Published in final edited form as:

Arterioscler Thromb Vasc Biol. 2017 March ; 37(3): 580–588. doi:10.1161/ATVBAHA.116.308704.

Lp(a) mass levels increase significantly according to *APOE* genotype: An analysis of 431,239 patients

Patrick M. Moriarty, MD[1], **Stephen A. Varvel, PhD**[2], **Philip L.S.M. Gords, PhD**[3,4], **Joseph P. McConnell, PhD**[2], and **Sotirios Tsimikas, MD**[5]

[1]Division of Clinical Pharmacology, Department of Internal Medicine, University of Kansas Medical Center, Kansas City, KS

[2]Salveo Diagnostics, Inc, Richmond Virginia

[3]Department of Cellular and Molecular Medicine, Glycobiology Research and Training Center

[4]Department of Medicine, Division of Endocrinology and Metabolism, University of California San Diego, La Jolla, California

[5]Division of Cardiovascular Medicine, Sulpizio Cardiovascular Center, University of California San Diego, La Jolla CA

Abstract

Objective—Lipoprotein(a) [Lp(a)] levels are genetically determined by hepatocyte apolipoprotein(a) synthesis, but catabolic pathways also influence circulating levels. *APOE* genotypes have different affinities for the LDL receptor (LDLR) and LDL related protein-1 (LRP-1), with ε2 having the weakest binding to LDLR at <2% relative to ε3 and ε4.

Approach and Results—*APOE* genotypes (ε2/ε2, ε2/ε3, ε2/ε4, ε3/ε3, ε3/ε4 and ε4/ε4), Lp(a) mass, directly-measured Lp(a) cholesterol (Lp(a)-C) levels and a variety of apoB-related lipoproteins were measured in 431,239 patients. The prevalence of *APOE* traits were: ε2:7.35%, ε3:77.56%, and ε4:15.09%. Mean (SD) Lp(a) levels were 65% higher in ε4/ε4 compared to ε2/ε2 genotypes and increased significantly according to *APOE* genotype: ε2/ε2: 23.4(29.2), ε2/ε3: 31.3(38.0), ε2/ε4: 32.8(38.5), ε3/ε3: 33.2(39.1), ε3/ε4: 35.5(41.6), and ε4/ε4: 38.5(44.1) mg/dL (P<0.0001). LDL-C, apoB, Lp(a)-C, LDL-C corrected for Lp(a)-C content, LDL particle number and small dense LDL also had similar patterns. Patients with LDL-C ≥250mg/dL, who are more likely to have LDLR mutations and reduced affinity for apoB, had higher Lp(a) levels across all apoE isoforms, but particularly in patients with ε2 alleles, compared to LDL <250mg/dL. The lowest Lp(a) mass levels were present in patients with ε2 isoforms and lowest LDL-C.

Conclusions—*APOE* genotypes strongly influence Lp(a) and apoB-related lipoprotein levels. This suggests that differences in affinity of apoE proteins for lipoprotein clearance receptors may

Corresponding Author: Sotirios Tsimikas, MD, Vascular Medicine Program, Department of Medicine, Sulpizio Cardiovascular Center, University of California San Diego, 9500 Gilman Drive, BSB 1080, La Jolla, CA 92093-0682. stsimikas@ucsd.edu.

Disclosures: PMM is a speaker/consultant to Genzyme, Regeneron, Sanofi, Ionis, Amgen, Esperion, Aegerion, Kowa, Eliaz, Alexion, Lilly, Catabasis and Pfizer. SV and JPMcC are employees of Salveo Diagnostics. Dr. Gordts reports no conflicts. Dr. Tsimikas is a co-inventor and receives royalties from patents owned by the University of California San Diego and has a dual appointment at UCSD and Ionis Pharmaceuticals.

affect Lp(a) catabolism, suggesting a competition between Lp(a) and apoE protein for similar receptors.

