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LIPID ABNORMALITIES AND CARDIOVASCULAR PREVENTION (G DE BACKER, SECTION EDITOR)

Genetics, Dyslipidemia, and Cardiovascular Disease: New Insights

Ricardo Stein [1,2,3,4] · Filipe Ferrari [1,2] · Fernando Scolari [1]

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Abstract

Purpose of Review The cardiovascular (CV) risk related to lipid disorders is well established and is based on a robust body of evidence from well-designed randomized clinical trials, as well as prospective observational studies. In the last two decades, significant advances have been made in understanding the genetic basis of dyslipidemias. The present review is intended as a comprehensive discussion of current knowledge about the genetics and pathophysiology of disorders that predispose to dyslipidemia. We also focus on issues related to statins and the proprotein convertase subtilisin/kexin type 9 (PCSK9) and some of its polymorphisms, as well as new cholesterol-lowering medications, including PCSK9 inhibitors.

Recent Finding Cholesterol is essential for the proper functioning of several body systems. However, dyslipidemia—especially elevated low-density lipoprotein (LDL-c) and triglyceride levels, as well as reduced lipoprotein lipase activity—is associated with an increased risk of coronary artery disease (CAD). High-density lipoprotein (HDL-c), however, seems to play a role as a risk marker rather than as a causal factor of the disease, as suggested by Mendelian randomization studies. Several polymorphisms in the lipoprotein lipase locus have been described and are associated with variations in the activity of this enzyme, producing high concentrations of triglycerides and increased risk of CAD.

Summary Dyslipidemia, especially increased LDL-c and triglyceride levels, continues to play a significant role in CV risk. The combination of genetic testing and counseling is important in the management of patients with dyslipidemia of genetic etiology. Strategies focused on primary prevention can offer an opportunity to reduce CV events.

Keywords Dyslipidemia · Familial hypercholesterolemia · Genetic test · Cardiovascular disease

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✉ Ricardo Stein
rstein@cardiol.br

Filipe Ferrari
ferrari.filipe88@gmail.com

Fernando Scolari
fernando.scolari@gmail.com

[1] Graduate Program in Cardiology and Cardiovascular Sciences, Hospital de Clínicas de Porto Alegre (HCPA), Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil

[2] Exercise Cardiology Research Group (CardioEx), Hospital de Clínicas de Porto Alegre, Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil

[3] School of Medicine, Hospital de Clínicas de Porto Alegre, Universidade Federal do Rio Grande do Sul, Porto Alegre, Brazil

[4] Serviço de Fisiatria e Reabilitação, Hospital de Clínicas de Porto Alegre, Rua Ramiro Barcelos, 2350, Porto Alegre, RS 90035-903, Brazil

Introduction

Dyslipidemia, defined as an abnormally high concentration of lipids in the blood, is one of the main risk factors for the development and progression of cardiovascular (CV) disease [1]. Although the prevalence of total blood cholesterol above desirable indices has decreased in the last decade, it still affects approximately 12% of adults, especially those in the fifth and sixth decades of life [2]. Dyslipidemia is associated not only with lifestyle but also with genetic disorders [1–5]. Familial hypercholesterolemia (FH) is an autosomal dominant inherited disease that affects low-density lipoprotein (LDL-c) through LDL-c receptor (LDLR) gene mutations and is associated with higher CV risk [3]. Besides monogenic disorders such as FH, polymorphisms involving not only LDL-c genes but also high-density lipoprotein (HDL-c), lipoprotein lipase (LPL), and apolipoproteins are also related to higher blood cholesterol levels, increased CV risk, and, therefore, worse outcomes [5–12].

Statins are the cornerstone of dyslipidemia treatment, but their clinical effect and adverse effect profile may differ depending on the genetic burden of the patient [13–18]. Genetic profiles that are associated with high blood cholesterol and higher CV risk can modify the clinical approach on the individual level [19, 20]. The discovery of a mutation on the proprotein convertase subtilisin/kexin type 9 (PCSK9) gene which is associated with LDL-c levels and CV risk has led to the development of a new class of agents to treat severe dyslipidemia [21, 22••].

In this review, we discuss the evidence for a causal role of cholesterol in the progression of CV disease, its relationship to genes and polymorphisms that change the lipid profile, and implications for treatment. Finally, we examine how precision medicine, specifically genetic testing and next-generation sequencing (NGS), is advancing knowledge on dyslipidemia and CV disease.

Familial Hypercholesterolemia: the Prototypical Form of Genetic Dyslipidemia

FH is the prototype of a monogenic disorder leading to dyslipidemia. This disease affects 1 in 200–250 individuals in general, and the vast majority of these individuals are heterozygotes, making it the most common monogenic disease [23]. Three major pathogenic defects are implicated: mutations in the LDLR gene, in apolipoprotein B (APOB), or in PCSK9 [24]. Homozygous FH is rarer, caused by pathogenic biallelic variants, usually in LDLR, with recent data suggesting a prevalence of approximately 1 in 160,000 to 450,000 individuals. The main characteristics of FH in its heterozygous and homozygous forms, as well as its implications for CV risk, are summarized in Table 1 [25–33]. If left undiagnosed, FH is associated with increased risk of fatal or nonfatal coronary events, reaching 50% at age 50 (in untreated women, this risk reaches 30% at 60 years) [34].

In individuals with LDL-c levels ≥ 190 mg/dL, the risk of coronary artery disease (CAD) is increased sixfold, but when a genetic variant causing FH is associated, the risk increases 22-fold at the same LDL-c level [35]. Unfortunately, genetic screening is not used as widely for the diagnosis of FH as desired, which can play a decisive role in risk stratification [36]. In addition, genetic screening allows assessment of the patient's relatives by cascade testing, making it clear whether more aggressive lipid-lowering treatment should be prescribed [37]. Some diagnostic criteria for heterozygous FH, according to the Dutch Lipid Clinic Network, are summarized in Table 2 [38, 39].

The Expert Consensus Panel recommends that genetic testing become standard in the care of patients with probable or definite FH, as well as for their relatives at risk (Fig. 1) [34]. As the costs of NGS have decreased, genetic testing in this scenario is becoming increasingly affordable, leading to a major advance in clinical diagnosis and public health surveillance [40].

Genes and Polymorphisms Related to Cholesterol Levels

LDL-c

The role of LDL-c in inflammation and pathogenesis of atherosclerosis has been evaluated extensively since the beginning of the twenty-first century. Consistent evidence from multiple clinical and genetic studies has established that LDL-c is a cornerstone in the genesis of CV disease. A meta-analysis enrolling 892,337 individuals without CV disease described a strong association between total cholesterol, LDL-c, and CAD-associated mortality [41]. The discovery of the LDLR provided more robust subsidies for this association [42].

Prospective epidemiological studies of Mendelian randomization have shown a consistent association between the absolute magnitude of LDL-c and CV risk. The longer an individual is exposed to high cholesterol levels, the greater the risk [43]. Silverman et al. [44•] evaluated more than 310,000 individuals (mean age 62 years, mean LDL-c 120 mg/dL) included in 49 studies. There were 39,645 major vascular events. In this meta-regression analysis, the authors reported that lower LDL-c levels were associated with lower rates of major CV events. Corroborating these findings, a recent meta-analysis and meta-regression of 34 studies ($n = 136{,}000$ patients) showed that a significant reduction in LDL-c was associated with a decrease in CV and all-cause mortality. However, these findings were more consistent in studies in which patients had baseline LDL-c > 100 mg/dL [45], suggesting that the greatest benefit of LDL-c reduction therapy may occur in patients with elevated baseline LDL-c levels.

LDLR mutations are the main cause of FH. Khera et al. [35] sequenced three genes causative of FH (LDLR, APOB, and PCSK9) in more than 26,000 participants from seven case-control studies and five prospective cohort studies. In their analysis, among participants with LDL-c ≥ 190 mg/dL, gene sequencing identified an FH mutation in < 2%. However, for any observed LDL-c level, FH mutation carriers had substantially increased risk of CAD. It is interesting to point out that 86% of patients with FH had their mutations located in LDLR, most of them missense, although the highest LDL-c levels were related to loss-of-function mutations.

