthy, even when receiving a high-potency statin in a large proportion and half of the population using ezetimibe.

A pooled analysis of two phase 3 trials (ORION-10 and -11 trials) evaluated the individual responses of patients on LDL-C reduction with inclisiran [125]. This analysis showed a highly consistent effect, with a safety and tolerability profile similar to placebo, on a twice-yearly dosing schedule after an initial dose and one 3 months later, across individual patients with ASCVD or risk equivalents over 17 months of treatment. At day 510, inclisiran reduced LDL-C levels by 52.3% (95% CI 48.8 to 55.7) in the ORION-10 trial and by 49.9% (95% CI 46.6 to 53.1) in the ORION-11 trial.

In a post hoc analysis of the ORION-9, -10 and -11 trials showed that inclisiran was well-tolerated and effective in LDL-C reduction in both patients with and without polyvascular disease (PVD) [126]. The mean placebo-corrected LDL-C % change from baseline to day 510 was -48.9% (95% CI -55.6 to -42.2) in patients with PVD and -51.5% (95% CI -53.9 to -49.1) in patients without PVD.

 doi:10.20944/preprints202312.1195.v1

Recently, a patient-level, pooled analysis of ORION-9, -10 and -11 trials showed a placebo-corrected percentage reduction in LDL-C with inclisiran (50.6% at day 90) [127]. Furthermore, inclisiran reduced composite MACE (non-adjudicated CV death, cardiac arrest, MI, and stroke) [OR 0.74, 95% CI 0.58 to 0.94], but not fatal and non-fatal MIs [OR 0.80, 95% CI 0.50 to 1.27] or fatal and non-fatal stroke [OR 0.86, 95% CI 0.41 to 1.81] over 18 months, suggesting a potential benefit of inclisiran for MACE.

The ongoing ORION-4 trial recruited more than 16,000 patients from 2018 to evaluate the reduction of clinical outcomes in patients at high risk for atherosclerotic disease [128].

Table 10. Characteristics of the main published trials involving inclisiran. Legend: CV: cardiovascular; NE: not evaluated.

Study	Sample size	Characteristics of patients	Comparison groups	Follow-up	Lipids effect	CV effects
ORION-1 (2017) [121]	501	Patients at high risk for CV disease and elevated LDL-C	Different doses of inclisiran vs. placebo	180 days	LDL-C 27.9% (200mg inclisiran); LDL-C 38.4% (300mg inclisiran); LDL-C 41.9% (500mg inclisiran); LDL-C 35.5% (double dose 100mg inclisiran); LDL-C 44.9% (double dose 200mg inclisiran); LDL-C 52.6% (double dose 300mg inclisiran)	NE
ORION-3 (2023) [122]	382	Patients at high risk for CV disease and elevated LDL-C	Inclisiran-only (patients who already received inclisiran continued to receive it) vs. switching-arm (patients who received placebo started receiving evolocumab for 1 year and then	4 years	LDL-C 44.2%	NE

 doi:10.20944/preprints202312.1195.v1

			switched to inclisiran)			
ORION-5 (2023) [123]	56	HoFH	Inclisiran 300 mg vs. placebo	150 days	LDL-C 1.68% PCSK9 60.6%	NE
ORION-9 (2020) [124]	482	HeFH	Inclisiran 300 mg vs. placebo	510 days	LDL-C 39.7%	NE
ORION-10 and ORION-11(2020) [125]	1,561 (ORION-10) and 1,617 (ORION-11)	Patients with atherosclerotic CV disease (ORION-10 trial) and patients with atherosclerotic CV disease or an atherosclerotic CV disease risk equivalent (ORION-11 trial) and elevated LDL-C despite receiving statin therapy at the maximum tolerated dose	Inclisiran 284 mg vs. placebo	510 days	LDL-C 52.3% (ORION-10) LDL-C 49.9% (ORION-11)	NE

13.2. ANGPTL3 inhibitors

ANGPTL3 is a type of protein from the group of angiopoietin-like proteins, produced exclusively by the liver and which acts by inhibiting lipoprotein lipase (LPL) – the enzyme responsible for catalyzing the hydrolysis of plasma triglycerides – and endothelial lipase, which appears to have a fundamental role in lipoprotein metabolism, cytokine expression and cellular lipid composition [129]. Two other angiopoietin-like proteins act in a complementary way to ANGPTL3: ANGPTL4, which is produced in different types of cells and acts mainly to inhibit LPL during fasting; and ANGPTL8, produced by the liver and adipose tissue, which, unlike ANGPTL3 and ANGPTL4, does not have the property of inhibiting LPL on its own, but can contribute to the increase or decrease of LPL inhibition through the complexes formed. The ANGPTL3/ANGPTL8 complex substantially increases affinity with LPL and creates a potent inhibitor of plasma triglyceride clearance, while the ANGPTL4/ANGPTL8 complex neutralizes the inhibitory action of the ANGPTL3/ANGPTL8 complex [130-132].

Preclinical study of a monoclonal antibody targeting the C-terminal LPL inhibitory domain of ANGPTL3 was published in 2015, which resulted in increased LPL activity and reduced triglycerides, LDL-C and HDL-C in rats and monkeys [133].

 doi:10.20944/preprints202312.1195.v1

Loss-of-function genetic variants of ANGPTL3 are associated with low levels of LDL-C, triglycerides and a lower risk of coronary artery disease despite the presence of low levels of HDL-C. In the DiscovEHR human genetics study, loss-of-function variants in ANGPTL3 were found in 0.33% of case patients with CAD and in 0.45% of controls (adjusted OR 0.59, 95% CI 0.41 to 0.85, p=0.004) [134].

In 2020, the Evinacumab Lipid Studies in Patients with Homozygous Familial Hypercholesterolemia (ELIPSE HoFH) trial evaluated the efficacy of evinacumab, a monoclonal antibody against ANGPTL3, in reducing LDL-C in patients with a genetic or clinical diagnosis of HoFH [135]. Even with the inclusion of patients without the need for genetic testing to confirm HoFH (32% of individuals had a clinical diagnosis), only 65 patients were included and randomized in a 2:1 ratio to receive an intravenous infusion of evinacumab every 4 weeks or placebo. At week 24, patients in the evinacumab group had a relative reduction from baseline in the LDL-C of 47.1%, as compared with an increase of 1.9% in the placebo group. Almost all of the trial patients (94%) were receiving a statin (a high-intensity statin in 77%). In addition, a PCSK9 inhibitor was being administered in 77% of the patients, ezetimibe in 75%, and lomitapide in 25%; 34% of the patients underwent apheresis. A total of 63% of the patients were taking at least three lipid modifying drugs. Even so, the placebo group showed a 1.9% increase in LDL-C when compared to baseline after 6 months. This data strengthens the hypothesis that current therapy (mainly LDLR-targeting therapies) is little or not effective for treating HoFH, especially those with null/null mutations.

The use of evinacumab was also evaluated in patients with refractory hypercholesterolemia, defined as an LDL-C level of 70 mg/dL or higher with clinical ASCVD or a level of 100 mg/dL or higher without clinical ASCVD and refractory to treatment with a PCSK9 inhibitor and a statin at a maximum tolerated dose, with or without ezetimibe; 72% of the sample had a diagnosis of HeFH [136]. After week 16 of use of evinacumab or placebo, both subcutaneous and intravenous administration of evinacumab resulted in a reduction of between 40 and 50% of LDL-C compared to baseline levels, versus an increase of 8.8% in the placebo group of the subcutaneous regimen and 0.6% of the placebo group of the intravenous regimen. However, this trial was small and underpowered for clinical outcomes.

To date, all clinical trials involving evinacumab have only evaluated the drug's efficacy and safety over a short period of time. These studies have confirmed the findings from animal models that the medication is not only capable of reducing LDL-C, but also triglycerides and HDL-C. The implications of concomitant HDL-C reduction remain to be explained, but in patients with genetic ANGPTL3 deficiency, reduced HDL-C levels are not associated with an increased risk of cardiovascular disease [134].

13.3. CRISPR/Cas9

In 2012, Doudna and Charpentier developed the gene editing technique called CRISPR/Cas9 [137]. CRISPR/Cas9 is the name of a molecular biology technique capable of editing (removing, adding, exchanging) DNA sequences located in any region of the genome. This technique is based on an immunological memory system present in bacteria, used to protect them from virus invasion [138,139].

The Clustered Regularly Interspaced Short Palindromic Repeat Associated System 9 (CRISPR/Cas9) is a promising tool for clinical applications in the treatment of genetic diseases such as FH [140].

Zhao et al. utilized a recombinant adeno-associated virus (AAV) vector carrying the CRISPR/Cas9 gene editor (AAV-CRISPR/Cas9) in LDLR loss-of-function mutant mice that exhibited severe atherosclerotic phenotypes when fed a diet rich in fat in vivo animal study [141]. AAV-CRISPR/Cas9-mediated gene editing partially corrected the point mutation in the LDLR gene expressed in hepatocytes and restored partial expression of the LDLR protein, and significantly reduced total cholesterol, triglycerides and LDL-C.

 doi:10.20944/preprints202312.1195.v1

Wang et al. showed, through a LDLR-deficient mouse model (Ldlr-/-, Apobec1-/-, double knockout), that AAV carrying an LDLR transgene at vector doses as low as 3×10^{11} increased transgene expression and decreased LDL-C [142].

13.4. Antisense Oligonucleotides (ASO)

ASO are analogues of single chains of nucleic acids, complementary to a specific mRNA (Watson-Crick hybridization), capable of inhibiting its expression and blocking protein synthesis. This technology is used to treat different diseases, including hypercholesterolemia.

13.4.1. Mipomersen

Apolipoprotein B (apoB) is the protein part of the lipoproteins considered atherogenic: VLDL-C, IDL-C and LDL-C. There are two forms of apoB: apoB-49, which is synthesized in enterocytes and is present in chylomicrons, and apoB-100, which is synthesized in hepatocytes and is present in VLDL and, consequently, in IDL and LDL.

The mutation in apoB that leads to a reduction in its synthesis, as in hypobetalipoproteinemia, also leads to a reduction in LDL-C levels. Therefore, inhibition of apoB synthesis could be a therapeutic target for reducing LDL-C.

Mipomersen is a second-generation antisense oligonucleotide that binds to the messenger RNA that encodes apoB 100, thus reducing its production.

A post hoc analysis of collected data from three RCTs showed that long-term mipomersen reduced cardiovascular events in patients with FH [143].

On the other hand, meta-analysis including 5 RCTs showed that mipomersen probably reduces LDL-C compared with placebo (mean difference: -24.79, 95% CI -30.15 to -19.43) but with a moderate level of certainty [144].

13.4.2. Volanesorsen and Olezarsen

Volanesorsen, a second-generation ASO, binds at base position 489-508 of the ApoC-III mRNA, allowing ribonuclease H1-mediated RNA degradation and consequently decreased ApoC-III synthesis. Its lipid-lowering action is LPL-independent [145].

The APPROACH study showed a mean decrement of TG levels of 77% in familial chylomicronemia patients [146]. The COMPASS study achieved similar results in patients who had TG levels >500 mg/dL [147].