Keywords

Lp(a); apoE; genotype; cardiovascular disease; prevalence; populations; guidelines

Introduction

Lipoprotein(a) [Lp(a)] is an independent and likely causal risk factor for cardiovascular disease (CVD) and calcific aortic valve stenosis (CAVS).[1-3] Lp(a) plasma levels are primarily (~90%) determined by variations in the *LPA* gene locus,[4] including the number of kringle IV_2 repeats and *LPA* single nucleotide polymorphisms. In addition, apolipoprotein(a) gene expression can be modulated by interlukin-6 that increases hepatocyte apolipoprotein(a) production, and by estrogen and bile acids that decrease production.[5, 6] Clearance mechanisms are not well defined but the major possibilities include the LDL receptor (LDLR), scavenger receptor B1,[7] plasminogen receptors and renal mechanisms.[8]

Apolipoprotein E (apoE) is polymorphic glycoprotein synthesized and secreted primarily by liver, brain, skin and macrophages.[9] ApoE is present on very low density lipoproteins (VLDL), remnant lipoproteins and HDL and facilitates their clearance via the LDL receptor (LDLR) and the LDL related protein-1 (LRP1) and syndecan-1 (SDC1).[10, 11] The 3 isoforms of *APOE*, ε2, ε3 and ε4, differ by single amino acid substitutions at two non-synonymous sites, which affect binding affinity for LDLR, LRP1 and SDC1. These variations are associated with differences in circulating VLDL-C, IDL-C and LDL-C levels, risk of CVD and the propensity to Alzheimer's disease.[12]

Although *APOE* genotypes have been associated with differences in lipoprotein levels, less is known about their role in influencing plasma Lp(a) levels.[13, 14] Previous studies reporting a relationship between Lp(a) and the apoE genotype are difficult to interpret as they suffer from low patient numbers in the less prevalent apoE genotypes. In particular controversy persists on whether the LDLR plays any significant role in Lp(a) clearance, and therefore if it influences plasma Lp(a) levels under physiological conditions.[8, 15] Furthermore, the relationship of *APOE* genotypes with Lp(a)-cholesterol [Lp(a)-C] and advanced measures of lipoproteins, such as plasma levels of apolipoprotein B-100, LDL particle number, small dense LDL and high-sensitivity C-reactive protein (hsCRP) has not been previously evaluated. In this study, we describe the relationship of *APOE* genotypes with Lp(a) levels and apoB-related lipoprotein measurements in a very large database from a referral laboratory.

Materials and Methods

Materials and Methods are available in the online-only Data Supplement Materials and Methods

Results

Baseline characteristics

The data is presented by APOE isoform genotype in Table 1. The prevalence of *APOE* traits were: ε2: 7.35%, ε3: 77.56%, and ε4: 15.09%. The prevalence of *APOE* genotypes were: ε2/ε2: 0.61%, ε2/ε3: 11.17%, ε2/ε4: 2.3%, ε3/ε3: 60.41%, ε3/ε4: 23.14% and ε4/ε4: 2.38%.. The mean age range was approximately 54-56, ~53% of patients were female and BMI was ~30.

Lipid and lipoprotein variables

Corresponding mean(SD) Lp(a) levels increased significantly according to *APOE* genotype ranging from 23.4 mg/dL for ε2/ε2 to 38.5(44.1) mg/dL for ε4/ε4 (P<0.0001 ANOVA) (Table 1 and Figure 1). Median Lp(a) levels also increased by genotype, from 11 mg/dL for ε2/ε2 to 20 mg/dL for ε4/ε4. Along with Lp(a) mass, Lp(a)-C also increased according to *APOE* genotype. Mean LDL-C also increased by *APOE* genotype ranging from 49.6 mg/dL for ε2/ε2 to 114.3 mg/dL for ε4/ε4 (P<0.0001 ANOVA). Corresponding mean apoB levels ranged from 56.6 for ε2/ε2 to 101.1 mg/dL for ε4/ε4 (p<0.0001 ANOVA). Striking increases in LDL particle number and small dense LDL were also noted according to genotype, with lowest values in ε2/ε2 to highest values in ε4/ε4.

In contrast, median triglyceride levels were inversely associated with *APOE* genotype from 140 mg/dL to 109 mg/dL for ε4/ε4 (p<0.0001 ANOVA). A similar but modest inverse association was noted with HDL-C (P<0.0001 ANOVA). Finally, hsCRP was also inversely associated with APOE genotype, with highest values in highest values in ε2/ε2 to lowest values in ε4/ε4 (Table 1).

