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Review

Remnant lipoproteins and atherosclerotic cardiovascular disease

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ABSTRACT

Lipoproteins are one of the major risk factors for atherosclerotic cardiovascular disease (ASCVD), among which, low-density lipoprotein (LDL) particles have been definitively shown to be causally associated with the development of ASCVD. Additionally, the concept of remnant lipoproteins has emerged as lipoprotein metabolism has been fully investigated. The principal concept of this lipoprotein category is triglyceride-rich lipoproteins significantly increase at the postprandial state. Although there is no clear definition of remnant lipoproteins, they typically include chylomicron remnants, which are lipolyzed particles from chylomicron, as well as very low-density lipoprotein (VLDL) and intermediate-density lipoprotein (IDL) remnants that are lipolyzed particles from VLDL and IDL particles. However, the most important factor of these lipoproteins is such remnant lipoproteins seem to be causally associated with ASCVD, independent of LDL particles or LDL cholesterol. It has been challenging to assert a causal association of remnant lipoproteins and ASCVD; however, accumulated evidence from epidemiological studies, as well as recent Mendelian randomization studies from common and rare genetic variations strongly support this association. In this article, a basic explanation of lipoprotein metabolism is presented, including remnant lipoproteins and the important causal associations with ASCVD from a clinical point of view.

1. Introduction

It has been definitively established that low-density lipoprotein (LDL) cholesterol is a causal risk factor for atherosclerotic cardiovascular disease (ASCVD). Over the decades, LDL-lowering therapies, including statins, ezetimibe, and proprotein convertase subtilisin-kexin type 9 (PCSK9) inhibitors have been introduced in clinical settings, and they have contributed to better prognosis [1–3]. In addition to LDL cholesterol, triglyceride-rich remnant lipoproteins are also known as important causal risk factors for ASCVD [4], and they are usually regarded as one of the major residual risk factors of standard LDL-lowering therapies [5]. However, triglycerides as well as remnant lipoproteins are not currently receiving adequate attention from researchers and physicians, especially from cardiologists who should carefully assess a patient's risk of ASCVD [6]. One of the major reasons for this oversight may be the difficulty of the concept of remnant lipoproteins, and another may be lack of evidence from randomized controlled trials (RCTs) supporting causal associations between those biomarkers and ASCVD. In this article, supporting evidence is presented that remnant lipoproteins are a causal residual risk factor for ASCVD from various aspects, including human genetics from common to rare variations associated with remnant lipoproteins and clinical data.

2. What are remnant lipoproteins?

It is important to define the concept of remnant lipoproteins, because the definition may vary somewhat among researchers [7]. The initial concept of remnant lipoproteins was intermediate-density lipoproteins (IDLs) isolated by an ultracentrifugation method, since remnant lipoproteins had been considered as smaller particles derived from chylomicrons and very low-density lipoproteins (VLDLs). In 1979, Zilversmit reported that postprandial remnant lipoproteins that increased after food intake were associated with the development of atherosclerosis, although only chylomicron and chylomicron remnants secreted from intestine were proposed as risk factors for atherosclerosis at that time [8]. On the other hand, Havel and Nakajima have shown that VLDL remnants secreted primarily from the liver, rather than chylomicron remnants, are the major postprandial remnant lipoproteins associated with the development of atherosclerosis [9,10]. According to some earlier studies, there are several ways to determine plasma remnant lipoproteins, including electrophoresis, nuclear magnetic resonance, and high-performance liquid chromatography. These methods