The BROADEN trial assessed the impact of the drug in 15 familial partial lipodystrophy patients. TG levels were decreased by 69%. Insulin action increased by 50% and hemoglobin A1c levels were decreased by 0.44% [148].

Olezarsen is another ASO targeting APOC-III, but differently from Volanesorsen, it is an N-acetyl-galactosamine (GalNAc)-conjugated ASO, presenting higher binding capacity and affinity with hepatic receptors. Therefore, lower doses may be necessary for a clinically significant effect, favoring safety. In a phase 2 study including individuals with high triglycerides, olezarsen promoted reduction of triglycerides in up to 60% relative to placebo, with mild injection-site reactions being the most common adverse event [149].

13.5. apoB and MTP Inhibitors

In recent years, the value of apoB – an important structural component of LDL-C – as a marker of CV risk has increased through epidemiological studies, clinical trials and Mendelian randomization analyses [150-157]. Based on this evidence, the European Society of Cardiology recommended apoB as a more accurate marker of CV risk when compared to LDL-C and non-HDL-C and a more accurate parameter for lipid-lowering therapy [158].

In 2006, the first human study of an antisense oligonucleotide targeting hepatic apoB mRNA to inhibit its synthesis was published, which was later named mipomersen. The reduction in apoB compared to baseline after 83 days of the first dose was 50% [159]. Meta-analysis observed an average

 doi:10.20944/preprints202312.1195.v1

reduction of 33% in apoB with the use of mipomersen with an associated reduction in LDL-C, non-HDL-C, triglycerides and Lp(a) [160]. More recently, another meta-analysis observed a 26% reduction in LDL-C in patients using mipomersen [161].

Although there was no significant discontinuation in the first study (even with 72% of the population presenting injection-site reactions), when used in studies with a larger number of patients and longer treatment duration, important adverse effects were consistently associated with mipomersen, since injection-site reactions up to flu-like symptoms and liver injury due to steatosis, which compromise its widespread use, currently being restricted to cases of HoFH with low response to other safer therapies, such as statins and PCSK9 inhibitors [161].

13.6. Inducible degrader of LDLR (IDOL)

IDOL (inducible degrader of the low-density lipoprotein receptor, LDLR) is an E3 ubiquitin ligase (a kind of enzyme which can covalently combine with the substrates of various ubiquitin proteins and promotes the degradation of the substrate proteins) regulated by liver X receptor (LXR) that promotes the ubiquitination and degradation of the LDLR through lysosomal degradation by binding to the cytoplasmic region of LDLR, regulating cholesterol metabolism through the LXR-IDOL-LDLR axis [166]. The activation of LXR by oxysterol ligands increases transcription of genes whose protein products work to reduce intracellular cholesterol levels [162]. Treatment of various cell types including macrophages and hepatocytes with LXR agonists markedly inhibits the binding and uptake of LDL by these cells. LXR activation was shown to lead to rapid elimination of LDLR protein from the cell surface. The importance of the LXR-IDOL-LDLR axis in vivo was provided by the observation that LXR agonists reduce LDLR protein levels in mice in a tissue-specific manner depending on the degree of IDOL induction [163].

Since the discovery of the IDOL gene in 2009 [164], population-based observational studies have shown that its genetic variants are strongly associated with plasma lipid levels [165,166]. Consequently, the search for compounds capable of silencing IDOL and consequently reducing LDLR degradation is ongoing.

14. Lomitapide

A microsomal triglyceride transfer protein (MTP) is a product of the MTTP gene and is essential for the mounting and secretion of apoB-containing lipoproteins. Genetic MTP deficiency is associated with abetalipoproteinemia, characterized by the virtual absence of apoB-containing lipoproteins in the circulation. Lomitapide is a selective inhibitor of MTP, responsible for transferring neutral lipids, predominantly triglyceride, to triglyceride-rich lipoproteins (TGRLs) intracellularly - chylomicrons in the intestine and very low-density lipoproteins (VLDL) in hepatocytes [167,168].

MTP becomes attractive as a therapeutic target because it potentially inhibits the production of TGRLs at the source (decreasing the formation of chylomicrons in the intestine and VLDL in the liver), without acting on other targets known as LDLR and LPL. However, this inhibition can be asymmetrical from a harmful point of view, causing damage due to the lack of absorption of fats (diarrhea and gastrointestinal symptoms) and the formation of VLDL in the liver (causing fat accumulation and steatosis). It is precisely these adverse effects that made it impossible to widely use lomitapide, a selective MTP inhibitor, which in a dose-escalation study to examine the safety, tolerability, and effects on lipid levels, reduced LDL-C by 50.9% and 55.6% apoB when comparing the levels after 4 weeks of use with the baseline levels [169].

Phase 3 study evaluated the mean reduction in LDL-C after starting the use of lomitapide in patients with HoFH and observed a 50% reduction in LDL-C in 26 weeks and progressively (at a lower mean percentage) until week 78 [170]. The main adverse effects were gastrointestinal symptoms (abdominal cramps, nausea, vomiting and diarrhea).

However, there are no randomized clinical trials that have evaluated the cardiovascular effects of lomitapide.

 doi:10.20944/preprints202312.1195.v1

15. Vaccines against PCSK9

Vaccination to produce antibodies against self-antigens has been shown to be effective in several diseases, such as cancer and hypertension. Vaccines against PCSK9 stimulate the immune system to produce specific antibodies against the PCSK9 protein. These antibodies are designed to bind to PCSK9 and neutralize it, preventing it from carrying out its normal function of regulating cholesterol levels. When PCSK9 is neutralized by antibodies produced in response to vaccination, the amount of PCSK9 available to bind to LDL receptors on cells is reduced. This results in an increase in the amount of receptors available to remove LDL-C from the bloodstream, helping to lower circulating LDL-C levels.

Preclinical studies have shown beneficial effects of inducing anti-PCSK9 antibody production by special epitope/peptide vaccines. However, clinical studies are still scarce.

Several experimental studies have been published with positive results. Landlinger et al. showed that the peptide-based AT04A vaccine promoted a sustained reduction in PCSK9 levels (≥49%) over 12 weeks [172].

L-IFPTA+ is a vaccine that incorporates an immunogenic peptide onto the surface of negatively charged nanoliposomes along with the adjuvant Alhydrogel®. Momtazi-Borojeni et al. showed that injections of L-IFPTA+ decreased PCSK9 plasma concentrations by up to 58.5%. After 8 weeks, LDL-C and TC levels were reduced by 51.7% and 44.7%, respectively, compared to controls [173].

Other studies have also shown success in lowering LDL-C levels through the use of PCSK9 vaccines [174-177].

Momtazi-Borojeni et al. showed that nanoliposomal anti-PCSK9 vaccines induced safe, durable and functional PCSK9-specific antibodies in hypercholesterolemic C57BL/6 mice [178].

Recently, Fowler et al. evaluated the effectiveness of virus-like particle (VLP)-based vaccines targeting epitopes in the LDL-R domain of PCSK9 [179]. VLP vaccine reduced LDL-C levels in combination with statins, whereas immunization with bivalent vaccine reduced LDL-C levels without requiring statin.

This approach aims to offer long-term control over cholesterol levels, potentially decreasing the need for frequent treatments. However, the development of these vaccines requires extensive studies to ensure their safety, effectiveness and long-term impact on patients' health.

16. Plasmapheresis

In 1980, the long-term effects of plasmapheresis for the treatment of hypercholesterolemia in two children were first reported [180]. Since then, several clinical trials have demonstrated the effectiveness and safety of the method, especially in children and patients with hypertriglyceridemia.

The most commonly used apheresis methods for removing lipoproteins from the blood are: (1) double filtration; (2) LDL-C adsorption due to binding based on immunoaffinity; (3) LDL-C adsorption due to binding based on immunoaffinity; (4) heparin-induced precipitation; and (5) direct adsorption of lipoproteins (DALI) [181].

Recently, retrospective analysis by Albayrak et al. showed that double filtration plasmapheresis reduced the levels of Lp(a), total cholesterol, LDL-C, HDL-C and triglycerides in patients with FH [182].

Plasmapheresis is an effective method for removing triglycerides in patients with severe hypertriglyceridemia. However, the studies that evaluated the safety of the method, especially in patients with hypertriglyceridemia-associated acute pancreatitis (HTG-AP) are small, retrospective, and often included mild cases.

Prior analysis of data collected for the PERFORM study showed that, after adjusting for confounders, plasmapheresis was not associated with the incidence and duration of organ failure, but with increased intensive care unit requirements in patients with hypertriglyceridemia-associated acute pancreatitis [183].

The ongoing Intensive insulin therapy versus plasmapheresis in the management of hypertriglyceridemia-induced acute pancreatitis (Bi-TPAI) trial aims to evaluate whether intensive insulin therapy is non-inferior to plasmapheresis in patients with HTG-AP [184].

 doi:10.20944/preprints202312.1195.v1

17. Conclusion

Advances in cholesterol-lowering therapy, especially in the context of FH, have highlighted the importance of genetic interventions and the development of specific action medications. Inclisiran, for example, with its action aimed at the lasting reduction of LDL-C, represents a paradigm shift, offering less frequent treatments with sustained effects.

Future perspectives suggest an increasing focus on personalized therapies, based on the patient's genetic profile. Furthermore, the search for drugs that can act on multiple targets for even greater efficacy and with a lower incidence of side effects has been a growing focus in research.

The integration of new genetic discoveries, advances in technology and an in-depth understanding of the mechanisms of action of drugs will allow the development of more effective therapies, reducing the cardiovascular risk of patients with dyslipidemia. However, further studies are needed to validate the long-term safety and efficacy of these new therapeutic approaches.