For the patients with ε2/ε2 phenotype, a portion of which can have elevated triglycerides when a secondary stimulus accelerates VLDL production, additional analyses were performed. Out of 2445 patients with ε2/ε2 phenotype, there were 1205, 782 and 93 patients with triglyceride levels ≥150 mg/dL, ≥200 mg/dL and ≥500 mg/dL, respectively. Patients with triglycerides < vs. ≥150 mg/dL (Lp(a) mean (SD) 23.1 (28.5 vs. 23.7 (30.0), p=0.54), <200 vs. ≥200 mg/dL (Lp(a) mean (SD) 23.6 (29.5 vs. 22.8 (9.2), p=0.53), or <500 vs. ≥500 mg/dL (Lp(a) mean (SD) 23.4 (29.1 vs. 23.5 (30.9), p=0.98), did not have significantly different levels. Figure 1 shows a graphical representation of some of the key relationships.

Correlations among variables

Using a validated Lp(a)-C assay,[16] the Spearman correlation between Lp(a) mass and Lp(a)-C was r^2=0.79 (p<0.001) and between Lp(a) mass and LDL-C r^2=0.008 (p<0.001) (Figure 2). All other correlations of Lp(a) mass with variables in Table 1 had r <0.10, but due to the large numbers all were statistically significant with p<0.001 (data not shown). Lp(a)-C also had a weak correlation with LDL-C (r^2=0.012, p<0.001), total cholesterol (r^2=0.018, p<0.001), HDL-C r^2=0.0.38, p<0.001), and triglycerides (r^2=-0.016, p<0.001). All other correlations of Lp(a)-C with variables in table 1 had r <0.10, but due to the large numbers all were statistically significant with p<0.001 (data not shown).

Relationship Of Apoe Isoforms To Ldl-C ≥190 Mg/Dl And Ldl-C ≥250 Mg/Dl

Because LDLR mutation status was not available, we used LDL-C ≥190 mg/dL and LDL-C ≥250 mg/dL as potential surrogates for LDLR deficiency to derive insights into the potential role of the LDLR vis-à-vis APOE genotypes and Lp(a) catabolism. The analysis showed that compared to patients with LDL-C <190 mg/dL or <250 mg/dL, respectively, patients with LDL-C ≥190 mg/dL (mean (SD) Lp(a) 33.4 (39.4) mg/dL vs. 47.2 (50.9) mg/dL, p<0.001) and LDL ≥250 (mean (SD) Lp(a) 33.6 (39.6) mg/dL vs. 49.4 (53.1) mg/dL, p<0.001) had significantly higher Lp(a) levels.

When analyzed by *APOE* isoform status and LDL-C cutoffs, the lowest Lp(a) levels were present in ε2 genotypes with concomitant low LDL-C (Table 2). In contrast, ε2 genotypes with concomitant high LDL-C had the highest Lp(a) levels. The differences in Lp(a) levels according to *APOE* genotypes and normal or elevated LDL-C were greatest in ε2 genotypes and smallest in ε4 genotypes. For example, in individuals with ε2/ε3 genotype (there was only one ε2/ε2 individual with LDL ≥190, so this genotype could not be examined), Lp(a) levels were 31.2 (37.9) for LDL-C <190 mg/dL and 63.3 (57.5) for LDL ≥190 mg/dL (p<0.001). In contrast, for ε4/ε4 Lp(a) levels were 38.1 (43.8) mg/dL for LDL-C <190 mg/dL and 50.2 (52.1) for LDL ≥190 mg/dL (p<0.001).