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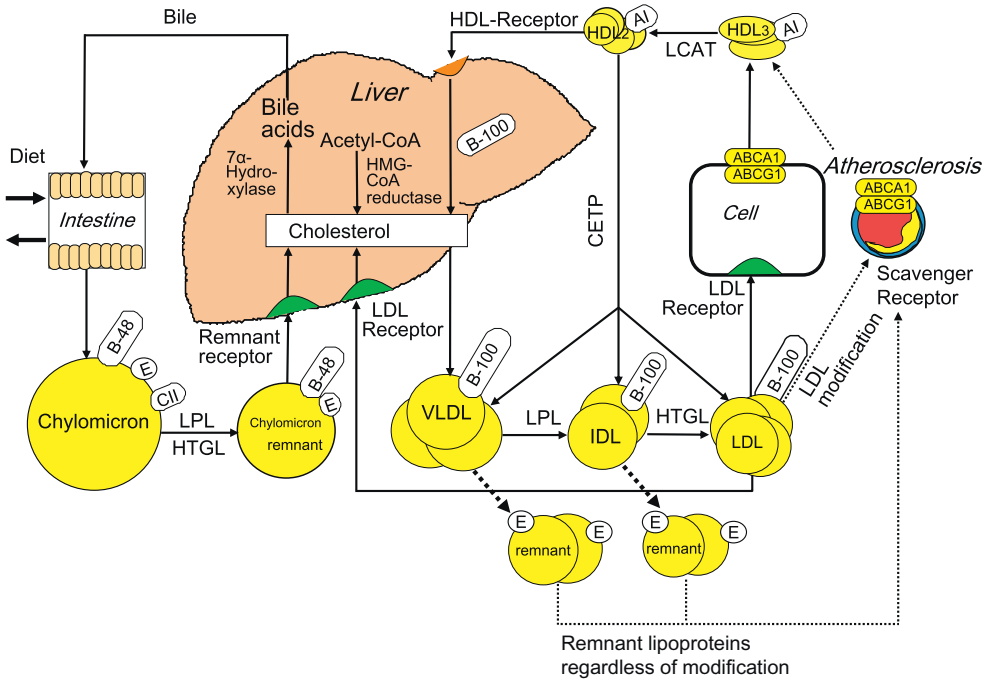

Fig. 1. Scheme of lipoprotein metabolism. The liver plays a central role in lipoprotein metabolism. Very low-density lipoprotein (VLDL) is secreted from the liver, then hydrolyzed to intermediate-density lipoprotein (IDL) and low-density lipoprotein (LDL) by lipoprotein lipase (LPL) and hepatic triglyceride lipase (HTGL). During this process, remnant lipoproteins, including VLDL remnants and IDL remnants, are introduced into circulation. If LDL particles and/or remnant lipoprotein particles are oxidized, those lipoproteins may lead to the development of atherosclerosis. Alternatively, chylomicron is secreted from the intestine, then hydrolyzed to chylomicron remnants by LDL and HTGL. The atherogenicity of chylomicron remnants is not yet clear.

are used to detect remnant lipoproteins by charge, particle size, estimated by the equations and the immuno-separation method [11–13]. However, the most important characteristic of remnant lipoproteins is triglyceride-rich lipoproteins that increase significantly after food intake. The diagram of lipoprotein metabolism is illustrated in Fig. 1.

3. Remnant lipoproteins and triglycerides

As stated above, remnant lipoproteins contain many triglycerides, and both biomarkers are significantly positively correlated in most cases. However, there are some rare conditions where triglycerides are significantly elevated and remnant lipoproteins are absent. The representative example is familial hyperchylomicronemia syndrome (FHS) [14]. Some patients with FHS without any lipoprotein lipase (LPL) activity are found to be rich in nascent chylomicrons and VLDL in plasma. Because of the lack of LPL activity, there is a lack of remnant lipoprotein formation on the endothelium [15,16]. And it has been shown that such patients do not always exhibit ASCVD, despite significant elevation of triglyceride levels. It is not clear if the lack of remnant lipoproteins is the cause of their protective phenotype. In this regard, it was reported in a study 215 Japanese patients that cases with plasma triglycerides concentration above 1000 mg/dl were frequently complicated with pancreatitis but rarely with cardiovascular diseases [17]. Considering those observations, remnant lipoproteins rather than triglycerides are a true causal factor for ASCVD.

4. Remnant lipoproteins and postprandial dyslipidemias

Plasma triglycerides are known to be a surrogate for triglyceride-rich lipoproteins and are present as lipoprotein subclasses in chylomicron, VLDL, and their remnants (Fig. 2). Triglyceride-rich lipoproteins and their remnants are not homogeneous lipoprotein fractions and are significantly increased in the postprandial plasma. Considering the extreme conditions caused by specific genetic mutations may help elucidate this issue. For example, sitosterolemia, which is a rare autosomal recessive disorder of lipid metabolism, has exhibited disturbance of postprandial remnant lipoprotein metabolism (Fig. 3) [18]. In addition, postprandial remnant lipoprotein metabolism in patients with FH has also been shown to be disturbed [19]. Accordingly, assessments for lipids are more accurate at postprandial state rather than fasting state, in at least some situations where postprandial remnant lipoprotein metabolism may be disturbed. Moreover, the authors have reported an extremely rare case of autosomal recessive hypercholesterolemia (ARH), which was caused by the protein truncating variations in LDL receptor adaptor protein 1 gene. This disease has been considered as a phenocopy of homozygous FH based on the significant disturbance of LDL metabolism. However, remnant lipoprotein metabolism is preserved in ARH, in contrast to FH, leading to a relatively milder phenotype (Fig. 4) [20]. Interestingly, observations from this study are consistent with the results obtained using stable isotope kinetic study in vivo [21]. Therefore, it is notable that remnant lipoprotein metabolism is different even among similar diseases, and it may lead to the difference of their phenotypes.