References

1. Nelson, D.L.; Cox, M.M. Lehninger Principles of Biochemistry, (5th Ed.). New York: W. H. Freeman and Company 2008, p. 343-389, p.831-850.
2. Mach, F.; Baigent, C.; Catapano, A.L.; Koskinas, K.C.; Casula, M.; Badimon, L.; Chapman, M.J.; De Backer, G.G.; Delgado, V.; Ference, B.A.; et al. 2019 ESC/EAS Guidelines for the management of dyslipidaemias: lipid modification to reduce cardiovascular risk. Eur. Heart J. 2020, 41(1), 111-188.
3. Grundy, S.M.; Stone, N.J.; Bailey, A.L.; Beam, C.; Birtcher, K.K.; Blumenthal, R.S.; Braun, L.T.; de Ferranti, S.; Faiella-Tommasino, J.; Forman, D.E.; et al. 2018AHA/ACC/AACVPR/AAPA/ABC/ACPM/ADA/AGS/APhA/ASPC/NLA/PCNA Guideline on the Management of Blood Cholesterol: Executive Summary: A Report of the American College of Cardiology/American Heart Association Task Force on Clinical Practice Guidelines. J. Am. Coll. Cardiol. 2019, 73(24), 3168-3209.
4. Shepherd, J.; Cobbe, S.M.; Ford, I.; Isles, C.G.; Lorimer, A.R.; MacFarlane, P.W.; McKillop, J.H.; Packard, C.J. Prevention of coronary heart disease with pravastatin in men with hypercholesterolemia. West of Scotland Coronary Prevention Study Group. N. Engl. J. Med. 1995, 333(20), 1301-1307.
5. Downs, J.R.; Clearfield, M.; Weis, S.; Whitney, E.; Shapiro, D.R.; Beere, P.A.; Langendorfer, A.; Stein, E.A.; Kruyer, W.; Gotto, A.M.Jr. Primary prevention of acute coronary events with lovastatin in men and women with average cholesterol levels: results of AFCAPS/TexCAPS. Air Force/Texas Coronary Atherosclerosis Prevention Study. J.A.M.A. 1998, 279(20), 1615-1622.
6. Bassler, D.; Briel, M.; Montori, V.M.; Lane, M.; Glasziou, P.; Zhou, Q.; Heels-Ansdell, D.; Walter, S.D.; Guyatt, G.H.; STOPIT-2 Study Group; Flynn, D.N.; et al. Stopping randomized trials early for benefit and estimation of treatment effects: systematic review and meta-regression analysis. J.A.M.A. 2010, 303(12), 1180-1187.
7. Sever, P.S.; Dahlöf, B.; Poulter, N.R.; Wedel, H.; Beevers, G.; Caulfield, M.; Collins, R.; Kjeldsen, S.E.; Kristinsson, A.; McInnes, G.T.; et al; ASCOT investigators. Prevention of coronary and stroke events with atorvastatin in hypertensive patients who have average or lower-than-average cholesterol concentrations, in the Anglo-Scandinavian Cardiac Outcomes Trial-Lipid Lowering Arm (ASCOT-LLA): a multicentre randomised controlled trial. Lancet 2003, 361(9364), 1149-1158.
8. Nakamura, H.; Arakawa, K.; Itakura, H.; Kitabatake, A.; Goto, Y.; Toyota, T.; Nakaya, N.; Nishimoto, S.; Muranaka, M.; Yamamoto, A.; et al; MEGA Study Group. Primary prevention of cardiovascular disease with pravastatin in Japan (MEGA Study): a prospective randomised controlled trial. Lancet 2006, 368(9542), 1155-1163.
9. Ridker, P.M.; Danielson, E.; Fonseca, F.A.; Genest, J.; Gotto, A.M. Jr.; Kastelein, J.J.; Koenig, W.; Libby, P.; Lorenzatti, A.J.; MacFadyen, J.G.; et al; JUPITER Study Group. Rosuvastatin to prevent vascular events in men and women with elevated C-reactive protein. N. Engl. J. Med. 2008, 359(21), 2195-2207.
10. Yusuf, S.; Bosch, J.; Dagenais, G.; Zhu, J.; Xavier, D.; Liu, L.; Pais, P.; López-Jaramillo, P.; Leiter, L.A.; Dans, A.; et al; HOPE-3 Investigators. Cholesterol Lowering in Intermediate-Risk Persons without Cardiovascular Disease. N. Engl. J. Med. 2016, 374(21), 2021-2031.
11. Randomised trial of cholesterol lowering in 4444 patients with coronary heart disease: the Scandinavian Simvastatin Survival Study (4S). Lancet 1994, 344(8934), 1383-1389.
12. Sacks, F.M.; Pfeffer, M.A.; Moye, L.A.; Rouleau, J.L.; Rutherford, J.D.; Cole, T.G.; Brown, L.; Warnica, J.W.; Arnold, J.M.; Wun, C.C.; et al. The effect of pravastatin on coronary events after myocardial infarction in patients with average cholesterol levels. Cholesterol and Recurrent Events Trial investigators. N. Engl. J. Med. 1996, 335(14), 1001-1009.

 doi:10.20944/preprints202312.1195.v1

13. Long-Term Intervention with Pravastatin in Ischaemic Disease (LIPID) Study Group. Prevention of cardiovascular events and death with pravastatin in patients with coronary heart disease and a broad range of initial cholesterol levels. N. Engl. J. Med. 1998, 339(19), 1349-1357.
14. Heart Protection Study Collaborative Group. MRC/BHF Heart Protection Study of cholesterol lowering with simvastatin in 20,536 high-risk individuals: a randomised placebo-controlled trial. Lancet 2002, 360(9326), 7-22.
15. Liem, A.; van Boven, A.J.; Withagen, A. P.; Robles de Medina, R.M.; Veeger, N.J.; Tijssen, J. G. Fluvastatin in Acute Myocardial Infarction: Effects on Early and Late Ischemia and Events: the FLORIDA Trial. Circulation 2000, 102, 2672.
16. Liem, A.H.; van Boven, A.J.; Veeger, N.J.; Withagen, A. J.; Robles de Medina, R.M.; Tijssen, J. G.; van Veldhuisen, D.J.; FLuvastatin On Risk Reduction after Acute myocardial infarction study group. Effect of fluvastatin on ischaemia following acute myocardial infarction: a randomized trial. Eur. Heart J. 2002, 23(24), 1931-1937.
17. Cannon, C.P.; Braunwald, E.; McCabe, C.H.; Rader, D.J.; Rouleau, J.L.; Belder, R.; Joyal, S.V.; Hill, K.A.; Pfeffer, M.A.; Skene, A.M.; Pravastatin or Atorvastatin Evaluation and Infection Therapy-Thrombolysis in Myocardial Infarction 22 Investigators. Intensive versus moderate lipid lowering with statins after acute coronary syndromes. N. Engl. J. Med. 2004, 350(15), 1495-1504.
18. Pedersen, T.R.; Faergeman, O.; Kastelein, J.J.; Olsson, A.G.; Tikkanen, M.J.; Holme, I.; Larsen, M.L.; Bendiksen, F.S.; Lindahl, C.; Szarek, M.; Incremental Decrease in End Points Through Aggressive Lipid Lowering (IDEAL) Study Group. High-dose atorvastatin vs usual-dose simvastatin for secondary prevention after myocardial infarction: the IDEAL study: a randomized controlled trial. J.A.M.A. 2005, 294(19), 2437-2445.
19. LaRosa, J.C.; Grundy, S.M.; Waters, D.D.; Shear, C.; Barter, P.; Fruchart, J.C.; Gotto, A.M.; Greten, H.; Kastelein, J.J.; Shepherd, J.; et al; Treating to New Targets (TNT) Investigators. Intensive lipid lowering with atorvastatin in patients with stable coronary disease. N. Engl. J. Med. 2005, 352(14), 1425-1435.
20. Study of the Effectiveness of Additional Reductions in Cholesterol and Homocysteine (SEARCH) Collaborative Group; Armitage, J.; Bowman, L.; Wallendszus, K.; Bulbulia, R.; Rahimi, K.; Haynes, R.; Parish, S.; Peto, R.; Collins, R. Intensive lowering of LDL cholesterol with 80 mg versus 20 mg simvastatin daily in 12,064 survivors of myocardial infarction: a double-blind randomised trial. Lancet 2010, 376(9753), 1658-1669.
21. Colhoun, H.M.; Betteridge, D.J.; Durrington, P.N.; Hitman, G.A.; Neil, H.A.; Livingstone, S.J.; Thomason, M.J.; Mackness, M.I.; Charlton-Menys, V.; Fuller, J.H.; CARDS investigators. Primary prevention of cardiovascular disease with atorvastatin in type 2 diabetes in the Collaborative Atorvastatin Diabetes Study (CARDS): multicentre randomised placebo-controlled trial. Lancet 2004, 364(9435), 685-696.
22. Knopp, R.H.; d'Emden, M.; Smilde, J.G.; Pocock, S.J. Efficacy and safety of atorvastatin in the prevention of cardiovascular end points in subjects with type 2 diabetes: the Atorvastatin Study for Prevention of Coronary Heart Disease Endpoints in non-insulin-dependent diabetes mellitus (ASPEN). Diabetes Care 2006, 29(7), 1478-1485.
23. Collins, R.; Armitage, J.; Parish, S.; Sleigh, P.; Peto, R.; Heart Protection Study Collaborative Group. MRC/BHF Heart Protection Study of cholesterol-lowering with simvastatin in 5963 people with diabetes: a randomised placebo-controlled trial. Lancet 2003, 361(9374), 2005-2016.
24. Sever, P.S.; Poulter, N.R.; Dahlöf, B.; Wedel, H.; Collins, R.; Beevers, G.; Caulfield, M.; Kjeldsen, S.E.; Kristinsson, A.; McInnes, G.T.; et al. Reduction in cardiovascular events with atorvastatin in 2,532 patients with type 2 diabetes: Anglo-Scandinavian Cardiac Outcomes Trial-lipid-lowering arm (ASCOT-LLA). Diabetes Care 2005, 28(5), 1151-1157.
25. Holdaas, H.; Fellström, B.; Jardine, A.G.; Holme, I.; Nyberg, G.; Fauchald, P.; Grönhagen-Riska, C.; Madsen, S.; Neumayer, H. H.; Cole, E.; et al; Assessment of LEscol in Renal Transplantation (ALERT) Study Investigators. Effect of fluvastatin on cardiac outcomes in renal transplant recipients: a multicenter, randomized, placebo-controlled trial. Lancet 2003, 361(9374), 2024-2031.
26. Wanner, C.; Krane, V.; März, W.; Olschewski, M.; Mann, J.F.; Ruf, G.; Ritz, E.; German Diabetes and Dialysis Study Investigators. Atorvastatin in patients with type 2 diabetes mellitus undergoing hemodialysis. N. Engl. J. Med. 2005, 353(3), 238-248.
27. Fellström, B.C.; Jardine, A.G.; Schmieder, R.E.; Holdaas, H.; Bannister, K.; Beutler, J.; Chae, D.W.; Chevaile, A.; Cobbe, S.M.; Grönhagen-Riska, C.; et al; AURORA Study Group. Rosuvastatin and cardiovascular events in patients undergoing hemodialysis. N. Engl. J. Med. 2009, 360(14), 1395-1407.
28. Baigent, C.; Landray, M.J.; Reith, C.; Emberson, J.; Wheeler, D.C.; Tomson, C.; Wanner, C.; Krane, V.; Cass, A.; Craig, J.; et al; SHARP Investigators. The effects of lowering LDL cholesterol with simvastatin plus ezetimibe in patients with chronic kidney disease (Study of Heart and Renal Protection): a randomised placebo-controlled trial. Lancet 2011, 377(9784), 2181-2192.