One study reported that common variants in 95 loci were associated with higher lipid levels. Individuals with an LDL-c allelic dosage score in the top quartile were 13 times as likely to have elevated LDL-c as individuals in the bottom quartile

Table 1 Main characteristics of FH in its heterozygous and homozygous forms

Disease	Mutation	Lipid phenotype	Other manifestations	Frequency	If untreated
FH [26, 27]	LDLR*, APOB, PCSK9, APOE, SREBP2, STAP1	Increased LDL-c	Xanthomas	1/250	Increase CV risk
Heterozygous FH [25, 28–30]	LDLR	LDL-c in the range of 155 to 500 mg/dL	Tendon/skin xanthomas	1/500	Develop CAD before the age of 55 years (men) and 60 years (women)
Homozygous FH [31–33]	LDLR, APOB, PCSK9, LDLRAP1	LDL-c can reach > 600 mg/dL	Planar and tendinous xanthomas, valvar and supravalvar atheroma	Rare (1/275,000–450,-000)	Rarely survive beyond the age of 30 years

FH familial hypercholesterolemia, *LDL-c* low-density lipoprotein, *LDLR* LDL-c receptor, *LDLRAP1* LDL-c receptor adaptor protein 1, *STAP1* signal transducing adaptor family member 1, *SREBP2* sterol regulatory element-binding protein 2, *APOB* apolipoprotein B, *PCSK9* proprotein convertase subtilisin/kexin type 9, *APOE* apolipoprotein E

*Occurs in 79% of cases

[46]. In another study, Talmud et al. [47] compared 321 mutation-negative subjects, 319 mutation-positive subjects, and 3020 controls from the UK Whitehall II cohort. Participants were genotyped for 12 common LDL-c-raising alleles, and a weighted LDL-c-raising gene score was constructed. In the top decile (LDL-c 189 mg/dL), the CV risk ratio was 4.17 (3.10–5.78) in comparison with that in the bottom decile (LDL-c 145 mg/dL). This finding has implications for familial cascade testing, which might reveal not a single mutation but rather a polygenic cause.

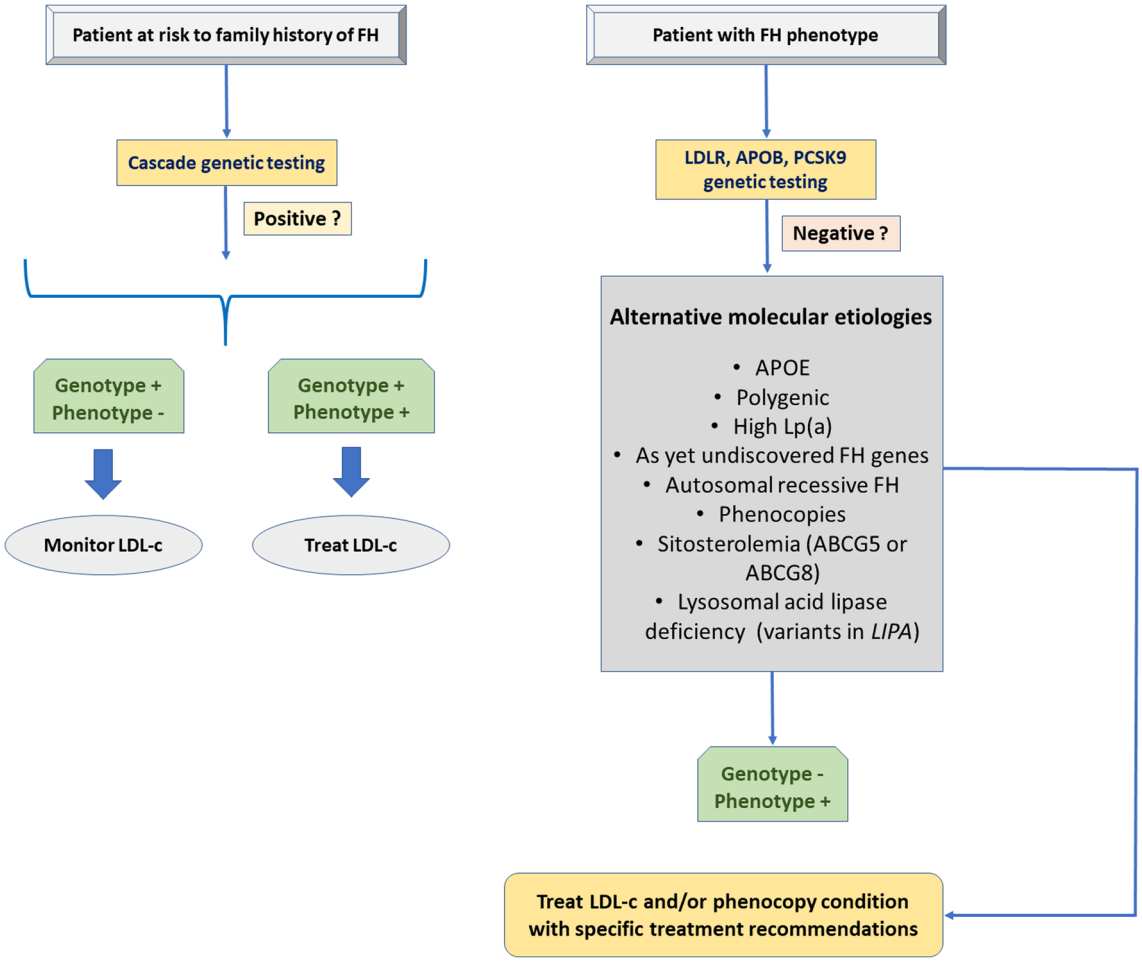
HDL-c

Over many years, epidemiological studies strongly suggested that HDL-c levels were inversely associated with the risk of CAD [48]. However, although this dogma shaped the panorama of dyslipidemia for several generations of physicians, the causal nature of this relationship has been questioned by studies of Mendelian randomization. In addition, genetic mechanisms that appear to increase HDL-c have no significant implications for the reduction of myocardial infarction rates,

Table 2 Score for diagnosis of FH [38, 39]

	Points
Family history	
First-degree relative known with premature coronary artery disease percentile (men < 55 years, women < 60 years)	1
First-degree relative with LDL-c > 95th First-degree relative with tendinous xanthomata and/or children aged < 18 years with LDL-c > 95th percentile	2
Clinical history	
Premature coronary artery disease (men < 55 years, women < 60 years)	2
Premature cerebral/peripheral vascular disease (men < 55 years, women < 60 years)	1
Physical examination	
Tendinous xanthomata	6
Arcus cornealis prior to age 45 years	4
LDL-c levels (mg/dL)	
> 330	8
~ 250–329	5
~ 190–249	3
~ 155–189	1
DNA analysis	
Causative mutation in the LDLR	8

< 3 points: no diagnosis; 3–5 points: possible FH; 6–8 points: probable FH; > 8 points: definite FH (reprinted with permission from the WHO Human Genetics Programme [38])



FH: Familial hypercholesterolemia; LDL-c: Low-density lipoprotein; LDLR: LDL-c receptor; PCSK9: Proprotein convertase subtilisin/kexin type 9; APOE: Apolipoprotein E; Lp(a): Lipoprotein (a).

Fig. 1 Patients with FH eligible for genetic testing (republished with permission of Elsevier Science and Technology Journals, from Sturm et al. [34]; permission conveyed through Copyright Clearance Center, Inc.). FH, familial hypercholesterolemia; LDL-c, low-density lipoprotein; LDLR, LDL-c receptor; PCSK9, proprotein convertase subtilisin/kexin type 9; APOE, apolipoprotein E; Lp(a), lipoprotein (a)

which may explain why clinical trials aimed at increasing HDL-c have failed [49–51].

Apolipoprotein A1 (APOA1) is a protein that is encoded by the APOA1 gene and plays a specific role in lipid metabolism, as the main protein component of plasma HDL-c. Interestingly, genetically elevated APOA1 did not reduce the risk of CAD [52]. In the same direction, three functional hepatic lipase variants associated with a (modest) increase in HDL-c also did not reduce CV risk [53]. Increases in HDL-c due to mutations or polymorphisms in genes that regulate HDL-c remodeling (such as cholesteryl ester transfer protein (CETP)) or clearance (scavenger receptor class B type 1 (SR-BI)) have not been clearly linked to vascular protection [54]. Similarly, trials of the plasma CETP inhibitor torcetrapib found that, although a significant increase (60% or more) in HDL-c was achieved, an increase in atherosclerosis burden was also observed [6, 7].