Discussion

The current data document that *APOE* isoforms strongly influence Lp(a) mass levels, with a 65% increase in ε4/ε4 compared to ε2/ε2 genotypes. Consistent with this, Lp(a)-C levels, determined with an assay that strongly reflects Lp(a) mass,[16] also increased according to *APOE* genotypes. In addition, a variety of measures of lipoprotein particle number, including apoB levels, LDL particle number and small dense LDL had similar statistically significant associations. Finally, it was documented that concomitantly highly elevated LDL-C, a surrogate for LDLR deficiency, also influenced Lp(a) levels overall with higher levels in subjects with elevated LDL, as shown previously with a gene-dose relationship in subjects with homozygous and heterozygous familial hypercholesterolemia.[17, 18] This relationship was particularly accentuated in patients with ε2/ε2 genotypes that have the lowest affinity for the LDLR and LRP1. Because apoE proteins are not thought to directly affect hepatocyte synthesis of apolipoprotein(a) or impact Lp(a) assembly or secretion, it suggests that Lp(a) catabolism is influenced by apoE affinities for lipoprotein receptor clearance pathways such as LDLR and LRP1. These data further suggest a competition for such receptors by apoE and Lp(a) that affect circulating Lp(a) levels. It may also explain the weak to modest inverse correlation noted with Lp(a) and triglycerides in this study and several other studies, with higher Lp(a) levels being associated with lower triglyceride levels.[19, 20] Figure 3 represents a hypothetical construct of these relationships.

ApoE isoforms at the protein level are characterized by differences in 1 or 2 amino acids: ε3 has Cys-112 and Arg-158, ε4 has Arg-112 and Arg-158 and ε2 has Cys-112 and Cys-158. The LDLR binding activity for ε3 and ε4 is reported to be normal but ε2 has <2% of normal LDLR binding activity and is associated with recessive inheritance and low penetrance (10%).[12, 21, 22] The binding of apoE proteins to LRP1 has not been well-studies, but ligand blotting experiments suggest that lipid-bound apoE2 has 30-50% of normal LRP1 binding

activity.[23] The defective binding of apoE2 to the LDLR results in clinical expression of type III hyperlipoproteinemia in the presence of another factor (diabetes, obesity, hypothyroidism or estrogen deficiency) that overwhelms the capacity of apoE2 to mediate remnant lipoprotein clearance due to increases in VLDL production or reduced LDLR expression. These amino acid differences also result in different affinities for triglyceride-rich lipoproteins that lead to different effects on remodeling of VLDL to LDL and in receptor-mediated remnant clearance.[12, 24-30] The apoE4 protein is reported to prefer large, triglyceride-rich lipoproteins (VLDL and chylomicrons, while apoE3 and apoE2 proteins preferentially bind to small spherical HDL particles.[31, 32] In that regard, the apoE4 isoform is associated with the lowest triglyceride levels but higher LDL-C. It is presumed that enrichment of apoE4 on VLDL accelerates its clearance from the circulation by liver LDLR, LRP1 and SDC1 consequently downregulating hepatic LDLR expression. In addition, the enrichment of apoE4 on VLDL can outcompete LDL binding to LDLR due to the 20-fold greater affinity of apoE3 and apoE4 for LDLR compared to apoB100, further increasing circulating LDL-C.[9, 33]

Controversy exists whether the LDLR is involved in clearance of Lp(a) and data from cell culture, animal and human studies have given conflicting results. Initial studies revealed that the clearance rate of Lp(a) was similar when radiolabeled Lp(a) is injected in very small numbers of human with homozygous or heterozygous familial hypercholesterolemia or mice with or without intact LDLR receptors.[15, 34] Additionally, statins increase LDLR density but a seemingly paradoxical effect of statin-treatment on Lp(a) levels occurs and statins do not lower plasma Lp(a) levels. In fact, a recent analysis of 3896 patients on multiple statins as well as different doses of statins modestly raised mean Lp(a) levels by a mean of 11% and up to 50% in some studies.[35] These studies argue against a major role of the LDLR in Lp(a) clearance.