29. Kjekshus, J.; Apetrei, E.; Barrios, V.; Böhm, M.; Cleland, J.G.; Cornel, J.H.; Dunselman, P.; Fonseca, C.; Goudev, A.; Grande, P.; et al; CORONA Group. Rosuvastatin in older patients with systolic heart failure. N. Engl. J. Med. 2007, 357(22), 2248-2261.
30. Tavazzi, L.; Maggioni, A.P.; Marchioli, R.; Barlera, S.; Franzosi, M.G.; Latini, R.; Lucci, D.; Nicolosi, G.L.; Porcu, M.; Tognoni, G.; Gissi-HF Investigators. Effect of rosuvastatin in patients with chronic heart failure (the GISSI-HF trial): a randomised, double-blind, placebo-controlled trial. Lancet 2008, 372(9645), 1231-1239.
31. Shepherd, J.; Blauw, G.J.; Murphy, M.B.; Bollen, E.L.; Buckley, B.M.; Cobbe, S.M.; Ford, I.; Gaw, A.; Hyland, M.; Jukema, J.W.; et al; PROSPER study group. PROspective Study of Pravastatin in the Elderly at Risk. Pravastatin in elderly individuals at risk of vascular disease (PROSPER): a randomised controlled trial. Lancet 2002, 360(9346), 1623-1630.
32. Grinspoon, S,K.; Fitch, K.V.; Zanni, M.V.; Fichtenbaum, C.J.; Umbleja, T.; Aberg, J.A.; Overton, E.T.; Malvestutto, C.D.; Bloomfield, G.S.; Currier, J.S.; et al; REPRIEVE Investigators. Pitavastatin to Prevent Cardiovascular Disease in HIV Infection. N. Engl. J. Med. 2023, 389(8), 687-699.
33. Cholesterol Treatment Trialists' (CTT) Collaboration; Baigent, C.; Blackwell, L.; Emberson, J.; Holland, L.E.; Reith, C.; Bhala, N.; Peto, R.; Barnes, E.H.; Keech, A.; Simes, J.; et al. Efficacy and safety of more intensive lowering of LDL cholesterol: a meta-analysis of data from 170,000 participants in 26 randomised trials. Lancet 2010, 376(9753), 1670-1681.
34. Tsouka, A.N.; Tellis, C.C.; Tselepis, A.D. Pharmacology of PCSK9 Inhibitors: Current Status and Future Perspectives. Curr. Pharm. Des. 2018, 24(31), 3622-3633.
35. Gaudet, D.; Kereiakes, D.J.; McKenney, J.M.; Roth, E.M.; Hanotin, C.; Gipe, D.; Du, Y.; Ferrand, A.C.; Ginsberg, H.N.; Stein, E.A. Effect of alirocumab, a monoclonal proprotein convertase subtilisin/kexin 9 antibody, on lipoprotein(a) concentrations (a pooled analysis of 150 mg every two weeks dosing from phase 2 trials). Am. J. Cardiol. 2014, 114(5), 711-715.
36. Sabatine, M.S.; Giugliano, R.P.; Keech, A.C.; Honarpour, N.; Wiviott, S.D.; Murphy, S.A.; Kuder, J.F.; Wang, H.; Liu, T.; Wasserman, S.M.; et al; FOURIER Steering Committee and Investigators. Evolocumab and Clinical Outcomes in Patients with Cardiovascular Disease. N. Engl. J. Med. 2017, 376(18), 1713-1722.
37. Schwartz, G.G.; Steg, P.G.; Szarek, M.; Bhatt, D.L.; Bittner, V.A.; Diaz, R.; Edelberg, J.M.; Goodman, S.G.; Hanotin, C.; Harrington, R.A.; et al; ODYSSEY OUTCOMES Committees and Investigators. Alirocumab and Cardiovascular Outcomes after Acute Coronary Syndrome. N. Engl. J. Med. 2018, 379(22), 2097-2107.
38. O'Donoghue, M.L.; Giugliano, R.P.; Wiviott, S.D.; Atar, D.; Keech, A.; Kuder, J.F.; Im, K.; Murphy, S.A.; Flores-Arredondo, J.H.; López, J.A.G.; et al. Long-Term Evolocumab in Patients With Established Atherosclerotic Cardiovascular Disease. Circulation 2022, 146(15), 1109-1119.
39. Cui, Y.; Zhao, X.; Qi, L.; Li, X.; Huang, Z.; Yang, G.; Qian, L.; Deng, H.; Li, H.; Huo, Y. A potential long-acting LDL-cholesterol-lowering PCSK9 monoclonal antibody: randomized, placebo-controlled phase 1 studies. J.A.C.C. Asia 2021, 1, 411-415.
40. Huo, Y.; Chen, B.; Lian, Q.; Wang, S.; Liu, L.; Lu, D.; Qu, Y.; Zheng, G.; Li, L.; Ji, Y.; et al. Tafolecimab in Chinese patients with non-familial hypercholesterolemia (CREDIT-1): a 48-week randomized, double-blind, placebo-controlled phase 3 trial. Lancet Reg. Health West Pac. 2023, 41, 100907.
41. Chai, M.; He, Y.; Zhao, W.; Han, X.; Zhao, G.; Ma, X.; Qiao, P.; Shi, D.; Liu, Y.; Han, W.; et al.; CREDIT-2 investigators. Efficacy and safety of tafolecimab in Chinese patients with heterozygous familial hypercholesterolemia: a randomized, double-blind, placebo-controlled phase 3 trial (CREDIT-2). B.M.C. Med. 2023, 21(1), 77.
42. Stein, E.; Toth, P.; Butcher, M.B.; Kereiakes, D.; Magnu, P.; Bays, H.; Zhou, R.; Turner, T. A. Safety, tolerability and LDL-C reduction with a novel anti-PCSK9 recombinant fusion protein (LIB003): results of a randomized, double-blind, placebo controlled, phase 2 study [abstract]. Atherosclerosis 2019, 287, e7.
43. Stein, E.A.; Turner, T.; Kereiakes, D.J.; Butcher, B.; Mangu, P.; Safety, Z.R. Tolerability and LDL-C reduction with LIB003 a novel anti-PCSK9 recombinant fusion protein: results of open-label extension phase 2B study [abstract]. Circulation 2019, 140, A17222.
44. Raal, F.; Fourie, N.; Scott, R.; Blom, R.; De Vries Basson, M.; Kayikcioglu, M.; Caldwell, K.; Kallend, D.; Stein, E.; LIBerate-HeFH Investigators. Long-term efficacy and safety of lerodalcibep in heterozygous familial hypercholesterolaemia: the LIBerate-HeFH trial. Eur. Heart J. 2023, 44(40), 4272-4280.
45. Ridker, P.M.; Lei, L.; Louie, M.J.; Haddad, T.; Nicholls, S.J.; Lincoff, A.M.; Libby, P.; Nissen, S. E. Inflammation and Cholesterol as Predictors of Cardiovascular Events Among 13970 Contemporary High-Risk Patients With Statin Intolerance. Circulation 2023, doi: 10.1161/CIRCULATIONAHA.123.066213
46. Phan, B.A.P.; Dayspring, T.D.; Toth, P.P. Ezetimibe therapy: mechanism of action and clinical update. Vasc. Health Risk Manag. 2012, 8, 415-427.
47. Dujovne, C.A.; Ettinger, M.P.; McNeer, J.F.; Lipka, L.J.; LeBeaut, A.P.; Suresh, R.; Yang, B.; Veltri, E.P.; Ezetimibe Study Group. Efficacy and safety of a potential new selective cholesterol absorption inhibitor, ezetimibe, in patients with primary hypercholesterolemia. Am. J. Cardiol. 2002, 90, 1092-1097.

48. Pearson, T.A.; Denke, M.A.; McBride, P.E.; Battisti, W.P.; Brady, W.E.; Palmisano, J. A community-based, randomized trial of ezetimibe added to statin therapy to attain NCEP ATP III goals for LDL cholesterol in hypercholesterolemic patients: the ezetimibe add-on to statin for effectiveness (EASE) trial. Mayo Clin. Proc. 2005, 80(5), 587-595.
49. Kastelein, J.J.; Akdim, F.; Stroes, E.S.; Zwinderman, A.H.; Bots, M.L.; Stalenhoef, A.F.; Visseren, F.L.; Sijbrands, E.J.; Trip, M.D.; Stein, E.A.; et al; ENHANCE Investigators. Simvastatin with or without ezetimibe in familial hypercholesterolemia. N. Engl. J. Med. 2008, 358(14), 1431-1443.
50. Cannon, C.P.; Blazing, M.A.; Giugliano, R.P.; McCagg, A.; White, J.A.; Theroux, P.; Darius, H.; Lewis, B.S.; Ophuis, T.O.; Jukema, J.W.; et al; IMPROVE-IT Investigators. Ezetimibe Added to Statin Therapy after Acute Coronary Syndromes. N. Engl. J. Med. 2015, 372(25), 2387-2397.
51. Hagiwara, N.; Kawada-Watanabe, E.; Koyanagi, R.; Arashi, H.; Yamaguchi, J.; Nakao, K.; Tobaru, T.; Tanaka, H.; Oka, T.; Endoh, Y.; et al. Low-density lipoprotein cholesterol targeting with pitavastatin + ezetimibe for patients with acute coronary syndrome and dyslipidaemia: the HIJ-PROPER study, a prospective, open-label, randomized trial. Eur. Heart J. 2017, 38(29), 2264-2276.
52. Ouchi, Y.; Sasaki, J.; Arai, H.; Yokote, K.; Harada, K.; Katayama, Y.; Urabe, T.; Uchida, Y.; Hayashi, M.; Yokota, N.; et al. Ezetimibe Lipid-Lowering Trial on Prevention of Atherosclerotic Cardiovascular Disease in 75 or Older (EWTOPIA 75): A Randomized, Controlled Trial. Circulation 2019, 140(12), 992-1003.
53. Lee, Y.J.; Cho, J.Y.; You, S.C.; Lee, Y.H.; Yun, K.H.; Cho, Y.H.; Shin, W.Y.; Im, S.W.; Kang, W.C.; Park, Y.; et al. Moderate-intensity statin with ezetimibe vs. high-intensity statin in patients with diabetes and atherosclerotic cardiovascular disease in the RACING trial. Eur. Heart J. 2023, 44(11), 972-983.
54. Pinkosky, S.L.; Newton, R.S.; Day, E.A.; Ford, R.J.; Lhotak, S.; Austin, R.C.; Birch, C.M.; Smith, B.K.; Filippov, S.; Groot, P.H.E.; et al. Liver-specific ATP-citrate lyase inhibition by bempedoic acid decreases LDL-C and attenuates atherosclerosis. Nat. Commun. 2016, 7, 13457-13457.
55. Nissen, S.E.; Lincoff, A.M.; Brennan, D.; Ray, K.K.; Mason, D.; Kastelein, J.J.P.; Thompson, P.D.; Libby, P.; Cho, L.; Plutzky, J.; et al.; for the CLEAR OUTCOMES Investigators. Bempedoic Acid and Cardiovascular Outcomes in Statin-Intolerant Patients. N. Engl. J. Med. 2023, 388,1353-1364.
56. Ballantyne, C.M.; Laufs, U.; Ray, K.K.; Leiter, L.A.; Bays, H.E.; Goldberg, A.C.; Stroes, E.S.; MacDougall, D.; Zhao, X.; Catapano, A.L. Bempedoic acid plus ezetimibe fixed-dose combination in patients with hypercholesterolemia and high CVD risk treated with maximally tolerated statin therapy. Eur. J. Prev. Cardiol. 2022, 27(6), 593-603.
57. Mutschlechner, D.; Tscharre, M.; Huber, K.; Gremmel, T. Cardiovascular events in patients treated with bempedoic acid vs. placebo: systematic review and meta-analysis. Eur. Heart J. Cardiovasc. Pharmacother. 2023, 9(6), 583-591.
58. Ridker, P.M.; Lei, L.; Ray, K.K.; Ballantyne, C.M.; Bradwin, G.; Rifai, N. Effects of bempedoic acid on CRP, IL-6, fibrinogen and lipoprotein(a) in patients with residual inflammatory risk: A secondary analysis of the CLEAR harmony trial. J. Clin. Lipidol. 2023, 17(2), 297-302.
59. Berg, K. A new serum type system in man - the Lp system. Acta Pathol. Microbiol. Scand. 1963. 59, 369-382.
60. Tsimikas, S. A test in context: Lipoprotein (a): Diagnosis, Prognosis, Controversies, and Emerging Therapies. J. Am. Coll. Cardiol. 2017, 69(6), 692-711.
61. Khera, A.V.; Everett, B.M.; Caulfield, M.P., Hantash, F.M.; Wohlgemuth, J.; Ridker, P.M.; Mora, S. Lipoprotein(a) concentrations, rosuvastatin therapy, and residual vascular risk: an analysis from the JUPITER Trial (Justification for the Use of Statins in Prevention: an Intervention Trial Evaluating Rosuvastatin). Circulation 2014, 129(6), 635-642.
62. Davidson, M.H.; Ballantyne, C.M.; Jacobson, T.A.; Bittner, V.A.; Braun, L.T.; Brown, A.S.; Brown, W.V.; Cromwell, W.C.; Goldberg, R.B.; McKenney, J.M.; et al. Clinical utility of inflammatory markers and advanced lipoprotein testing: Advice from an expert panel of lipid specialists. J. Clin. Lipidol. 2011, 5, 338-367.
63. Catapano, A.L.; Graham, I.; Backer, G.D.; Wiklund, O.; Chapman, M.J.; Drexel, H.; Hoes, A.W.; Jennings, C.S.; Landmesser, U.; Pedersen, T.R.; Reiner, Z.; et al. 2016 ESC/EAS Guidelines for the Management of Dyslipidemias. Eur. Heart J. 2016, 37(39), 2999-3058.
64. Yeang, C.; Hung, M.Y.; Byun, Y.S.; Clopton, P.; Yang, X.; Witztum, J.L.; Tsimikas, S. Effect of therapeutic interventions on oxidized phospholipids on apolipoprotein B100 and lipoprotein(a). J. Clin. Lipidol. 2016, 10(3), 594-603.
65. Yeang, C.; Witztum J.L.; Tsimikas, S. 'LDL-C' = LDL-C + Lp(a)-C: implications of achieved ultra-low LDL-C levels in the proprotein convertase subtilisin/kexin type 9 era of potent LDL-C lowering. Curr. Opin. Lipidol. 2015, 26, 169-178.
66. AIM-HIGH Investigators; Boden, W.E.; Probstfield, J.L.; Anderson, T.; Chaitman, B.R.; Desvignes-Nickens, P.; Koprowicz, K.; McBride, R.; Teo, K.; Weintraub, W. Niacin in patients with low HDL cholesterol levels receiving intensive statin therapy. N. Engl. J. Med. 2011, 365, 2255-2267.