On the other hand, low HDL-c levels are an important risk factor for atherosclerosis [48]. Rare mutations of APOA1, ATP-binding cassette protein A1 (ABCA1), and lecithin: cholesterol acyltransferase (LCAT) can contribute to low plasma levels of HDL-c [4, 5, 55]. These mutations are present in at least 40% of probands, with reductions in HDL-c levels ranging from 0.44 mmol/L (~17 mg/dL) to 0.69 mmol/L (~27 mg/dL). Interestingly, they were associated with CAD risk only when HDL-c levels were below the 5th percentile (0.9 mmol/L, ~35 mg/dL) [4]. In another study, ABCA1 and APOA1 mutations were evaluated by NGS in 72 patients with HDL-c below the 10th percentile. Of these individuals, 22% had a probable or known pathogenic variant and 83% had

evidence of atherosclerosis when HDL-c was < 23 mg/dL, compared with those without mutations, of whom only 39% had evidence of atherosclerosis when HDL-c was lower than 27 mg/dL [56]. These data show that, even in patients with low HDL-c levels, the presence of such mutations increases the risk of atherosclerosis. This robust evidence strengthens the hypothesis that HDL-c levels can be regarded as a marker of CV risk, but not as a specific causal risk factor for CV disease [57].

Lipoprotein Lipase

The complete absence of LPL in individuals who are homozygous for the G188E LPL gene mutation causes complete loss of enzyme activity [58]. This leads to a phenotype known as hyperlipoproteinemia type 1 [59]. Several polymorphisms in the LPL locus are associated with variations in its activity, serum lipid concentrations, and risk of CAD [60]. The presence of two mutated alleles in the LPL gene locus causes a series of deficiencies in enzyme activity, producing marked fasting hypertriglyceridemia [61]. The H2H2 genotype has been associated with high fasting triglycerides and LDL-c and reduced levels of HDL-c, resulting in CAD [8]. In previous studies, three common LPL variants—Ser447Ter, PvuII, and HindIII—were associated with higher plasma lipid levels and CAD [62–64]. In addition, the HindIII polymorphism was assessed in a recent meta-analysis, and the genotype HindIII H+H+ and H+ allele were found to be associated with increased CV risk [9].

Apolipoprotein C3

There is a possible association between the rare allele G3238 and high levels of total cholesterol, triglycerides, apolipoprotein C3 (APOC3), and increased CV risk [65]. More recently, the APOC3 polymorphisms were found to be associated with stroke risk in Chinese women [66]. The R19X mutation in the APOC3 gene promoted a 40–50% reduction in APOC3 levels, in addition to reductions in fasting and postprandial triglycerides. In this population, a significant reduction in coronary artery calcification was observed [10]. Four mutations in the APOC3 gene were identified as being associated with plasma triglyceride levels in participants of the Exome Sequencing Project. These mutations resulted in a 46% decrease in circulating APOC3 levels, leading to a 39% decrease in triglyceride levels. According to the authors, APOC3 deficiency conferred a 40% reduction in CV disease risk [11].

Finally, a meta-analysis investigated the three main APOC3 polymorphisms (Ssti, T-455C, and C-482T) in all studies published up to 2016. The authors found that the Ssti and T-455C polymorphisms increased susceptibility to CAD significantly, but no association was observed with the C-482T polymorphism [12].

PCSK9

The PCSK9 gene encodes the proprotein convertase subtilisin/kexin type 9, an enzyme that reduces the presence of LDLR on the surface of hepatocytes, reducing LDLR availability [67]. This leads to alterations in the lipid profile and elevation of LDL-c levels. In fact, the discovery of the first PCSK9 gene mutation in 2003, a variant that causes autosomal dominant hypercholesterolemia, was the first step toward closing a scientific gap in the understanding of cholesterol metabolism [68]. A meta-analysis of 32 studies showed a consistent association between the G allele variant of PCSK9 rs505151 and higher serum LDL-c levels in a Caucasian population. In addition, this polymorphism was also related to an increased incidence of CV events. On the other hand, the T allele variant of PCSK9 rs11591147 was associated with reduced serum levels of total cholesterol and LDL-c, as well as reduced CV risk [69]. Other meta-analyses have shown an association of the PCSK9 rs505151 variant with increased serum levels of total cholesterol and LDL-c, as well as increased CV risk [70, 71]. Therefore, these variants can be used as genetic biomarkers for the assessment of CV risk and may become targets for diagnosis and more specific therapeutic intervention.

Dyslipidemia Treatment and Genetics

Statins

Statins, the most widely prescribed drugs for treatment of dyslipidemia, reduce intrahepatic cholesterol synthesis by inhibiting the enzyme 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase [72]. This leads to upregulation of LDLR on the hepatocyte surface, increasing LDL-c uptake and lowering serum levels of this lipoprotein [73]. Statins are associated with CV risk reduction both as primary and as secondary prevention [74, 75]. However, some patients do not achieve cholesterol reduction goals, and others cannot tolerate these drugs due to adverse effects. The use of pharmacogenetic information to individualize drug treatment in clinical practice can maximize efficacy and prevent adverse events and is an important component of precision medicine.

Pharmacogenomics of Statin Adverse Effects

Muscle symptoms are one of the most common statin-related side effects. They are directly responsible for nonadherence to lipid-lowering treatment in up to 15% of patients, who are especially likely to discontinue therapy if muscle pain develops [13]. Some genes that are involved in higher susceptibility to this association have been reported, such as AMPD1, COQ2, CPT2, and CYP2D6 [76], and there is evidence that

the presence of some polymorphisms may also be associated with intolerance to this drug class. The variant c.521C>T (Val174Ala; rs4149056) is the best known such polymorphism, reducing the activity of solute carrier organic anion transporter family member 1B1 (SLCO1B1) and increasing plasma levels of statins [14]. Different clinical expressions of statin intolerance are seen, depending on whether the carrier is heterozygous or homozygous (ratio of 1.0 and 17, respectively) [14, 15, 77].

A genotype-based approach for prescribing simvastatin was proposed by the US National Institutes of Health Pharmacogenomics Research Network and the Pharmacogenomics Knowledge Base (PharmGKB®), where simvastatin dose should be lowered in the intermediate (genotype TC) and high-risk (genotype CC) groups [15, 78]. Such genotype-informed statin therapy (GIST) may decrease LDL-c levels and improve adherence in primary care [65]. In a recent study, 159 patients with a history of statin-induced side effects were randomized to receive SLCO1B1-directed GIST or usual care [79]. The primary endpoint was statin adherence using the Morisky Medication Adherence Scale. SLCO1B1*5 was found in 25% of the participants. Although no difference was seen on adherence between the two arms, the GIST arm had more new statin prescriptions compared with the usual care group (55.4% versus 38.0%; $p = 0.04$), lower LDL-c levels at 3 months (131.9 versus 144.4 mg/dL; $p = 0.04$), and lower levels at 8 months (128.6 versus 141.0; $p = 0.12$) [55]. Thus, SLCO1B1 testing can help doctors restart statins and improve patient perception of therapy with this class of drug.

The ABCB1 gene, related to hepatobiliary and renal–urinary transport of statins, is also associated with statin-induced myopathy, but there are no recommendations based on the genotype due to inconsistent results [80, 81]. Statins are metabolized via cytochrome P450 (CYP) 3A isoenzymes, and concomitant use of CYP3A-inhibiting medications is associated with increased risk of myotoxicity. Polymorphisms that reduce the function of CYP3A are also associated with increased risk, such as CYP3A4*22 (rs35599367) and CYP3A5*3 (rs776746). Again, there are no formal recommendations on genotype-based therapy for these polymorphisms [14, 82, 83].

Decreased Statin Effect due to Genetic Polymorphisms

The clinical response to statins may vary according to the genetic burden of the patient [17, 18, 84]. This interindividual effect may be explained by polymorphisms related to statin absorption and metabolism [85]. At least 40 genes are thought to be related to different statin effects.

The gene that encodes the CETP, which facilitates triglyceride and cholesteryl ester exchange between HDL-c and apolipoprotein B100-containing lipoproteins, has been extensively studied [16]. Polymorphisms on this gene, such as the rs3764261 variant, are associated with higher HDL-c and total cholesterol and with lower LDL-c and triglycerides. On the other hand, some polymorphisms may reduce HDL-c and elevate LDL-c and triglyceride levels, such as the rs1800775 variant, which is associated with higher CV risk [17]. Rs708272, known as Taq1B, is a common polymorphism in intron 1 that relates to statin response. In a meta-analysis, the Taq1B genotype was significantly associated with HDL-c levels. B2B2 individuals had higher HDL-c levels than did B1B1 subjects, with an impact on CAD rates (odds ratio = 0.78 (0.66 to 0.93); $p = 0.008$) [18]. In contrast with these results, the REGRESS trial found that statin treatment reduced CV and CAD death in B1B1 individuals [86].