5. Remnant lipoproteins and atherosclerotic cardiovascular disease

Remnant lipoproteins are larger than LDL particles, and it has been believed that their penetration into arterial walls would be limited from biophysical considerations alone. However, both apoB100 and apoB48 can be extracted from atherosclerotic plaque [22]. There is much experimental evidence suggesting that remnant lipoproteins can enter the arterial intima, contributing to atherosclerosis; whereas, chylomicron and chylomicron remnants are too large to penetrate the endothelial layer. In addition, the remnant lipoproteins do not require oxidative modification in order to be taken by macrophages, because the macrophages recognize apolipoprotein E on the surface of these lipoproteins, triggering lipoprotein uptake [23,24]. Hence, it is biologically plausible that remnant lipoproteins are in fact atherogenic.

Until recently, the association between remnant lipoproteins and ASCVD remained unknown; however, several clinical studies have shown that remnant lipoproteins are significantly associated with ASCVD events now that lipoproteins can be measured in clinical settings [25–28]. It is noteworthy that remnant lipoproteins are associated with ASCVD events independent of triglycerides, although remnant lipoproteins contain most of the triglycerides, and they are correlated with each other. Moreover, remnant lipoproteins are significantly associated with risk for ASCVD, even in patients with FH where LDL

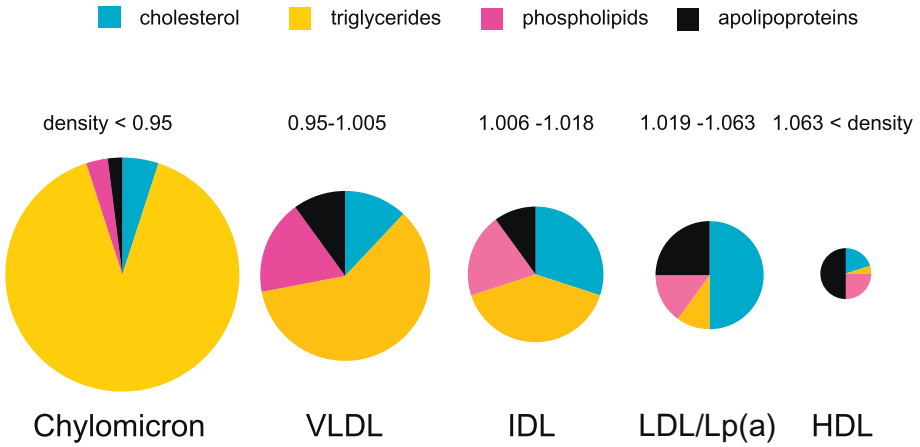

Fig. 2. Lipid content of each lipoprotein.
Light blue indicates cholesterol. Orange indicates triglycerides. Pink indicates phospholipids. Black indicates apolipoproteins. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

cholesterol is critically increasing their ASCVD risk [29].

In addition to these clinical data, a useful technique has been introduced to establish a causal relationship between a risk factor (usually a biomarker) and an outcome. Randomized controlled trials (RCTs), which require a large amount of time and effort, are the gold standard. In contrast, the Mendelian randomization study is a technique that uses genotypes as instruments to assess a causal relationship between biomarkers and outcomes [30]. In a Mendelian randomization study, a genetic variant associated with a specific biomarker is used as a proxy for the biomarker. Outcomes are compared between the group harboring the effect allele and a group with the reference allele. This approach can be considered a proxy for an RCT, in which the randomized groups have similar confounding variables. Accordingly, a Mendelian randomization study can be regarded as a natural RCT. In a case with remnant lipoproteins and triglycerides, recent Mendelian randomization studies effectively showed the causal relationships between those lipids and ASCVD. Using common genetic variations, Do et al. performed an interesting study to determine if triglyceride levels causally influenced the risk of ASCVD. They constructed a model adjusting the effects of LDL cholesterol and/or high-density lipoprotein (HDL) cholesterol levels on the risk of ASCVD and found that the genetic impact of single nucleotide polymorphisms (SNPs) on triglyceride levels was independently associated with the risk of ASCVD [31]. Moreover, they also showed robust associations between rare genetic variations associated with triglycerides and the risk of ASCVD in targeted analysis and in exome-wide analysis [32,33]. Interestingly, the gene-based association testing indicated that rare genetic variants associated with lower triglycerides were consistently associated with reduced cardiovascular risk, although those effects on the occurrence of diabetes were not consistent (Table 1). In addition to triglycerides, remnant lipoprotein cholesterol level driven by a genetic variant in apolipoprotein E gene has been shown to be associated with ASCVD [28].