 doi:10.20944/preprints202312.1195.v1

67. Shlipak, M.G.; Simon, J.A.; Vittinghoff, E.; Lin, F.; Barrett-Connor, E.; Knopp, R.H.; Levy, R.I.; Hulley, S.B. Estrogen and Progestin, Lipoprotein(a), and the Risk of Recurrent Coronary Heart Disease Events After Menopause. J.A.M.A. 2000, 283(14), 1845-1852.
68. Tsimikas, S.; Karwatowska-Prokopczuk, E.; Gouni-Berthold, I.; Tardif, J.C.; Baum, S.J.; Steinhagen-Thiessen, E.; Shapiro, M.D.; Stroes, E.S.; Moriarty, P.M.; Nordestgaard, B.G.; et al; AKCEA-APO(a)-LRx Study Investigators. Lipoprotein(a) Reduction in Persons with Cardiovascular Disease. N. Engl. J. Med. 2020, 382(3), 244-255.
69. O'Donoghue, M.L.; Rosenson, R.S.; Gencer, B.; López, J.A.G.; Lepor, N.E.; Baum, S.J.; Stout, E.; Gaudet, D.; Knusel, B.; Kuder, J.F.; et al; OCEAN(a)-DOSE Trial Investigators. Small Interfering RNA to Reduce Lipoprotein(a) in Cardiovascular Disease. N. Engl. J. Med. 2022, 387(20), 1855-1864.
70. Einarsson, K.; Ericsson, S.; Ewerth, S.; Reihner, E.; Rudling, M.; Stahlberg, D.; Angelin, B. Bile acid sequestrants: mechanisms of action on bile acid and cholesterol metabolism. Eur. J. Clin. Pharmacol. 1991, 40(1), S53-58.
71. The Lipid Research Clinics Coronary: Primary Prevention Trial Results. J.A.M.A. 1984, 251(3), 365-374.
72. Heel, R.C.; Brogden, R.N.; Pakes, G.E.; Speight, T.M.; Avery, G.S. Colestipol: a review of its pharmacological properties and therapeutic efficacy in patients with hypercholesterolaemia. Drugs 1980, 19, 161-180.
73. Davidson, M.H.; Dillon, M.A.; Gordon, B.; Jones, P.; Samuels, J.; Weiss, S.; Isaacsohn, J.; Toth, P.; Burke, S.K. Colesevelam hydrochloride (cholestagel): a new, potent bile acid sequestrant associated with a low incidence of gastrointestinal side effects. Arch. Intern. Med. 1999, 159, 1893-1900.
74. Insull, W. Jr; Toth, P.; Mullican, W.; Hunninghake, D.; Burke, S.; Donovan, J.M.; Davidson, M.H. Effectiveness of colesevelam hydrochloride in decreasing LDL cholesterol in patients with primary hypercholesterolemia: a 24-week randomized controlled trial. Mayo Clin. Proc. 2001, 76, 971-982.
75. Ross, S.; D'Mello, M.; Anand, S.S.; Eikelboom, J.; Stewart, A.F.R.; Samani, N.J.; Roberts, R.; Paré, G. The Effect of Bile Acid Sequestrants on the Risk of Cardiovascular Events: A Mendelian Randomization Analysis. Circ. Cardiovasc. Genet. 2015, 8(4), 618-627.
76. Clofibrate and niacin in coronary heart disease. JAMA 1975, 231, 360-381.
77. Carlson, L.A.; Danielson, M.; Ekberg, I.; Klintemar, B.; Rosenhamer, G. Reduction of myocardial reinfarction by the combined treatment with clofibrate and nicotinic acid. Atherosclerosis 1977, 28, 81-86.
78. Blankenhorn, D.H.; Nessim, S.A.; Johnson, R.L.; Sanmarco, M.E.; Azen, S.P.; Cashin-Hemphill, L. Beneficial effects of combined colestipol-niacin therapy on coronary atherosclerosis and coronary venous bypass grafts. JAMA 1987, 257, 3233-3240.
79. Brown, B.G.; Zhao, X.Q.; Chait, A.; Fisher, L.D.; Cheung, M.C.; Morse, J.S.; Dowdy, A.A.; Marino, E.K.; Bolson, E.L.; Alaupovic, P.; et al. Simvastatin and niacin, antioxidant vitamins, or the combination for the prevention of coronary disease. N. Engl. J. Med. 2001, 345, 1583-1589.
80. Taylor, A.J.; Sullenberger, L.E.; Lee, H.J.; Grace, K.A. Arterial biology for the investigation of the treatment effects of reducing cholesterol (ARBITER) 2: a double-blind, placebo-controlled study of extended release niacin on atherosclerosis progression in secondary prevention patients treated with statins. Circulation 2004, 110, 3512-3517.
81. Taylor, A.J.; Villines, T.C.; Stanek, E.J.; Devine, P.J.; Griffen, L.; Miller, M.; Weissman, N.J.; Turco, M. Extended-release niacin or ezetimibe and carotid intima-media thickness. N. Engl. J. Med. 2009, 361, 2113-2122.
82. Whitney, E.J.; Krasuski, R.A.; Personius, B.E.; Michalek, J.E.; Maranian A.M.; Kolasa, M.W.; Monick, E.; Brown, G.; Gotto Jr, A.M. A randomized trial of a strategy for increasing high-density lipoprotein cholesterol levels: effects on progression of coronary heart disease and clinical events. Ann. Intern. Med. 2005, 142, 95-104.
83. HPS2-THRIVE Collaborative Group; Landray, M.J.; Haynes, R.; Hopewell, J.C.; Parish, S.; Aung, T.; Tomson, J.; Wallendszus, K.; Craig, M.; Jiang, L.; Collins, R.; et al: Effects of extended-release niacin with laropiprant in high-risk patients. N. Engl. J. Med. 2014, 371, 203-212.
84. D'Andrea, E.; Hey, S.P.; Ramirez, C.L.; Kesselheim, A.S. Assessment of the Role of Niacin in Managing Cardiovascular Disease Outcomes. J.A.M.A. Netw. Open 2019, 2(4), e192224.
85. Ronsein, G.E.; Vaisar, T.; Davidson, W.S.; Bornfeldt, K.E.; Probstfield, J.L.; O'Brien, K.D.; Zhao, X.Q.; Heinecke, J.W. Niacin Increases Atherogenic Proteins in High-Density Lipoprotein of Statin-Treated Subjects. Arterioscler. Thromb. Vasc. Biol. 2021, 41(8), 2330-2341.
86. Frick, M.H.; Elo, O.; Haapa, K.; Heinonen, O.P.; Heinsalmi, P.; Helo, P.; Huttunen, J.K.; Kaitaniemi, P.; Koskinen, P.; Manninen, V.; et al. Helsinki Heart Study: primary-prevention trial with gemfibrozil in middle-aged men with dyslipidemia. Safety of treatment, changes in risk factors, and incidence of coronary heart disease. N. Engl. J. Med. 1987, 317, 1237-1245.
87. Rubins, H.B.; Robins, S.J.; Collins, D.; Fye, C.L.; Anderson, J.W.; Elam, M.B.; Faas, F.H.; Linares, E.; Schaefer, E.J.; Schectman, G.; et al. Gemfibrozil for the secondary prevention of coronary heart disease in men with low levels of high-density lipoprotein cholesterol. Veterans Affairs High-Density Lipoprotein Cholesterol Intervention Trial Study Group. N. Engl. J. Med. 1999, 341, 410-418.