However, Lp(a) was shown to compete for LDL for binding to human LDLR in intact fibroblasts and that overexpression of the human LDLR in mice led to faster clearance of Lp(a).[36] Additionally, patients with familial hypercholesterolemia with abnormalities in the LDLR not only have higher LDL-C but also 1.5-2 times higher Lp(a) in a gene-dose relationship compared to their unaffected siblings.[17, 18] These studies argue for a role of the LDLR in clearance of Lp(a). More recent cell culture studies have also had conflicting results, with one study showing that Lp(a) catabolism in human hepatoma cell lines and primary fibroblasts is inhibited by PCSK9 via the LDLR, which mediated the effects of PCSK9 on Lp(a) internalization.[8] However, another study suggests the LDLR plays no role, but that PCSK9 may potentiate Lp(a) secretion, a pathway inhibited by PCSK9 antibodies.[37] The current data also favor that the LDLR is involved in clearing Lp(a), but that this is also influenced by the underlying *APOE* genotype that also competes for the same receptor. However, it is not possible to quantitate this effect, or the effect of non-LDLR pathways, such as plasminogen and SRB1 receptors.[7, 8]

The apolipoprotein(a) molecule is large with molecular mass ~200-900 KD and often larger than apolipoprotein B-100. It is covalently linked by a disulfide bond at cysteine number 4326 on apoB which is near the apoB docking site that binds to the LDLR. Thus, the apolipoprotein(a) of Lp(a) may cause the apoB of Lp(a) to be sterically hindered in its

interaction with the LDLR and thus slow Lp(a)'s clearance. This is supported by the fact that Lp(a) has slower clearance rate than LDL, approximately a day longer with a fractional catabolic rate (FCR) of 0.22 pools/day versus FCR of 0.37 pools/day for LDL,[38, 39] and that when apolipoprotein(a) synthesis is inhibited by antisense oligonucleotides (ASO), LDL-C is significantly reduced. The effect of the apolipoprotein(a) ASO on LDL is independent of effects on apoB synthesis and LDL- production,[40, 41] which is in agreement with the different metabolic production routes for Lp(a) and LDL production.[42]

This study also demonstrates that LDL-C levels and apolipoprotein B-100 variables distribute in the same direction as Lp(a) mass and Lp(a)-C. Despite this, there is almost negligible correlation between Lp(a) and LDL-C or apoB due to different genetic influences in production of each. Despite the fact that Lp(a), unlike LDL, is not derived from a VLDL precursor the mechanisms associated with elevation of each may be similar. One may postulate that when apoE is present on larger chylomicron remnants and VLDL particles, it competes for binding to LDLR and LRP1, thereby slowing their clearance and leading to higher LDL-C and Lp(a) levels. When there is relatively high affinity for these receptors, such as apoE4(and apoE3, it leads to higher LDL-C, apoB and Lp(a). In contrast, when there is lower affinity, such as with apoE2, LDL and Lp(a) particles can clear faster. In addition, the slower conversion of VLDL to LDL with ε2 genotype, leads to lower LDL levels, which further enhances Lp(a) clearance due to less LDL competition. The additional increment in circulating Lp(a) levels in ε4/ε4 patients is likely explained by the fact the ε4 partitions preferentially on triglyceride-rich lipoproteins and thus generates a bigger mass of lipoproteins to outcompete Lp(a) binding.

It is also possible that LRP1 can be responsible for the observed apoE-Lp(a) correlation. The apoE2 isoform has negligible binding to LDLR (< 2% compared to ε3). If LDLR was responsible for our observed association one could expect to see a reduction in plasma Lp(a) and Lp(a)-cholesterol levels between ε3/ε3 and ε2/ε2 patients greater than the observed 27% and 14%. In contrast LDL-C, LDL-P and apoB levels, both cleared by LDLR, are 50-55% lower in ε2/ε2 patients. Binding of the ε2 isoform to LRP1 is only reduced by 50-70% and can possibly explain the less drastic reduction in Lp(a) levels in ε2/ε2 patients compared to LDL and apoB. The underlying mechanisms of elevation of Lp(a) in different APOE isoforms are evidently more complex and whether LDLR and LRP1 receptors pay a role in this relationship needs to be determined in experimental studies.

Limitations of this study are the lack of data on race and lipid-modifying therapies. This study provides a rationale to study the role of apoE in explaining differences in Lp(a) and in therapeutic interventions such as with PCSK9 inhibitors and antisense oligonucleotides to Lp(a) lowering.[40, 43]

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

none.