Single-nucleotide polymorphisms (SNP) in the gene that encodes apolipoprotein E (APOE) are associated with total cholesterol and LDL-c serum levels. APOE *ε3* homozygotes derive greater benefit from LDL-c reduction with statins than *ε4* homozygotes, and *ε2* homozygotes experience greater reductions than *ε3* homozygotes [87]. One study evaluated 43 SNPs in 16 genes implicated with statin response. Among them, only APOE2 (rs7412) heterozygotes had lower LDL-c levels compared with common allele carriers [85]. In another study, 23 candidate genes were analyzed in 5745 individuals. Three SNPs in APOE were associated with LDL-c reduction: rs4112, SNP17, and rs429358 [84]. A recent meta-analysis enrolling 1171 individuals assessed the lipid response to fluvastatin according to SLCO1B1, APOE, and CYP2C9 genotypes. SLCO1B1 521TT was associated with greater change in total cholesterol and LDL-c levels compared with 521TC or CC, and APOE ε2/ε3 was associated with greater HDL-c improvement in comparison with APOE *ε3/ε3*, *ε3/ε4*, or *ε4/ε4* [88].

PCSK9 Inhibitors

The discovery of PCSK9, a serine protease which binds to the LDLR and targets these receptors for lysosomal degradation, created an additional route through which plasma LDL-c levels can be controlled [89].

Two major studies (mean follow-up 2–3 years) tested monoclonal antibody PCSK9 inhibitors in patients with established CV disease already on statins: the FOURIER trial of evolocumab [19] and the ODYSSEY trial of alirocumab [20]. Both showed extreme reductions in LDL-c levels, on the order of 60% greater than statins alone. In the FOURIER study, more than 27,000 patients were evaluated, and those randomized to receive the PCSK9 inhibitor had a risk reduction of 15% (combined outcome of myocardial infarction, stroke, CV death, coronary revascularization, and hospitalization for unstable angina) [19]. In the ODYSSEY OUTCOMES study, which enrolled approximately 19,000

patients, those treated with the PCSK9 inhibitor also presented a 15% reduction in the primary combined outcome (CAD death, nonfatal myocardial infarction, ischemic stroke, or hospitalization for unstable angina) [20]. In the secondary analysis, the clinical benefit of evolocumab was also consistent in patients with low baseline LDL-c levels (< 70 mg/dL). In this subgroup of patients, LDL-c was reduced to 20 mg/dL, with a 30% reduction in the risk of CV death, myocardial infarction, or stroke in comparison with placebo [90].

It is important to point out that greater reductions in LDL-c entail a proportional reduction in CV risk [91]. Humphries et al. [92] evaluated a large cohort of patients with severe FH followed from 1992 to 2016. The authors observed that CAD mortality remained higher in patients with severe FH who were on statins when compared with those who did not have this diagnosis. Of note, the importance of a more "radical" reduction of LDL-c, as can be achieved with PCSK9 inhibitors, places these drugs as a very interesting option for the clinician. Indeed, these therapeutic agents offer the opportunity for earlier intervention in order to reduce LDL-c and decrease the impact of CAD on public health. However, it is important that the price of PCSK9 inhibitors become more affordable, since long-term follow-up is necessary [93].

Inclisiran

Small interfering RNA (siRNA) molecules have been used to target the hepatic production of PCSK9. These molecules interfere with the expression of specific genes by affecting the posttranscriptional degradation of mRNA, thus preventing translation [94, 95]. In a small phase I randomized clinical trial, subcutaneous doses of 300 mg or more of inclisiran, a PCSK9-targeted siRNA, significantly reduced LDL-c levels for at least 6 months in patients with a baseline LDL-c of at least 100 mg/dL. Furthermore, no evidence of major adverse events was observed [67]. In a multicenter, double-blind, placebo-controlled phase II trial of 500 patients, administration of inclisiran to patients with high CV risk led to significant reductions in LDL-c levels [96]. Through analysis of these two studies, one can assume that a single 300-mg dose of inclisiran administered every 6 months may result in a mean 50% reduction in plasma levels of LDL-c. Moreover, the benefit seems similar in the presence or absence of diabetes, which may make it an especially attractive treatment option for this subgroup [97]. Robust phase III clinical trials are already underway, with evaluation of hard clinical endpoints, encompassing 15,000 patients and a planned duration of 5 years [98]. Inclisiran is still an experimental agent and has not been approved by the US Food and Drug Administration or any other regulatory authorities [99].

Genetic Tests and Precision Medicine

High Genetic Risk

As noted above, the presence of certain gene polymorphisms not only increases CV risk but can also affect treatment with statins. To assess whether a genetic risk-based treatment approach would be clinically feasible, Natarajan et al. [21] proposed a polygenic risk score derived from up to 57 common DNA sequence variants associated with CAD. They compared the efficacy of statin therapy in those with high genetic risk (top quintile) with all others. For this analysis, 4910 participants from the WOSCOPS study were selected. Those in the placebo group with high genetic risk had an increased hazard ratio for the first CAD event, with a 25% increase in risk for each standard deviation. In the treatment group, those with high genetic risk experienced a 44% reduction in first coronary event, versus 24% in all others. The number needed to treat to prevent one coronary event was 13 among participants at high genetic risk and 38 among all others. The achieved LDL-c reduction was similar among groups. The authors also tested whether this high genetic risk group would be predisposed to subclinical atherosclerosis. To asses this hypothesis, the CARDIA and BioImage cohorts were analyzed. The authors found higher coronary artery calcification and carotid artery plaque burden in the high genetic risk group [21]. This approach was also proposed by Khera et al. [22••] who analyzed 50 SNPs in a cohort of 55,685 participants from four trials (ARIC, WGHS, MDCS, and BioImage). The aim of the study was to determine the extent to which a healthy lifestyle is associated with a reduced risk of CAD among participants at high genetic risk. The relative risk of incident coronary events was 91% higher among participants at high genetic risk; moreover, in this group, a favorable lifestyle was associated with a 46% lower relative risk of coronary events than an unhealthy lifestyle.

Precision Medicine and Familial Hypercholesterolemia

Some studies have shown that NGS can be used in primary care for the diagnosis of FH [100, 101]. Clinical practice guidelines based on systematic reviews supported by the US Centers for Disease Control and Prevention place cascade screening for close relatives of individuals with FH as a level 1 intervention [102]. Cascade testing using DNA analysis is also recommended in the clinical guidelines of the UK National Institute for Health and Care Excellence and in the consensus statement of the European Atherosclerosis Society on FH [103, 104]. Genetic testing may also be useful in therapeutic management of FH, since it is able to differentiate patients with compound heterozygous FH, heterozygous FH, homozygous FH, double heterozygous FH, or autosomal recessive FH [25, 28]. In addition, genetic testing may be

helpful in identifying probably or definitely pathogenic variants that indicate higher CV risk. In these cases, an earlier, more aggressive approach and strict adherence to therapy are indicated [34]. It is important to note that a successful molecular diagnosis depends on the ability of the designated method to evaluate both the heterogeneity of the locus and the allele associated with FH [37]. In many high-risk individuals, definitive diagnosis of FH cannot be made solely on the basis of clinical criteria. In a Latvian study enrolling patients with elevated LDL-c levels, the use of NGS identified FH-related mutations in 7.6% of all subjects [105].

Multicenter studies with larger sample sizes and better designs should be conducted to establish the feasibility and cost-effectiveness of this approach. It is noteworthy that, although genetic testing can improve patient identification and care in the setting of FH, clinical diagnosis is still the gold standard [106]. Finally, data suggest that a DNA-based diagnosis of FH appears to have a minimal adverse psychological impact, not being perceived as an anxiety trigger [107, 108].

Conclusion

The field of cardiovascular genetics is growing worldwide and establishing itself as a reality in the diagnostic and therapeutic armamentarium of cardiologists. The classification of primary dyslipidemia by heredity (monogenic or polygenic) is essential to its understanding. Furthermore, knowledge of the molecular basis of disease in individual patients allows the implementation of a precision medicine approach, thus promoting correct diagnosis, institution of optimized drug therapy based on pharmacogenetics, and prognostication. In addition, genetic diagnosis of an index case can trigger a family-wide investigation process (cascade screening), allowing early detection, guidance, therapy, and, consequently, reduction of CV risk in these individuals.