These findings, ranging from clinical data to genetic studies with common and rare variants, collectively suggest that triglycerides as well as remnant lipoproteins are causally associated with ASCVD.

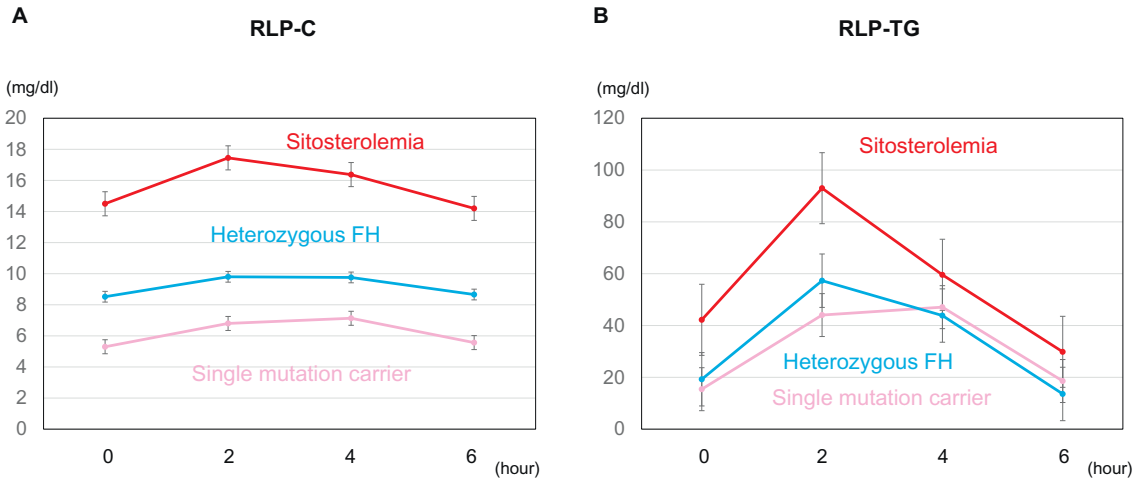

Fig. 3. Remnant-like lipoprotein metabolism in sitosterolemia, heterozygous familial hyperchylomicronemia syndrome (FHS), and in single-mutation carriers (ABCG5 or ABCG8 gene).
Bars indicate standard errors.
Red: Sitosterolemia.
Pink: Single-mutation carrier.
Light blue: Heterozygous FHS. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

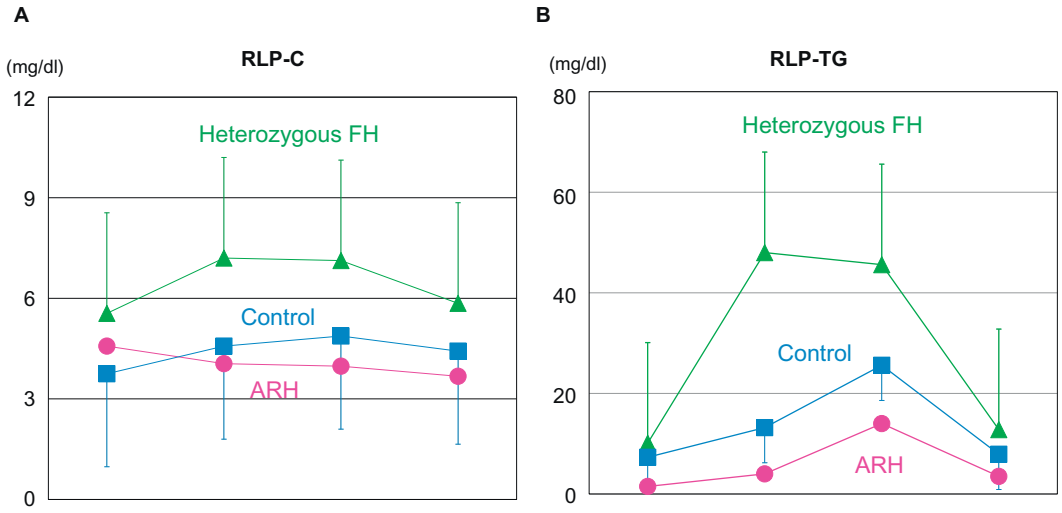

Fig. 4. Remnant-like lipoprotein metabolism in autosomal recessive hypercholesterolemia (ARH), heterozygous familial hyperchylomicronemia syndrome (FHS), and controls.
Bars indicate standard errors.
Green: Heterozygous FHS.
Pink: ARH.
Blue: Controls. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