Funding source: Dr. Gordts is supported by the AHA grant 15BGIA25550111.

Dr Tsimikas is supported by NIH grants HL119828, HL055798, HL088093, HL106579, HL078610, and HL124174.

References

1. Kronenberg F. Human genetics and the causal role of Lipoprotein(a) for various diseases. Cardiovasc Drugs Ther. 2016; 30:87–100. [PubMed: 26896185]
2. Thanassoulis G. Lipoprotein (a) in calcific aortic valve disease: from genomics to novel drug target for aortic stenosis. J Lipid Res. 2016; 57:917–924. [PubMed: 26685327]
3. Capoulade R, Chan KL, Yeang C, Mathieu P, Bosse Y, Dumesnil JG, Tam JW, Teo KK, Mahmut A, Yang X, Witztum JL, Arsenault BJ, Despres JP, Pibarot P, Tsimikas S. Oxidized phospholipids, lipoprotein(a), and progression of calcific aortic valve stenosis. J Am Coll Cardiol. 2015; 66:1236–1246. [PubMed: 26361154]
4. Schmidt K, Noureen A, Kronenberg F, Utermann G. Structure, function, and genetics of lipoprotein (a). J Lipid Res. 2016; 57:1339–1359. [PubMed: 27074913]
5. Hoover-Plow J, Huang M. Lipoprotein(a) metabolism: potential sites for therapeutic targets. Metabolism: Clin Exper. 2013; 62:479–491.
6. Chennamsetty I, Claudel T, Kostner KM, Baghdasaryan A, Kratky D, Levak-Frank S, Frank S, Gonzalez FJ, Trauner M, Kostner GM. Farnesoid X receptor represses hepatic human APOA gene expression. J Clin Invest. 2011; 121:3724–3734. [PubMed: 21804189]
7. Yang XP, Amar MJ, Vaisman B, Bocharov AV, Vishnyakova TG, Freeman LA, Kurlander RJ, Patterson AP, Becker LC, Remaley AT. Scavenger receptor-BI is a receptor for lipoprotein(a). J Lipid Res. 2013; 54:2450–2457. [PubMed: 23812625]
8. Romagnuolo R, Scipione CA, Boffa MB, Marcovina SM, Seidah NG, Koschinsky ML. Lipoprotein(a) catabolism is regulated by proprotein convertase subtilisin/kexin type 9 through the low density lipoprotein receptor. J Biol Chem. 2015; 290:11649–11662. [PubMed: 25778403]
9. Mahley RW. Apolipoprotein E: from cardiovascular disease to neurodegenerative disorders. J Mol Med (Berl). 2016; 94:739–746. [PubMed: 27277824]
10. Li J, Kanekiyo T, Shinohara M, Zhang Y, LaDu MJ, Xu H, Bu G. Differential regulation of amyloid-beta endocytic trafficking and lysosomal degradation by apolipoprotein E isoforms. J Biol Chem. 2012; 287:44593–44601. [PubMed: 23132858]
11. Gonzales JC, Gordts PL, Foley EM, Esko JD. Apolipoproteins E and AV mediate lipoprotein clearance by hepatic proteoglycans. J Clin Invest. 2013; 123:2742–2751. [PubMed: 23676495]
12. Huang Y. Mechanisms linking apolipoprotein E isoforms with cardiovascular and neurological diseases. Curr Opin Lipidol. 2010; 21:337–345. [PubMed: 20531185]
13. de Knijff P, Kaptein A, Boomsma D, Princen HM, Frants RR, Havekes LM. Apolipoprotein E polymorphism affects plasma levels of lipoprotein(a). Atherosclerosis. 1991; 90:169–174. [PubMed: 1836947]
14. Horita K, Eto M, Saito M, Nakata H, Iwashima Y, Ito H, Takahashi M, Kondo A, Morikawa A, Makino I. Effects of apolipoprotein E polymorphism on plasma lipoprotein(a) levels. Artery. 1993; 20:324–336. [PubMed: 8010888]
15. Rader DJ, Mann WA, Cain W, Kraft HG, Usher D, Zech LA, Hoeg JM, Davignon J, Lupien P, Grossman M, Wilson JM, Brewer HB. The low density lipoprotein receptor is not required for normal catabolism of Lp(a) in humans. J Clin Invest. 1995; 95:1403–1408. [PubMed: 7883987]
16. Baudhuin LM, Hartman SJ, O'Brien JF, Meissner I, Galen RS, Ward JN, Hogen SM, Branum EL, McConnell JP. Electrophoretic measurement of lipoprotein(a) cholesterol in plasma with and without ultracentrifugation: comparison with an immunoturbidimetric lipoprotein(a) method. Clin Biochem. 2004; 37:481–488. [PubMed: 15183296]
17. Kraft HG, Lingenhel A, Raal FJ, Hohenegger M, Utermann G. Lipoprotein(a) in homozygous familial hypercholesterolemia. Arterioscler Thromb Vasc Biol. 2000; 20:522–528. [PubMed: 10669652]