Finally, as with any other test, proper interpretation of the results of genetic screening is essential not only to establish the correct diagnosis but also to properly guide patients and their families. Therefore, careful assessment of the pathogenesis of any detected variants is a key aspect. All information available at major databases and publications should be taken into consideration, and information should be analyzed by a skilled team to ensure a reliable result.

Compliance with Ethical Standards

Conflict of Interest Ricardo Stein, Filipe Ferrari, and Fernando Scolari declare that they have no conflict of interest.

Human and Animal Rights and Informed Consent This article does not contain any studies with human or animal subjects performed by any of the authors.

References

Papers of particular interest, published recently, have been highlighted as:
- • Of importance
- •• Of major importance

1 Pol T, Held C, Westerbergh J, Lindbäck J, Alexander JH, Alings M, et al. Dyslipidemia and risk of cardiovascular events in patients with atrial fibrillation treated with oral anticoagulation therapy: insights from the ARISTOTLE (Apixaban for Reduction in Stroke and Other Thromboembolic Events in Atrial Fibrillation) Trial. J Am Heart Assoc. 2018;7(3):e007444. https://doi.org/10.1161/JAHA.117.007444.

2 Carroll MD, Fryar CD, Nguyen DT. Total and high-density lipoprotein cholesterol in adults: United States, 2015-2016. NCHS Data Brief. 2017;290:1–8 Available from https://www.cdc.gov.

3 Faggiano P, Pirillo A, Griffo R, Ambrosetti M, Pedretti R, Scorcu G, et al. Prevalence and management of familial hypercholesterolemia in patients with coronary artery disease: the heredity survey. Int J Cardiol. 2018;252:193–8. https://doi.org/10.1016/j.ijcard.2017.10.105.

4 Tietjen I, Hovingh GK, Singaraja R, Radomski C, McEwen J, Chan E, et al. Increased risk of coronary artery disease in Caucasians with extremely low HDL cholesterol due to mutations in ABCA1, APOA1, and LCAT. Biochim Biophys Acta. 2012;1821:416–24. https://doi.org/10.1016/j.bbalip.2011.08.006.

5 Sadananda SN, Foo JN, Toh MT, Cermakova L, Trigueros-Motos L, Chan T, et al. Targeted next-generation sequencing to diagnose disorders of HDL cholesterol. J Lipid Res. 2015;56:1993–2001. https://doi.org/10.1194/jlr.P058891.

6 Barter PJ, Caulfield M, Eriksson M, Grundy SM, Kastelein JJ, Komajda M, et al. Effects of torcetrapib in patients at high risk for coronary events. N Engl J Med. 2007;357(21):2109–22. https://doi.org/10.1056/NEJMoa0706628.

7 Tall AR, Yvan-Charvet L, Wang N. The failure of torcetrapib: was it the molecule or the mechanism? Arterioscler Thromb Vasc Biol. 2007;27(2):257–60. https://doi.org/10.1161/01.ATV.0000256728.60226.77.

8 Mattu RK, Needham EW, Morgan R, Rees A, Hackshaw AK, Stocks J, et al. DNA variants at the LPL gene locus associate with angiographically defined severity of atherosclerosis and serum lipoprotein levels in a Welsh population. Arterioscler Thromb. 1994;14(7):1090–7. https://doi.org/10.1161/01.ATV.14.7.1090.

9 Xie L, Li YM. Lipoprotein lipase (LPL) polymorphism and the risk of coronary artery disease: a meta-analysis. Int J Environ Res Public Health. 2017;14(1). https://doi.org/10.3390/ijerph14010084.

10 Kohan AB. Apolipoprotein C-III: a potent modulator of hypertriglyceridemia and cardiovascular disease. Curr Opin Endocrinol Diabetes Obes. 2015;22(2):119–25. https://doi.org/10.1097/MED.0000000000000136.

11 TG and HDL Working Group of the Exome Sequencing Project, National Heart, Lung, and Blood Institute Crosby J Peloso G Auer P Crosslin D, et al. Loss-of-function mutations in APOC3, triglycerides, and coronary disease. N Engl J Med. 2014;371(1):22–31. https://doi.org/10.1056/NEJMoa1307095.

12 Zhang JZ, Xie X, Ma YT, Zheng YY, Yang YN, Li XM, et al. Association between apolipoprotein C-III gene polymorphisms and coronary heart disease: a meta-analysis. Aging Dis. 2016;7(1):36–44. https://doi.org/10.14336/AD.2015.0709.

13 Rasmussen JN, Chong A, Alter DA. Relationship between adherence to evidence-based pharmacotherapy and long-term mortality

after acute myocardial infarction. JAMA. 2007;297:177–86. https://doi.org/10.1001/jama.297.2.177.

14 Kitzmiller JP, Mikulik EB, Dauki AM, Murkherjee C, Luzum JA. Pharmacogenomics of statins: understanding susceptibility to adverse effects. Pharmacogenomics Pers Med. 2016;9:97–106. https://doi.org/10.2147/PGPM.S86013.

15 Ramsey LB, Johnson SG, Caudle KE, Haidar CE, Voora D, Wilke RA, et al. The Clinical Pharmacogenetics Implementation Consortium (CPIC) guideline for SLCO1B1 and simvastatin-induced myopathy: 2014 update. Clin Pharmacol Ther. 2014;96(4):423–8. https://doi.org/10.1038/clpt.2014.125.

16 Armitage J, Holmes MV, Preiss D. Cholesteryl ester transfer protein inhibition for preventing cardiovascular events: JACC Review Topic of the Week. J Am Coll Cardiol. 2019;73(4):477–48. https://doi.org/10.1016/j.jacc.2018.10.072.

17 Webb TR, Erdmann J, Stirrups KE, Stitziel NO, Masca N1, Jansen H, et al. Systematic evaluation of pleiotropy identifies 6 further loci associated with coronary artery disease. J Am Coll Cardiol. 2017;69:823–36. https://doi.org/10.1016/j.jacc.2016.11.056.

18 Boekholdt SM, Sacks FM, Jukema JW, Shepherd J, Freeman DJ, McMahon AD, et al. Cholesteryl ester transfer protein TaqIB variant, high-density lipoprotein cholesterol levels, cardiovascular risk, and efficacy of pravastatin treatment: individual patient meta-analysis of 13,677 subjects. Circulation. 2005;111:278–87. https://doi.org/10.1161/01.CIR.0000153341.46271.40.

19 Sabatine MS, Giugliano RP, Keech AC, Honarpour N, Wiviott SD, Murphy SA, et al. Evolocumab and clinical outcomes in patients with cardiovascular disease. N Engl J Med. 2017;376(18):1713–22. https://doi.org/10.1056/NEJMoa1615664.

20 Schwartz GG, Steg PG, Szarek M, Bhatt DL, Bittner VA, Diaz R, et al. Alirocumab and cardiovascular outcomes after acute coronary syndrome. N Engl J Med. 2018;379(22):2097–107. https://doi.org/10.1056/NEJMoa1801174.

21 Natarajan P, Young R, Stitziel NO, Padmanabhan S, Baber U, Mehran R, et al. Polygenic risk score identifies subgroup with higher burden of atherosclerosis and greater relative benefit from statin therapy in the primary prevention setting. Circulation. 2017;135(22):2091–101. https://doi.org/10.1161/CIRCULATIONAHA.116.024436.

22•• Khera AV, Emdin CA, Drake I, Natarajan P, Bick AG, Cook NR, et al. Genetic risk, adherence to a healthy lifestyle, and coronary disease. N Engl J Med. 2016;375(24):2349–58. https://doi.org/10.1056/NEJMoa1605086 **The results of this study showed that in individuals with high genetic risk for coronary artery disease, the adoption of healthy lifestyle habits can reduce the relative risk of coronary heart disease by 50%, when compared with those individuals with unhealthy lifestyle.**

23 Sjouke B, Kusters DM, Kindt I, Besseling J, Defesche JC, Sijbrands EJ, et al. Homozygous autosomal dominant hypercholesterolaemia in the Netherlands: prevalence, genotype-phenotype relationship, and clinical outcome. Eur Heart J. 2015;36(9):560–5. https://doi.org/10.1093/eurheartj/ehu058.

24 Turgeon RD, Barry AR, Pearson GJ. Familial hypercholesterolemia: review of diagnosis, screening, and treatment. Can Fam Physician. 2016;62(1):32–7 Availabe at http://www.cfp.ca.