 doi:10.20944/preprints202312.1195.v1

88. Bezafibrate Infarction Prevention (BIP) study. Secondary prevention by raising HDL cholesterol and reducing triglycerides in patients with coronary artery disease. Circulation 2000, 102, 21-27.
89. Meade, T.; Zuhrie, R.; Cook, C.; Cooper, J. Bezafibrate in men with lower extremity arterial disease: randomised controlled trial. B.M.J. 2002, 325, 1139.
90. Keech, A.; Simes, R.J.; Barter, P.; Best, J.; Scott, R.; Taskinen, M.R.; Forder, P.; Pillai, A.; Davis, T.; Glasziou, P.; et al.; FIELD study investigators. Effects of long-term fenofibrate therapy on cardiovascular events in 9795 people with type 2 diabetes mellitus (the FIELD study): randomised controlled trial. Lancet 2005, 366, 1849-1861.
91. The ACCORD Study Group. Effects of Combination Lipid Therapy in Type 2 Diabetes Mellitus. N. Engl. J. Med. 2010, 362, 1563-1574.
92. Kim, N.H.; Han, K.H.; Choi, J.; Lee, J.; Kim, S.G. Use of fenofibrate on cardiovascular outcomes in statin users with metabolic syndrome: propensity matched cohort study. B.M.J. 2019, 366, l5125.
93. Jakob, T.; Nordmann, A.J.; Schandelmaier, S.; Ferreira-González, I.; Briel, M. Fibrates for primary prevention of cardiovascular disease events. Cochrane Database Syst. Rev. 2016, 11(11), CD009753.
94. Millan, J.; Ointó, X.; Brea, A.; Blasco, M.; Hernández-Mijares, A.; Ascaso, J.; Diaz, A.; Mantilla, T.; Pedro-Botet, J. Fibrates in the secondary prevention of cardiovascular disease (infarction and stroke). Results of a systematic review and meta-analysis of the Cochrane collaboration. Clin. Investig. Arterioscler. 2018, 30(1), 30-35.
95. Das Pradhan, A.; Glynn, R.J.; Fruchart, J-C.; MacFadyen, J.G.; Zaharris, E.S.; Everett, B.M.; Campbell, S.E.; Oshima, R.; Amarenco, P.; Blom, D.J.; et al. Triglyceride Lowering with Pemafibrate to Reduce Cardiovascular Risk. N. Engl. J. Med. 2022, 387, 1923-1934.
96. Le Jossic-Corcos, C.; Gonthier, C.; Zaghini, I.; Logette, E.; Shechter, I.; Bournot, P. Hepatic farnesyl diphosphate synthase expression is suppressed by polyunsaturated fatty acids. Biochem. J. 2005, 385(Pt 3), 787-794.
97. Yokoyama, M.; Origasa, H.; Matsuzaki, M.; Matsuzawa, Y.; Saito, Y.; Ishikawa, Y.; Oikawa, S.; Sasaki, J.; Hishida, H.; Itakura, H.; et al; Japan EPA lipid intervention study (JELIS) Investigators. Effects of eicosapentaenoic acid on major coronary events in hypercholesterolaemic patients (JELIS): a randomized open-label, blinded endpoint analysis. Lancet 2007, 369(9567), 1090-1098.
98. Rauch, B.; Schiele, R.; Schneider, S.; Diller, F.; Victor, N.; Gohlke, H.; Gottwik, M.; Steinbeck, G.; Del Castillo, U.; Sack, R.; et al; OMEGA Study Group. OMEGA, a randomized, placebo-controlled trial to test the effect of highly purified omega-3 fatty acids on top of modern guideline-adjusted therapy after myocardial infarction. Circulation 2010, 122(21), 2152-2159.
99. Manson, J.E.; Cook, N.R.; Lee, I-M.; Christen, W.; Bassuk, S.S.; Mora, S.; for the VITAL Research Group. Marine n-3 Fatty Acids and Prevention of Cardiovascular Disease and Cancer. N. Engl. J. Med. 2019, 380, 23-32.
100. Bhatt, D.L.; Steg, P.G.; Miller, M.; Brinton, E.A.; Jacobson, T.A.; Ketchum, S.B.; Doyle, R.T. Jr.; Juliano, R.A.; Jiao, L.; Granowitz, C.; et al.; REDUCE-IT Investigators. Cardiovascular risk reduction with icosapent ethyl for hypertriglyceridemia. N. Engl. J. Med. 2019, 380(1), 11-22.
101. Ridker, P.M.; Rifai, N.; MacFadyen, J.; Glynn, R.J.; Jiao, L.; Steg, P. G.; Miller, M.; Brinton, E. A.; Jacobson, T.A.; Tardif, J-C.; et al. Effects of randomized treatment with icosapent ethyl and a mineral oil comparator on interleukin-1β, interleukin-6, C-reactive protein, oxidized low-density lipoprotein cholesterol, homocysteine, lipoprotein(a), and lipoprotein-associated phospholipase A2: the REDUCE-IT biomarker substudy. Circulation 2022, 146, 372-379.
102. Nicholls, S.J.; Lincoff, A.M.; Garcia, M.; Bash, D.; Ballantyne, C.M; Barter, P.J.; Davidson, M.H.; Kastelein, J.J.P.; Koenig, W.; McGuire, D.K.; et al. Effect of High-Dose Omega-3 Fatty Acids vs Corn Oil on Major Adverse Cardiovascular Events in Patients at High Cardiovascular Risk. J.A.M.A. 2020, 324(22), 2268-2280.
103. Kalstad, A.A.; Myhre, P.L.; Laake, K.; Tvelt, S.H.; Schmidt, E.B.; Smith P.; Nilsen, D.W.T.; Tveit, A.; Fagerland, M.W.; Solheim, S.; et al.; and On behalf of the OMEMI Investigators. Effects of n-3 Fatty Acid Supplements in Elderly Patients After Myocardial Infarction. Circulation 2021, 143, 528-539.
104. Khan, S.U.; Lone, A.N.; Khan, M.S.; Virani, S.S.; Blumenthal, R.S.; Nasir, K.; Miller, M.; Michos, E.D.; Ballantyne, C.M.; Boden, W.E.; et al. Effect of omega-3 fatty acids on cardiovascular outcomes: A systematic review and meta-analysis. EClinicalMedicine 2021, 38, 100997.
105. Hunt, J.A.; Lu, Z. Cholesteryl ester transfer protein (CETP) inhibitors. Curr. Top. Med. Chem. 2009, 9(5), 419-427.
106. Barter, P.J.; Caulfield, M.; Eriksson, M.; Grundy, S. M.; Kastelein, J.J.; Komajda, M.; Lopez-Sendon, J.; Mosca, L.; Tardif, J.C.; Waters, D.D.; et al; ILLUMINATE Investigators. Effects of torcetrapib in patients at high risk for coronary events. N. Engl. J. Med. 2007, 357(21), 2109-2122.
107. Schwartz, G.G.; Olsson, A. G.; Abt, M.; Ballantyne, C. M.; Barter, P.J.; Brumm, J.; Chaitman, B. R.; Holme, I.M.; Kallend, D.; Leiter, L.A.; et al; dal-OUTCOMES Investigators. Effects of dalcetrapib in patients with a recent acute coronary syndrome. N. Engl. J. Med. 2012, 367(22), 2089-2099.

108. Lincoff, A.M.; Nicholls, S.J.; Riesmeyer, J.S.; Barter, P.J.; Brewer, H. B.; Fox, K.A.A.; Gibson, C. M.; Granger, C.; Menon, V.; Montalescot, G.; et al; ACCELERATE Investigators. Evacetrapib and Cardiovascular Outcomes in High-Risk Vascular Disease. N. Engl. J. Med. 2017, 376(20), 1933-1942.
109. HPS3/TIMI55–REVEAL Collaborative Group; Bowman, L.; Hopewell, J. C.; Chen, F.; Wallendszus, K.; Stevens, W.; Collins, R.; Wiviott, S. D.; Cannon, C.P.; Braunwald, E.; Sammons, E.; et al. Effects of Anacetrapib in Patients with Atherosclerotic Vascular Disease. N. Engl. J. Med. 2017, 377(13), 1217-1227.
110. Nicholls, S.J.; Ditmarsch, M.; Kastelein, J.J.; Rigby, S.P.; Kling, D.; Curcio, D.L.; Alp, N.J.; Davidson, M.H. Lipid lowering effects of the CETP inhibitor obicetrapib in combination with high-intensity statins: a randomized phase 2 trial. Nat. Med. 2022, 28(8), 1672-1678.
111. Ballantyne, C.M.; Ditmarsch, M.; Kastelein, J.J.; Nelson, A.J.; Kling, D.; Hsieh, A.; Curcio, D.L.; Maki, K.C.; Davidson, M.H.; Nicholls, S.J. Obicetrapib plus ezetimibe as an adjunct to high-intensity statin therapy: A randomized phase 2 trial. J. Clin. Lipidol. 2023, 17(4), 491-503.
112. Grossman, M.; Rader, D.J.; Muller, D.W.; Kolansky, D.M.; Kozarsky, K.; Clark, B.J.3rd.; Stein, E.A.; Lupien, P.J.; Brewer, H.B.Jr.; Raper, S.E.; et al. A pilot study of ex vivo gene therapy for homozygous familial hypercholesterolaemia. Nat. Med. 1995, 1(11), 1148-1154.
113. Jiang, L.; Wang, L.Y.; Cheng, X.S. Novel Approaches for the Treatment of Familial Hypercholesterolemia: Current Status and Future Challenges. J. Atheroscler. Thromb. 2018, 25(8), 665-673.
114. Martínez, T.; Jiménez, A.I.; Pañeda, C. Short-interference RNAs: becoming medicines. EXCLI J. 2015, 14, 714-746.
115. Nissen, S.E.; Linnebjerg, H.; Shen, X.; Wolski, K.; Ma, X.; Lim, S.; Michael, L.F.; Ruotolo, G.; Gribble, G.; Navar, A.M.; Nicholls, S.J. Lepodisiran, an Extended-Duration Short Interfering RNA Targeting Lipoprotein(a): A Randomized Dose-Ascending Clinical Trial. J.A.M.A. 2023, doi: 10.1001/jama.2023.21835
116. Schwartz, G.G.; Steg P.G.; Szarek, M.; Bhatt, D.L.; Bittner, V.A.; Diaz, R.; Edelberg, J.M.; Goodman, S.G.; Hanotin, C.; Harrington R.A.; et al. Alirocumab and Cardiovascular Outcomes after Acute Coronary Syndrome. N. Eng. J. Med. 2018, 379, 2097-2107.
117. Sabatine, M.S.; Giugliano, R.P.; Keech, A.C.; Honarpour, N.; Wiviott, S.D.; Murphy, S.A.; Kuder, J.F.; Wang, H.; Liu, T.; Wasserman, S.M.; et al. Evolocumab and Clinical Outcomes in Patients with Cardiovascular Disease. N. Eng. J. Med. 2017, 376, 1713-1722.
118. Khan, S.A.; Naz, A; Masood, M.Q.; Shah, R. Meta-Analysis of Inclisiran for the Treatment of Hypercholesterolemia. Am. J. Cardiol. 2020, 00, 1-5.
119. Kosmas, C.E.; Estrella, A.M.; Sourlas, A.; Silverio, D.; Hilario, E.; Montan, P.D.; Guzman, E. Inclisiran: A New Promising Agent in the Management of Hypercholesterolemia. Diseases 2018, 6(3), 63.
120. Fitzgerald, K.; Frank-Kamenetsky, M.; Shulga-Morskaya, S.; Liebow, A.; Bettencourt, B.R.; Sutherland, J.E.; Hutabarat, R.M.; Clausen, V.A.; Karsten, V.; Cehelsky, J.; et al. Effect of an RNA interference drug on the synthesis of proprotein convertase subtilisin/kexin type9 (PCSK9) and the concentration of serum LDL cholesterol in healthy volunteers: A randomised, single-blind, placebo-controlled, phase 1 trial. Lancet 2014, 383, 60-68.
121. Ray, K.K.; Landmesser, U.; Leiter, L.A.; Kallend, D.; Dufour, R.; Karakas, M.; Hall, T.; Troquay, R.P.T.; Turner, T.; Visseren, F.L.J.; et al. Inclisiran in Patients at High Cardiovascular Risk with Elevated LDL Cholesterol. N. Engl. J. Med. 2017, 376, 1430-1440.
122. Ray, K.K.; Troquay, R.P.T.; Visseren, F.L.J.; Leiter, L.A.; Wright, R.S.; Vikarunnessa, S.; Talloczy, Z.; Zang, X.; Maheux, P.; Lesogor, A.; et al. Long-term efficacy and safety of inclisiran in patients with high cardiovascular risk and elevated LDL cholesterol (ORION-3): results from the 4-year open-label extension of the ORION-1 trial. Lancet Diabetes Endoncrinol. 2023, 11, 109-119.
123. Raal, F.; Durst, R.; Bi, R.; Talloczy, Z.; Maheux, P.; Lesogor, A.; Kastelein, J.J.P.; on behalf of the ORION-5 Study Investigators. Efficacy, Safety, and Tolerability of Inclisiran in Patients With Homozygous Familial Hypercholesterolemia: Results From the ORION-5 Randomized Clinical Trial. Circulation 2023, doi: 10.1161/CIRCULATIONAHA.122.063460.
124. Raal, F.J.; Kallend, D.; Ray, K.K.; Turner, T.; Koenig, W.; Wright, R.S.; Wijngaard, P.L.J.; Curcio, D.; Jaros, M.J.; Leiter, L.A.; et al. Inclisiran for the Treatment of Heterozygous Familial Hypercholesterolemia. N. Eng. J. Med. 2020, 382, 1520-1530.
125. Ray, K.K.; Phil, M.; Wright, R.S.; Kallend, D.; Koenig, W.; Leiter, L.A.; Raal, F.J.; Bisch, J.A.; Richardson, T.; Jaros, M.; et al. Two Phase 3 Trials of Inclisiran in Patients with Elevated LDL Cholesterol. N. Eng. J. Med. 2020, 382, 1507-1519.
126. Koenig, W.; Conde, L.G.; Landmesser, U.; Leiter, L.A.; Ray, K.K.; Schwartz, G.G.; Wright, R.S.; Han, J.; Raal, F.J. Efficacy and Safety of Inclisiran in Patients with Polyvascular Disease: Pooled, Post Hoc Analysis of the ORION-9, ORION-10, and ORION-11 Phase 3 Randomized Controlled Trials. Cardiovasc. Drugs Ther. 2022, epub ahead of print.
127. Ray, K.K.; Raal, F.J.; Kallend, D.G.; Jaros, M.J.; Koenig, W.; Leiter, L.A.; Landmesser, U.; Schwartz, G.G.; Lawrence, D.; Friedman, A.; et al. Inclisiran and cardiovascular events: a patient-level analysis of phase III trials. Eur. Heart J. 2023, 44(2), 129-138.