6. Remnant lipoproteins or triglycerides? Fasting state or postprandial state?

As stated above, remnant lipoproteins and triglycerides have been shown to be associated with ASCVD. In addition, remnant lipoproteins and triglycerides levels are elevated at postprandial state, and it has been shown that assessment of such lipoproteins at postprandial state may predict future ASCVD events [34]. Conversely, assessment at fasting state is sometimes important for accurate diagnosis, especially in a case with inherited lipid disorders [35]. Accordingly, remnant lipoproteins and triglycerides at fasting and postprandial state should be measured for accurate diagnosis of lipid disorders and to fully assess the risk for ASCVD. However, there is no prior data investigating the clinical benefit of measuring both markers beyond the assessment of either of those markers. In addition, triglycerides are the major component of remnant lipoproteins, and both are typically significantly correlated. Therefore, assessments together with intervention for either marker may be adequate in most clinical situations. The important point is remnant lipoproteins as well as triglycerides are modifiable causal risk factors for ASCVD, and the current management status is not always sufficient [36].

7. Lipoprotein lipase (LPL) pathway and ASCVD

LPL has been shown to be one of the major drivers of triglyceride-rich lipoproteins, including remnant lipoproteins [37]. However, it has long been quite difficult to find the causal association between LPL pathway and the development of ASCVD. Recently, Kathiresan and colleagues investigated whether molecules in the LPL pathway were causally associated with ASCVD in large Mendelian randomization studies. They showed that individuals with apolipoprotein C3 loss-of function mutations exhibited lower triglycerides and lower incidence of ASCVD [38]. In addition, they found that apolipoprotein A5 missense mutations were significantly associated with early onset of myocardial infarction [39]. Moreover, rare variations in the LPL gene were significantly associated with ASCVD [32]. The discovery that molecules in LPL pathways are consistently associated with ASCVD suggests a strong causal association between the LPL pathway and the development of ASCVD.

8. Conclusion

In this paper, we have repeatedly emphasized that remnant

Table 1
Characteristics of participants.

Gene	Number of variants	Triglycerides	HDL cholesterol	LDL cholesterol	Odds ratio for ASCVD	Odds ratio for T2DM
LPL	7	−0.138 ± 0.002 ($< 1.0 \times 10^{-237}$)	0.939 ± 0.011 ($< 1.0 \times 10^{-237}$)	0.025 ± 0.012 (−0.03)	0.66	0.8
ANGPTL4	1	−0.273 ± 0.01 (4.2×10^{-175})	0.891 ± 0.035 (4.8×10^{-146})	−0.014 ± 0.036 (−0.7)	0.6	0.67
APOA5	7	−0.227 ± 0.002 ($< 1.0 \times 10^{-237}$)	0.453 ± 0.009 ($< 1.0 \times 10^{-237}$)	−0.145 ± 0.009 (8.4×10^{-59})	0.76	0.88
APOC3	3	−1.069 ± 0.032 (3.2×10^{-237})	0.695 ± 0.03 (9.0×10^{-120})	−0.106 ± 0.03 (4.7×10^{-4})	0.85	1.07
ANGPTL3	1	−0.077 ± 0.003 (6.1×10^{-170})	−0.136 ± 0.035 (1.2×10^{-4})	−0.588 ± 0.037 (4.8×10^{-58})	0.89	1.18
ANGPTL8	1	−0.353 ± 0.051 (2.8×10^{-12})	1.221 ± 0.141 (5.0×10^{-18})	−0.167 ± 0.146 (−0.25)	0.74	1.58

Values are expressed as effect size ± standard error (*p*-value).
LPL: lipoprotein lipase; ANGPTL4: angiopoietin-like 4; APOA5: apolipoprotein A5; APOC3: apolipoprotein C3;
ANGPTL3: angiopoietin-like 3; ANGPTL8: angiopoietin-like 8; ASCVD: atherosclerotic cardiovascular diseases; T2DM: type 2 diabetes mellitus.

lipoproteins and triglycerides are residual, modifiable, and causal risk factors for ASCVD. Much more attention should be given to remnant lipoproteins as well as LDL cholesterol for additional reduction of ASCVD events.

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Declarations of interest

None

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