25 Migliara G, Baccolini V, Rosso A, D'Andrea E, Massimi A, Villari P, et al. Familial hypercholesterolemia: a systematic review of guidelines on genetic testing and patient management. Front Public Health. 2017;5:252. https://doi.org/10.3389/fpubh.2017.00252.

26 Onorato A, Sturm AC. Heterozygous familial hypercholesterolemia. Circulation. 2016;133(14):e587–9. https://doi.org/10.1161/CIRCULATIONAHA.115.020701.

27 Poonia A, Giridhara P. Xanthomas in familial hypercholesterolemia. N Engl J Med. 2017;377(5):e7. https://doi.org/10.1056/NEJMicm161614.

28 Wang X, Jiang L, Sun LY, Wu Y, Wen WH, Wang XF, et al. Genetically confirmed familial hypercholesterolemia in outpatients with hypercholesterolemia. J Geriatr Cardiol. 2018;15(6):434–40. https://doi.org/10.11909/j.issn.1671-5411.2018.06.006.

29 Bouhairie EV, Goldberg AC. Familial hypercholesterolemia. Cardiol Clin. 2015;33(2):169–79. https://doi.org/10.1016/j.ccl.2015.01.001.

30 Kataoka Y, Harada-Shiba M, Nakao K, Nakashima T, Kawakami S, Fujino M, et al. Mature proprotein convertase subtilisin/kexin type 9, coronary atheroma burden, and vessel remodeling in heterozygous familial hypercholesterolemia. J Clin Lipidol. 2017;11(2):413–421.e3. https://doi.org/10.1016/j.jacl.2017.01.005.

31 Thompson GR, Blom DJ, Marais AD, Seed M, Pilcher GJ, Raal FJ. Survival in homozygous familial hypercholesterolaemia is determined by the on-treatment level of serum cholesterol. Eur Heart J. 2018;39(14):1162–8. https://doi.org/10.1093/eurheartj/ehx317.

32 Aljenedil S, Ruel I, Watters K, Genest J. Severe xanthomatosis in heterozygous familial hypercholesterolemia. J Clin Lipidol. 2018;12(4):872–7. https://doi.org/10.1016/j.jacl.2018.03.087.

33 Li JJ, Li S, Zhu CG, Wu NQ, Zhang Y, Guo YL, et al. Familial hypercholesterolemia phenotype in Chinese patients undergoing coronary angiography. Arterioscler Thromb Vasc Biol. 2017;37(3):570–9. https://doi.org/10.1161/ATVBAHA.116.308456.

34 Sturm AC, Knowles JW, Gidding SS, Ahmad ZS, Ahmed CD, Ballantyne CM, et al. Clinical genetic testing for familial hypercholesterolemia: JACC Scientific Expert Panel. J Am Coll Cardiol. 2018;72(6):662–80. https://doi.org/10.1016/j.jacc.2018.05.044.

35 Khera AV, Won HH, Peloso GM, Lawson KS, Bartz TM, Deng X, et al. Diagnostic yield and clinical utility of sequencing familial hypercholesterolemia genes in patients with severe hypercholesterolemia. J Am Coll Cardiol. 2016;67(22):2578–89. https://doi.org/10.1016/j.jacc.2016.03.520.

36 Henderson R, O'Kane M, McGilligan V, Watterson S. The genetics and screening of familial hypercholesterolaemia. J Biomed Sci. 2016;23:39. https://doi.org/10.1186/s12929-016-0256-1.

37 Iacocca MA, Hegele RA. Recent advances in genetic testing for familial hypercholesterolemia. Expert Rev Mol Diagn. 2017;17(7):641–51. https://doi.org/10.1080/14737159.2017.1332997.

38 WHO Human Genetics Programme. Familial hypercholesterolaemia (FH): report of a second WHO consultation [online]. http://apps.who.int/iris/handle/10665/66346 (1998).

39 Defesche JC, Lansberg PJ, Umans-Eckenhausen MA, Kastelein JJ. Advanced method for the identification of patients with inherited hypercholesterolemia. Semin Vasc Med. 2004;49(1):59–65.

40 Besser J, Carleton HA, Gerner-Smidt P, Lindsey RL, Trees E. Next-generation sequencing technologies and their application to the study and control of bacterial infections. Clin Microbiol Infect. 2018;24(4):335–41. https://doi.org/10.1016/j.cmi.2017.10.013.

41 Prospective Studies Collaboration, Lewington S, Whitlock G, Clarke R, Sherliker P, Emberson J, et al. Blood cholesterol and vascular mortality by age, sex, and blood pressure: a meta-analysis of individual data from 61 prospective studies with 55,000 vascular deaths. Lancet. 2007;370(9602):1829–39. https://doi.org/10.1016/S0140-6736(07)61778-4.

42 Catapano AL, Pirillo A, Norata GD. Vascular inflammation and low-density lipoproteins: is cholesterol the link? A lesson from the clinical trials. Br J Pharmacol. 2017;174(22):3973–85. https://doi.org/10.1111/bph.

43 Ference BA, Ginsberg HN, Graham I, Ray KK, Packard CJ, Bruckert E, et al. Low-density lipoproteins cause atherosclerotic

cardiovascular disease. 1. Evidence from genetic, epidemiologic, and clinical studies. A consensus statement from the European Atherosclerosis Society Consensus Panel. Eur Heart J. 2017;38(32):2459–72. https://doi.org/10.1093/eurheartj/ehx144.

44• Silverman MG, Ference BA, Im K, Wiviott SD, Giugliano RP, Grundy SM, et al. Association between lowering LDL-C and cardiovascular risk reduction among different therapeutic interventions: a systematic review and meta-analysis. JAMA. 2016;316(12):1289–97. https://doi.org/10.1001/jama.2016.13985 **In this analysis, with more than 300,000 individuals included, it confirms that achieving lower levels of LDL is associated with lower rates of major coronary events. A reduction of 1.5% and 4.6% in the rate of major cardiovascular events (cardiovascular death and acute myocardial infarction) was observed for each reduction of 1 mmol/L in LDL in primary and secondary prevention studies, respectively**.

45 Navarese EP, Robinson JG, Kowalewski M, Kolodziejczak M, Andreotti F, Bliden K, et al. Association between baseline LDL-C level and total and cardiovascular mortality after LDL-C lowering: a systematic review and meta-analysis. JAMA. 2018;319(15):1566–79. https://doi.org/10.1001/jama.2018.2525.

46 Teslovich TM, Musunuru K, Smith AV, Edmondson AC, Stylianou IM, Koseki M, et al. Biological, clinical and population relevance of 95 loci for blood lipids. Nature. 2010;466(7307):707–13. https://doi.org/10.1038/nature09270.

47 Talmud PJ, Shah S, Whittall R, Futema M, Howard P, Cooper JA, et al. Use of low-density lipoprotein cholesterol gene score to distinguish patients with polygenic and monogenic familial hypercholesterolaemia: a case-control study. Lancet. 2013;381:1293–301. https://doi.org/10.1016/S0140-6736(12)62127-8.

48 Di Angelantonio E, Sarwar N, Perry P, Kaptoge S, Ray KK, Thompson A, et al. Major lipids, apolipoproteins, and risk of vascular disease. JAMA. 2009;302:1993–2000. https://doi.org/10.1001/jama.2009.1619.

49 Rosenson RS. The high-density lipoprotein puzzle: why classic epidemiology, genetic epidemiology, and clinical trials conflict? Arterioscler Thromb Vasc Biol. 2016;36(5):777–82. https://doi.org/10.1161/ATVBAHA.116.307024.

50 Voight BF, Peloso GM, Orho-Melander M, Frikke-Schmidt R, Barbalic M, Jensen MK, et al. Plasma HDL cholesterol and risk of myocardial infarction: a Mendelian randomisation study. Lancet. 2012;380(9841):572–80. https://doi.org/10.1016/S0140-6736(12)60312-2.

51 Kawashiri MA, Tada H, Nomura A, Yamagishi M. Mendelian randomization: its impact on cardiovascular disease. J Cardiol. 2018;72(4):307–13. https://doi.org/10.1016/j.jjcc.2018.04.007.

52 Haase CL, Tybjærg-Hansen A, Grande P, Frikke-Schmidt R. Genetically elevated apolipoprotein A-I, high-density lipoprotein cholesterol levels, and risk of ischemic heart disease. J Clin Endocrinol Metab. 2010;95:E500–10. https://doi.org/10.1210/jc.2010-0450.