128. Stoekenbroek, R.M.; Kallend, D.; Wijngaard, P.L.; Kastelein, J.J. Inclisiran for the treatment of cardiovascular disease: the ORION clinical development program. Future Cardiol. 2018, 14(6), 433-442.
129. Kersten, S. ANGPTL3 as therapeutic target. Curr. Opin. Lipidol. 2021, 32, 335-341.
130. Chen, Y.Q.; Pottanat, T.G.; Siegel, R.W.; Ehsani, M.; Qian, Y.; Zhen, E.Y.; Regmi, A.; Roell, W.C.; Guo, H.; Luo, M.L.; et al. Angiopoietin-like protein 8 differentially regulates ANGPTL3 and ANGPTL4 during postprandial partitioning of fatty acids. J. Lipid. Res. 2020, 61, 1203-1220.
131. Oldoni, F.; Cheng, H.; Banfi, S.; Gusarova, V.; Cohen, J.C.; Hobbs, H.H. ANGPTL8 has both endocrine and autocrine effects on substrate utilization. J.C.I. insight 2020, 5, e138777.
132. Kovrov, O.; Kristensen, K.K.; Larsson, E.; Ploug, M.; Olivecrona, G. On the mechanism of angiopoietin-like protein 8 for control of lipoprotein lipase activity. J. Lipid. Res. 2019, 60, 783-793.
133. Gusarova, V.; Alexa, C.A.; Wang, Y.; Rafique, A.; Kim, J.H.; Buckler, D.; Mintah, I.J.; Shihanian, L.M.; Cohen, J.C.; Hobbs, H.H.; et al. ANGPTL3 blockade with a human monoclonal antibody reduces plasma lipids in dyslipidemic mice and monkeys. J. Lipid. Res. 2015, 56, 1308-1317.
134. Dewey, F.E.; Gusarova, V.; Dunbar, R.L.; O'Dushlaine, C.; Schurmann, C.; Gottesman, O.; McCarthy, S.; Hout, C.V.V.; Bruse, S.; Dansky, H.M.; et al. Genetic and Pharmacologic Inactivation of ANGPTL3 and Cardiovascular Disease. N. Engl. J. Med. 2017, 377, 211-221.
135. Raal, F.J.; Rosenson, R.S.; Reeskamp, L.F.; Hovingh, G.K.; Kastelein, J.J.P.; Rubba, P.; Ali, S.; Banerjee, P.; Chan, K.C.; Gipe, D.A.; et al.; ELIPSE HoFH Investigators. Evinacumab for Homozygous Familial Hypercholesterolemia. N. Eng. J. Med. 2020, 383, 711-720.
136. Rosenson, R.S.; Burgess, L.J.; Ebenbichler, C.F.; Baum, S.J.; Stroes, E.S.G.; Ali, S.; Khilla, N.; Hamlin, R.; Pordy, R.; Dong, Y.; et al. Evinacumab in Patients with Refractory Hypercholesterolemia. N. Eng. J. Med. 2020, 383, 2307-2319.
137. Jinek, M.; Chylinski, k.; Fonfara, I.; Hauer, M.; Doudna, J. A.; Charpentier, E. A programmable dual-RNA-guided DNA endonuclease in adaptive bacterial immunity. Science 2012, 337(6096), 816-821.
138. El-Mounadi, K.; Morales-Floriano, M.L.; Garcia-Ruiz, H. Principles, Applications, and Biosafety of Plant Genome Editing Using CRISPR-Cas9. Front. Plant. Sci. 2020, 11, 56.
139. Sun, J.; Wang, J.; Zheng, D.; Hu, X. Advances in therapeutic application of CRISPR-Cas9. Brief. Funct. Genomics 2020, 19(3), 164-174.
140. Liang, P.; Xu, Y.; Zhang, X.; Ding, C.; Huang, R.; Zhang, Z.; Lv, J.; Xie, X.; Chen, Y.; Li, Y.; et al. CRISPR/Cas9-mediated gene editing in human tripronuclear zygotes. Protein Cell 2015, 6, 363-372.
141. Zhao, H.; Li, Y.; He, L.; Pu, W.; Yu, W.; Li, Y.; Wu, Y-T.; Xu, C.; Wei, Y.; Ding, Q.; et al. In Vivo AAV-CRISPR/Cas9-Mediated Gene Editing Ameliorates Atherosclerosis in Familial Hypercholesterolemia. Circulation 2020, 141, 67-79.
142. Wang, L.; Muthuramu, I.; Somanathan, S.; Zhang, H.; Bell, P.; He, Z.; Yu, H.; Zhu, Y.; Tretiakova, A.P.; Wilson, J.M. Developing a second-generation clinical candidate AAV vector for gene therapy of familial hypercholesterolemia. Mol. Ther. Methods Clin. Dev. 2021, 22, 1-10.
143. Duell, P.; Santos, R.; Kirwan, B.; Witztum, J.; Tsimikas, S.; Kastelein, J. Long-term mipomersen treatment is associated with a reduction in cardiovascular events in patients with familial hypercholesterolemia. J. Clin. Lipidol. 2016, 10(4), 1011-1021.
144. Astaneh, B.; Makhdami, N.; Astaneh, V.; Guyatt, G. The Effect of Mipomersen in the Management of Patients with Familial Hypercholesterolemia: A Systematic Review and Meta-Analysis of Clinical Trials. J. Cardiovasc. Dev. Dis. 2021, 8(7), 82.
145. Aguilar-Salinas, C.A.; Gómez-Díaz, R.A.; Corral, P. New Therapies for Primary Hyperlipidemia. J. Clin. Endocrinol. Metab. 2022, 107(5), 1216-1224.
146. Blom, D.J.; O'Dea, L.; Digenio, A.; Alexander, V.J.; Karwatowska-Prokopczuk, E.; Williams, K.R.; Hemphill, L.; Muñiz-Grijalvo, O.; Santos, R.D.; Baum, S.; et al. Characterizing familial chylomicronemia syndrome: Baseline data of the APPROACH study. J Clin Lipidol. 2018;12(5):1234-1243.e5.
147. Gouni-Berthold, I.; Alexander, V.J.; Yang, Q.; Hurh, E.; Steinhagen-Thiessen, E.; Moriarty, P.M.; Hughes, S.G.; Gaudet, D.; Hegele, R.A.; O'Dea, L.S.L.; et al.; COMPASS study group. Efficacy and safety of volanesorsen in patients with multifactorial chylomicronaemia (COMPASS): a multicentre, double-blind, randomised, placebo-controlled, phase 3 trial. Lancet Diabetes Endocrinol. 2021, 9(5), 264-275.
148. Oral, E.A.; Garg, A.; Tami, J.; Huang, E.A.; O'Dea, L.S.L.; Schmidt, H.; Tiulpakov, A.; Mertens, A.; Alexander, V.J.; Watts, L.; et al. Assessment of efficacy and safety of volanesorsen for treatment of metabolic complications in patients with familial partial lipodystrophy: Results of the BROADEN study: Volanesorsen in FPLD; The BROADEN Study. J. Clin. Lipidol. 202, 16(6), 833-849.
149. Tardif, J.C.; Karwatowska-Prokopczuk, E.; Amour, E.S.; Ballantyne, C.M.; Shapiro, M.D.; Moriarty, P.M.; Baum, S.J.; Hurh, E.; Bartlett, V.J.; Kingsbury, J.; et al. Apolipoprotein C-III reduction in subjects with moderate hypertriglyceridaemia and at high cardiovascular risk. Eur. Heart J. 2022, 43(14), 1401-1412.
150. Sniderman, A.D.; Navar, A.M.; Thanassoulis, G. Apolipoprotein B vs. low density lipoprotein cholesterol and non-high-density lipoprotein cholesterol as the primary measure of apolipoprotein B lipoprotein-related risk: the debate is over. J.A.M.A. Cardiol. 2022, 7, 257-258.