53 Vergeer M, Holleboom AG, Kastelein JJ, Kuivenhoven JA. The HDL hypothesis: does high-density lipoprotein protect from atherosclerosis? J Lipid Res. 2010;51(8):2058–73. https://doi.org/10.1194/jlr.R001610.

54 Zhong S, Sharp DS, Grove JS, Bruce C, Yano K, Curb JD, et al. Increased coronary heart disease in Japanese-American men with mutation in the cholesteryl ester transfer protein gene despite increased HDL levels. J Clin Invest. 1996;97(12):2917–23. https://doi.org/10.1172/JCI118751.

55 Cohen JC, Kiss RS, Pertsemlidis A, Marcel YL, McPherson R, Hobbs HH. Multiple rare alleles contribute to low plasma levels of HDL cholesterol. Science. 2004;305:869–72. https://doi.org/10.1126/science.1099870.

56 Abdel-Razek O, Sadananda SN, Li X, Cermakova L, Frohlich J, Brunham LR. Increased prevalence of clinical and subclinical atherosclerosis in patients with damaging mutations in ABCA1 or APOA1. J Clin Lipidol. 2018;12(1):116–21. https://doi.org/10.1016/j.jacl.2017.10.010.

57 Hassan M, Philip P. CANHEART: is HDL cholesterol a cardiovascular specific risk factor? Glob Cardiol Sci Pract. 2016;2016(4):e201634. https://doi.org/10.21542/gcsp.2016.34.

58 Hooper AJ, Crawford GM, Brisbane JM, Robertson K, Watts GF, van Bockxmeer FM, et al. Familial lipoprotein lipase deficiency caused by known (G188E) and novel (W394X) LPL gene mutations. Ann Clin Biochem. 2008;45(Pt 1):102–5. https://doi.org/10.1258/acb.2007.007080.

59 Ashraf AP, Beukelman T, Pruneta-Deloche V, Kelly DR, Garg A. Type 1 hyperlipoproteinemia and recurrent acute pancreatitis due to lipoprotein lipase antibody in a young girl with Sjogren's syndrome. J Clin Endocrinol Metab. 2011;96(11):3302–7. https://doi.org/10.1210/jc.2011-1113.

60 Zhang WS, Zhang WH, Liu QJ. Lipoprotein lipase gene Hind III polymorphism was associated with hemorrhagic stroke. Int J Clin Exp Med. 2015;8(6):9575–9 Available at http://www.ijcem.com/.

61 Benlian P, Foubert L, Gagné E, Bernard L, De Gennes JL, Langlois S, et al. Complete paternal isodisomy for chromosome 8 unmasked by lipoprotein lipase deficiency. Am J Hum Genet. 1996;59(2):431–6 Available at www.cell.com.

62 Ahmadi Z, Senemar S, Toosi S, Radmanesh S. The association of lipoprotein lipase genes, HindIII and S447X polymorphisms with coronary artery disease in Shiraz City. J Cardiovasc Thorac Res. 2015;7(2):63–7. https://doi.org/10.15171/jcvtr.2015.14.

63 Daoud MS, Ataya FS, Fouad D, Alhazzani A, Shehata AI, Al-Jafari AA. Associations of three lipoprotein lipase gene polymorphisms, lipid profiles and coronary artery disease. Biomed Rep. 2013;1(4):573–82. https://doi.org/10.3892/br.2013.126.

64 Heng CK, He X, Saha N, Low PS, Demirci FY, Kamboh MI. Association of three lipoprotein lipase polymorphisms with coronary artery disease in Chinese and Asian Indians. Int J Cardiol. 2010;144(1):142–3. https://doi.org/10.1016/j.ijcard.2008.12.121.

65 Cui F, Li K, Li Y, Zhang X, An C. Apolipoprotein C3 genetic polymorphisms are associated with lipids and coronary artery disease in a Chinese population. Lipids Health Dis. 2014;13:170. https://doi.org/10.1186/1476-511X-13-170.

66 Wang Y, Yin X, Li L, Deng S, He Z. Association of apolipoprotein C3 genetic polymorphisms with the risk of ischemic stroke in the northern Chinese Han population. PLoS One. 2016;11(9):e0163910. https://doi.org/10.1371/journal.pone.0163910.

67 Fitzgerald K, White S, Borodovsky A, Bettencourt BR, Strahs A, Clausen V, et al. A highly durable RNAi therapeutic inhibitor of PCSK9. N Engl J Med. 2017;376(1):41–51. https://doi.org/10.1056/NEJMc1703361.

68 Abifadel M, Varret M, Rabes JP, Allard D, Ouguerram K, Devillers M, et al. Mutations in PCSK9 cause autosomal dominant hypercholesterolemia. Nat Genet. 2003;34:154–6. https://doi.org/10.1038/ng1161.

69 Qiu C, Zeng P, Li X, Zhang Z, Pan B, Peng Z, et al. What is the impact of PCSK9 rs505151 and rs11591147 polymorphisms on serum lipids level and cardiovascular risk: a meta-analysis. Lipids Health Dis. 2017;16(1):111. https://doi.org/10.1186/s12944-017-0506-6.

70 Cai G, Zhang B, Shi G, Weng W, Ma C, Song Y. The associations between proprotein convertase subtilisin/kexin type 9 e670g polymorphism and the risk of coronary artery disease and serum lipid levels: a meta-analysis. Lipids Health Dis. 2015;14:149. https://doi.org/10.1186/s12944-015-0154-7.

71 Adi D, Xie X, Liu F, Ma YT, Abudoukelimu M, Wu Y. Relationships between genetic polymorphisms of e670g in pcsk9

gene and coronary artery disease: a meta-analysis. Int J Clin Exp Med. 2015;8:13251–8 Available at http://www.ijcem.com/.

72 Grundy SM, Stone NJ, Bailey AL, Beam C, Birtcher KK, Blumenthal RS, et al. 2018 AHA/ACC/AACVPR/AAPA/ABC/ACPM/ADA/AGS/APhA/ ASPC/NLA/PCNA guideline on the management of blood cholesterol. Circulation. 2018. https://doi.org/10.1161/CIR.0000000000000625.

73 Grundy SM, Stone NJ, Bailey AL, Beam C, Birtcher KK, Blumenthal RS, et al. 2018 AHA/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. 2018. https://doi.org/10.1016/j.jacc.2018.11.002.

74 Yebyo HG, Aschmann HE, Kaufmann K, Puhan MA. Comparative effectiveness and safety of statins as a class and of specific statins for primary prevention of cardiovascular disease: a systematic review, meta-analysis and network meta-analysis of randomized trials with 94,283 participants. Am Heart J. 2019;210:18–28. https://doi.org/10.1016/j.ahj.2018.12.007.

75 Koskinas KC, Siontis GCM, Piccolo R, Mavridis D, Räber L, Mach F, et al. Effect of statins and non-statin LDL-lowering medications on cardiovascular outcomes in secondary prevention: a meta-analysis of randomized trials. Eur Heart J. 2018;39(14):1172–80. https://doi.org/10.1093/eurheartj/ehx566.

76 García-Giustiniani D, Stein R. Genetics of dyslipidemia. Arq Bras Cardiol. 2016;106(5):434–8. https://doi.org/10.5935/abc.20160074.

77 Josephine LH, Joy SV, Haga SB, Orlando LA, Kraus WE, Ginsburg GS, et al. Genetically guided statin therapy on statin perceptions, adherence, and cholesterol lowering: a pilot implementation study in primary care patients. J Pers Med. 2014;4(2):147–62. https://doi.org/10.3390/jpm4020147.

78 Relling MV, Krauss RM, Roden DM, Klein TE, Fowler DM, Terada N, et al. New pharmacogenomics research network: an open community catalyzing research and translation in precision medicine. Clin Pharmacol Ther. 2017;102(6):897–902. https://doi.org/10.1002/cpt.755.

79 Peyser B, Perry EP, Singh K, Gill RD, Mehan MR, Haga SB, et al. Effects of delivering SLCO1B1 pharmacogenetic information in randomized trial and observational settings. Circ Genom Precis Med. 2018;11(9):e002228. https://doi.org/10.1161/CIRCGEN.118.002228.

80 Hoenig MR, Walker PJ, Gurnsey C, Beadle K, Johnson L. The C3435T polymorphism in ABCB1 influences atorvastatin efficacy and muscle symptoms in a high-risk vascular cohort. J Clin Lipidol. 2011;5(2):91–6. https://doi.org/10.1016/j.jacl.2011.01.001.

81 DeGorter MK, Tirona RG, Schwarz UI, Choi YH, Dresser GK, Suskin N, et al. Clinical and pharmacogenetic predictors of circulating atorvastatin and rosuvastatin concentrations in routine clinical care. Circ Cardiovasc Genet. 2013;6(4):400–8. https://doi.org/10.1161/CIRCGENETICS.113.000099.

82 Wang D, Guo Y, Wrighton SA, Cooke GE, Sadee W. Intronic polymorphism in CYP3A4 affects hepatic expression and response to statin drugs. Pharmacogenomics J. 2011;11(4):274–86. https://doi.org/10.1038/tpj.2010.28.

83 Tsamandouras N, Dickinson G, Guo Y, Hall S, Rostami-Hodjegan A, Galetin A, et al. Identification of the effect of multiple polymorphisms on the pharmacokinetics of simvastatin and simvastatin acid using a population-modeling approach. Clin Pharmacol Ther. 2014;96(1):90–100. https://doi.org/10.1038/clpt.2014.55.

84 Thompson JF, Hyde CL, Wood LS, Paciga SA, Hinds DA, Cox DR, et al. Comprehensive whole-genome and candidate gene analysis for response to statin therapy in the Treating to New Targets (TNT) cohort. Circ Cardiovasc Genet. 2009;2:173–81. https://doi.org/10.1161/CIRCGENETICS.108.818062.

85 Thompson JF, Man M, Johnson KJ, Wood LS, Lira ME, Lloyd DB, et al. An association study of 43 SNPs in 16 candidate genes with atorvastatin response. Pharmacogenomics J. 2005;5:352–8. https://doi.org/10.1038/sj.tpj.6500328.

86 Regieli JJ, Jukema JW, Grobbee DE, Kastelein JJ, Kuivenhoven JA, Zwinderman AH, et al. CETP genotype predicts increased mortality in statin-treated men with proven cardiovascular disease: an adverse pharmacogenetic interaction. Eur Heart J. 2008;29:2792–9. https://doi.org/10.1093/eurheartj/ehn465.

87 Nieminen T, Kähönen M, Viiri LE, Grönroos P, Lehtimäki T. Pharmacogenetics of apolipoprotein E gene during lipid-lowering therapy: lipid levels and prevention of coronary heart disease. Pharmacogenomics. 2008;9(10):1475–86. https://doi.org/10.2217/14622416.9.10.1475.

88 Xiang Q, Zhang X, Ma L, Hu K, Zhang Z, Mu G, et al. The association between the SLCO1B1, apolipoprotein E, and CYP2C9 genes and lipid response to fluvastatin: a meta-analysis. Pharmacogenet Genomics. 2018;28(12):261–7. https://doi.org/10.2217/14622416.9.10.1475.

89 Ferrari F, Stein R, Motta MT, Moriguchi EH. PCSK9 inhibitors: clinical relevance, molecular mechanisms, and safety in clinical practice. Arq Bras Cardiol. 2019. https://doi.org/10.5935/abc.20190029.

90 Giugliano RP, Keech A, Murphy SA, Huber K, Tokgozoglu SL, Lewis BS, et al. Clinical efficacy and safety of evolocumab in high-risk patients receiving a statin: secondary analysis of patients with low LDL cholesterol levels and in those already receiving a maximal-potency statin in a randomized clinical trial. JAMA Cardiol. 2017;2(12):1385–91. https://doi.org/10.1001/jamacardio.2017.3944.

91 Leibowitz M, Cohen-Stavi C, Basu S, Balicer RD. Targeting LDL cholesterol: beyond absolute goals toward personalized risk. Curr Cardiol Rep. 2017;19(6):52. https://doi.org/10.1007/s11886-017-0858-6.

92 Humphries SE, Cooper JA, Seed M, Capps N, Durrington PN, Jones B, et al. Coronary heart disease mortality in treated familial hypercholesterolaemia: update of the UK Simon Broome FH register. Atherosclerosis. 2018;274:41–6. https://doi.org/10.1016/j.atherosclerosis.2018.04.040.

93 Sabatine MS. PCSK9 inhibitors: clinical evidence and implementation. Nat Rev Cardiol. 2019;16(3):155–65. https://doi.org/10.1038/s41569-018-0107-8.

94 Kosmas CE, Muñoz Estrella A, Sourlas A, Silverio D, Hilario E, Montan PD, et al. Inclisiran: a new promising agent in the management of hypercholesterolemia. Diseases. 2018;6(3):E63. https://doi.org/10.3390/diseases6030063.

95 Stoekenbroek RM, Kallend D, Wijngaard PL, Kastelein JJ. Inclisiran for the treatment of cardiovascular disease: the ORION clinical development program. Futur Cardiol. 2018;14(6):433–42. https://doi.org/10.2217/fca-2018-0067.

96 Ray KK, Landmesser U, Leiter LA, Kallend D, Dufour R, Karakas M, et al. Inclisiran in patients at high cardiovascular risk with elevated LDL cholesterol. N Engl J Med. 2017;376(15):1430–40. https://doi.org/10.1056/NEJMoa1615758.

97 Leiter LA, Teoh H, Kallend D, Wright RS, Landmesser U, Wijngaard PLJ, et al. Inclisiran lowers LDL-C and PCSK9 irrespective of diabetes status: the ORION-1 randomized clinical trial. Diabetes Care. 2019;42(1):173–6. https://doi.org/10.2337/dc18-1491.

98 TIMI Study Group. HPS-4/TIMI 65 ORION-4. TIMI.org http://www.timi.org/index.php?page=orion-4 (2018).

99 Nishikido T, Ray KK. Inclisiran for the treatment of dyslipidemia. Expert Opin Investig Drugs. 2018;27(3):287–94.

100 Vandrovcova J, Thomas ER, Atanur SS, Norsworthy PJ, Neuwirth C, Tan Y, et al. The use of next-generation sequencing in clinical diagnosis of familial hypercholesterolemia. Genet Med. 2013;15(12):948–57. https://doi.org/10.1038/gim.2013.55.

101 Norsworthy PJ, Vandrovcova J, Thomas ER, Campbell A, Kerr SM, Biggs J, et al. Targeted genetic testing for familial hypercholesterolaemia using next generation sequencing: a population-based study. BMC Med Genet. 2014;15:70. https://doi.org/10.1186/1471-2350-15-70.

102 CDC Office of Public Health Genomics Tier Table Database. Available at: https://phgkb.cdc. gov/PHGKB/topicStartPage. action. Accessed 16 Dec 2018.

103 National Institute for Health and Care Excellence: clinical guidelines. Available at: https://www.ncbi. nlm.nih.gov/books/NBK11822/. Accessed 18 Dec 2018.

104 Nordestgaard BG, Chapman MJ, Humphries SE, Ginsberg HN, Masana L, Descamps OS, et al. Familial hypercholesterolaemia is underdiagnosed and undertreated in the general population: guidance for clinicians to prevent coronary heart disease: consensus statement of the European Atherosclerosis Society. Eur Heart J. 2013;34:3478–90a. https://doi.org/10.1093/eurheartj/eht273.

105 Radovica-Spalvina I, Latkovskis G, Silamikelis I, Fridmanis D, Elbere I, Ventins K, et al. Next-generation-sequencing-based identification of familial hypercholesterolemia-related mutations in subjects with increased LDL-C levels in a Latvian population. BMC Med Genet. 2015;16:86. https://doi.org/10.1186/s12881-015-0230-x.

106 Hopkins PN, Lane SR. Genotype-guided diagnosis in familial hypercholesterolemia: clinical management and concerns. Curr Opin Lipidol. 2017;28(2):144–51. https://doi.org/10.1097/MOL.0000000000000397.

107 Marteau T, Senior V, Humphries SE, Bobrow M, Cranston T, Crook MA, et al. Psychological impact of genetic testing for familial hypercholesterolemia within a previously aware population: a randomized controlled trial. Am J Med Genet Part A. 2004;128A:285–93. https://doi.org/10.1002/ajmg.a.30102.

108 Hallowell N, Jenkins N, Douglas M, Walker S, Finnie R, Porteous M, et al. A qualitative study of patients' perceptions of the value of molecular diagnosis for familial hypercholesterolemia (FH). J Commun Genet. 2017;8:45–52. https://doi.org/10.1007/s12687-016-0286-0.