 doi:10.20944/preprints202312.1195.v1

151. Richardson, T.G.; Sanderson, E.; Palmer, T.M.; Ala-Korpela, M.; Ference, B.A.; Davey, S.G.; Holmes, M.V.; Rader, D.J. Evaluating the relationship between circulating lipoprotein lipids and apolipoproteins with risk of coronary heart disease: a multivariable Mendelian randomisation analysis, PLoS Med. 2020, 17, e1003062.
152. Zuber, V.; Gill. D.; Ala-Korpela, M.; Langenberg, C.; Butterworth, A.; Bottolo, L.; Burgess, S. High-throughput multivariable Mendelian randomization analysis prioritizes apolipoprotein B as key lipid risk factor for coronary artery disease. Int. J. Epidemiol. 2021, 50, 893-901.
153. Yuan, S.; Tang, B.; Zheng, J.; Larsson, S.C. Circulating lipoprotein lipids, apolipoproteins and ischemic stroke. Ann. Neurol. 2020, 88, 1229-1236.
154. Levin, M.G.; Zuber, V.; Walker, V.M.; Klarin, D.; Lynch, J.; Malik, R.; Aday, A.W.; Bottolo, L.; Pradhan, A.D.; Dichgans, M.; et al. Prioritizing the role of major lipoproteins and subfractions as risk factors for peripheral artery disease. Circulation 2021, 144, 353-364.
155. Richardson, T.G.; Wang, Q.; Sanderson, E.; Mahajan, A.; McCarthy, M.I.; Frayling, T.M.; Ala-Korpela, M.; Sniderman, A.; Smith, G.D.; Holmes, M.V. Effects of apolipoprotein B on lifespan and risks of major diseases including type 2 diabetes: a mendelian randomisation analysis using outcomes in first-degree relatives. Lancet Healthy Longev. 2021, 2, e317-e326.
156. Johannesen, C.D.; Mortensen, M.B.; Langsted, A.; Nordestgaard, B. Apolipoprotein B and Non-HDL-C better reflect residual risk than LDL cholesterol in statin-treated patients with atherosclerosis. J. Am. Coll. Cardiol. 2021, 77, 1439-1450.
157. Marston, N.A.; Giugliano, R.P.; Melloni, G.; Park, J.G.; Morrill, V.; Blazing, M.A.; Ference, G.A.; Stein, E.; Stroes, E.; Braunwald, E.; et al. Association of apolipoprotein-B-containing lipoproteins and risk of myocardial infarction in individuals with and without atherosclerosis: distinguishing between particle concentration, Type and Content. J.A.M.A. Cardiol. 2022, 7, 250-256.
158. Mach, F.; Baigent, C.; Catapano, A.L.; Koskinas, K.C.; Casula, M.; Badimon, L.; Chapman, M.J.; De Backer, G.G.; Delgado, V.; Ference, B.A.; et al. 2019 ESC/EAS guidelines for the management of dyslipidaemias: lipid modification to reduce cardiovascular risk. Eur. Heart J. 2020, 41, 111-188.
159. Kastelein, J.J.P.; Wedel, M.K.; Baker, B.F.; Su, J.; Bradley, J.D.; Yu, R.Z.; Chuang, E. Graham, M.J.; Crooke, R.M. Potent Reduction of Apolipoprotein B and Low-Density Lipoprotein Cholesterol by Short-Term Administration of an Antisense Inhibitor of Apolipoprotein B. Circulation 2006, 114(16), 1729-1735.
160. Li, N.; Li, Q.; Tian, X.Q.; Qian, H.Y.; Yang, Y.J. Mipomersen is a Promising Therapy in the Management of Hypercholesterolemia: A Meta-Analysis of Randomized Controlled Trials. Am. J. Cardiovasc. Drugs 2014, 14(5), 367-376.
161. Fogacci, F.; Ferri, N.; Toth, P.P.; Ruscica, M.; Corsini, A.; Cicero, A.F.G. Efficacy and Safety of Mipomersen: A Systematic Review and Meta-Analysis of Randomized Clinical Trials. Drugs 2019, 79(7), 751-766.
162. Liang, C.; Wang, X.; Peng, K.; Lai, P.; Liu, Z.; Ma, J.; Chen, X.; Liu, G.; Zheng, M., Wang, Y.; et al. Idol Depletion Protects against Spontaneous Atherosclerosis in a Hamster Model of Familial Hypercholesterolemia. Oxid. Med. Cell. Longev. 2022, 2022, 1889632.
163. Janowski, B.A.; Grogan, M.J.; Jones, S.A.; Wisely, G.B.; Kliewer, S.A.; Corey, E.J.; Mangelsdorf, D.J. Structural requirements of ligands for the oxysterol liver X receptors LXRalpha and LXRbeta. Proc. Natl. Acad. Sci. U.S.A. 1999, 96(1), 266-271.
164. Boyadjian, R.; Tontonoz, P. Lxr regulates cholesterol uptake through idol-dependent ubiquitination of the ldl receptor. Science 2009, 325, 100-104.
165. Zelcer, N.; Hong, C.; Boyadjian, R.; Tontonoz, P. LXR Regulates Cholesterol Uptake through Idol-dependent Ubiquitination of the LDL Receptor. Science 2009, 325(5936), 100-104.
166. Waterworth, D.M.; Ricketts, S.L.; Song, K.; Chen, L.; Zhao, J.H.; Ripatti, S.; Aulchenko, Y.S.; Zhang, W.; Yuan, X.; Lim, N.; et al. Genetic variants influencing circulating lipid levels and risk of coronary artery disease. Arterioscler. Thromb. Vasc. Biol. 2010, 30(11), 2264-2276.
167. Blom, D.J.; Raal, F.J.; Santos, R.D.; Marais, A.D. Lomitapide and Mipomersen-Inhibiting Microsomal Triglyceride Transfer Protein (MTP) and apoB100 Synthesis. Curr. Atheroscler. Rep. 2019, 21(12), 48.
168. Hooper, A.J.; Burnett, J.R.; Watts, G.F. Contemporary aspects of the biology and therapeutic regulation of the microsomal triglyceride transfer protein. Circ. Res. 2015, 116, 193-205.
169. Cuchel, M.; Bloedon, L.T.; Szapary, P.O.; Kolansky, D.M.; Wolfe, M.L.; Sarkis, A.; Millar, J.S.; Ikewaki, K.; Siegelman, E.S.; Gregg, R.E. Inhibition of microsomal triglyceride transfer protein in familial hypercholesterolemia. N. Engl. J. Med. 2007, 356(2), 148-156.
170. Cuchel, M.; Meagher, E.A.; Theron, H.T.; Blom, D.J.; Marais, A.D.; Hegele, R.A.; Averna, M.R.; Sirtori, C.R.; Shah, P.K.; Gaudet, D.; et al. Efficacy and safety of a microsomal triglyceride transfer protein inhibitor in patients with homozygous familial hypercholesterolaemia: a single-arm, open-label, phase 3 study. Lancet 2013, 381(9860), 40-46.
171. Adi, D.; Abuzhalihan, J.; Wang, Y.H.; Baituola, G.; Wu, Y.; Xie, X.; Fu, Z.Y.; Yang, Y.N.; Ma, X.; Li, X.M.; Chen, B.D.; et al. IDOL gene variant is associated with hyperlipidemia in Han population in Xinjiang, China. Sci. Rep. 2020, 10(1), 14280.

 doi:10.20944/preprints202312.1195.v1

172. Landlinger, C.; Pouwer, M.G.; Juno, C.; van der Hoorn, J.W.A.; Pieterman, E.J.; Jukema, J.W.; Staffler, G.; Princen, H.M.G.; Galabova, G. The AT04A vaccine against proprotein convertase subtilisin/kexin type 9 reduces total cholesterol, vascular inflammation, and atherosclerosis in APOE*3Leiden.CETP mice. Eur. Heart J. 2017, 38(32), 2499-2507.
173. Momtazi-Borojeni, A.A.; Jaafari, M.R.; Badiee, A.; Banach, M.; Sahebkar, A. Therapeutic effect of nanoliposomal PCSK9 vaccine in a mouse model of atherosclerosis. B.M.C. Med. 2019, 17(1), 1-5.
174. Crossey, E.; Amar, M.J.A.; Sampson, M.; Peabody, J.; Schiller, J.T.; Chackerian, B.; Remaley, A.T. A cholesterol-lowering VLP vaccine that targets PCSK9. Vaccine 2015, 33(43), 5747-5755.
175. Kawakami, R.; Nozato, Y.; Nakagami, H.; Ikeda, Y.; Shimamura, M.; Yoshida, S.; Sun, J.; Kawano, T.; Takami, Y.; Noma, T.; et al. Development of vaccine for dyslipidemia targeted to a proprotein convertase subtilisin/kexin type 9 (PCSK9) epitope in mice. PLoS One 2018, 13(2), e0191895.
176. Fattori, E.; Cappelletti, M.; Lo Surdo, P.; Calzetta, A.; Bendtsen, C.; Ni, Y.G.; Pandit, S.; Sitlani, A.; Mesiti, G.; Carfí, A.; et al. Immunization against proprotein convertase subtilisin-like/kexin type 9 lowers plasma LDL-cholesterol levels in mice. J. Lipid Res. 2012, 53(8), 1654-1661.
177. Pan, Y.; Zhou, Y.; Wu, H.; Chen, X.; Hu, X.; Zhang, H.; Zhou, Z.; Qiu, Z.; Liao, Y. A therapeutic peptide vaccine against PCSK9. Sci. Rep. 2017, 7(1), 1-10.
178. Momtazi-Borojeni, A.A.; Jaafari, M.R.; Afshar, M.; Banach, M.; Sahebkar, A. PCSK9 immunization using nanoliposomes: preventive efficacy against hypercholesterolemia and atherosclerosis. Arch. Med. Sci. 2021, 17(5), 1365-1377.
179. Fowler, A.; Van Rompay, K.K.A.; Sampson, M.; Leo, J.; Watanabe, J.K.; Usachenko, J.L.; Immareddy, R.; Lovato, D.M.; Schiller, J.T.; Remaley, A.T.; et al. A Virus-like particle-based bivalent PCSK9 vaccine lowers LDL-cholesterol levels in Non-Human Primates. npj Vaccines 2023, 8, 142.
180. King, M.E.; Breslow, J.L.; Lees, R.S. Plasma-exchange therapy of homozygous familial hypercholesterolemia. N. Engl. J. Med. 1980, 302, 1457-1459.
181. Taylan, C.; Weber, L.T. An update on lipid apheresis for familial hypercholesterolemia. Pediatr. Nephrol. 2023, 38(2), 371-382.
182. Albayrak, M.; Yıldız, A.; Ateş, N.; Pala, Ç. The efficacy of double filtration plasmapheresis in the treatment of homozygous familial hypercholesterolemia: A single-center experience. Transfus. Apher. Sci. 2019, 58(1), 61-64.
183. Cao, L.; Chen, Y.; Liu, S.; Huang, W.; Wu, D.; Hong, D.; Wang, Z.; Sun, Y.; Qin, K.; Guo, F.; et al. Chinese Acute Pancreatitis Clinical Trials Group (CAPCTG). Early Plasmapheresis Among Patients With Hypertriglyceridemia-Associated Acute Pancreatitis. J.A.M.A. Netw. Open 2023, 6(6), e2320802.
184. Song, X.; Shi, D.; Cui, Q.; Yu, S.; Yang, J.; Song, P.; Walline, J.; Xu, J.; Zhu, H.; Yu, X. Intensive insulin therapy versus plasmapheresis in the management of hypertriglyceridemia-induced acute pancreatitis (Bi-TPAI trial): study protocol for a randomized controlled trial. Trials 2019, 20(1), 365.