18. Alonso R, Andres E, Mata N, Fuentes-Jimenez F, Badimon L, Lopez-Miranda J, Padro T, Muniz O, Diaz-Diaz JL, Mauri M, Ordovas JM, Mata P, Investigators S. Lipoprotein(a) levels in familial hypercholesterolemia: an important predictor of cardiovascular disease independent of the type of LDL receptor mutation. J Am Coll Cardiol. 2014; 63:1982–1989. [PubMed: 24632281]
19. Tsimikas S, Witztum JL, Miller ER, Sasiela WJ, Szarek M, Olsson AG, Schwartz GG. High-dose atorvastatin reduces total plasma levels of oxidized phospholipids and immune complexes present on apolipoprotein B-100 in patients with acute coronary syndromes in the MIRACL trial. Circulation. 2004; 110:1406–1412. [PubMed: 15353498]
20. Tsimikas S, Clopton P, Brilakis ES, Marcovina SM, Khera A, Miller ER, de Lemos JA, Witztum JL. Relationship of oxidized phospholipids on apolipoprotein B-100 particles to race/ethnicity, apolipoprotein(a) isoform size, and cardiovascular risk factors: Results from the Dallas Heart Study. Circulation. 2009; 119:1711–1719. [PubMed: 19307470]
21. Toops KA, Tan LX, Lakkaraju A. Apolipoprotein E isoforms and AMD. Adv Exp Med Biol. 2016; 854:3–9. [PubMed: 26427386]
22. Mahley RW. Apolipoprotein E: cholesterol transport protein with expanding role in cell biology. Science. 1988; 240:622–630. [PubMed: 3283935]
23. Kowal RC, Herz J, Weisgraber KH, Mahley RW, Brown MS, Goldstein JL. Opposing effects of apolipoproteins E and C on lipoprotein binding to low density lipoprotein receptor-related protein. J Biol Chem. 1990; 265:10771–10779. [PubMed: 2355022]
24. Wu G, Yuan J, Hunninghake DB. Effect of human apolipoprotein E isoforms on plasma lipids, lipoproteins and apolipoproteins in apolipoprotein E-deficient mice. Atherosclerosis. 1998; 141:287–296. [PubMed: 9862177]
25. Phillips MC. Apolipoprotein E isoforms and lipoprotein metabolism. IUBMB Life. 2014; 66:616–623. [PubMed: 25328986]
26. Knouff C, Hinsdale ME, Mezdour H, Altenburg MK, Watanabe M, Quarfordt SH, Sullivan PM, Maeda N. Apo E structure determines VLDL clearance and atherosclerosis risk in mice. J Clin Invest. 1999; 103:1579–1586. [PubMed: 10359567]
27. Mamotte CD, Sturm M, Foo JI, van Bockxmeer FM, Taylor RR. Comparison of the LDL-receptor binding of VLDL and LDL from apoE4 and apoE3 homozygotes. Am J Physiol. 1999; 276:E553–557. [PubMed: 10070023]
28. Martins IJ, Hone E, Chi C, Seydel U, Martins RN, Redgrave TG. Relative roles of LDLr and LRP in the metabolism of chylomicron remnants in genetically manipulated mice. J Lipid Res. 2000; 41:205–213. [PubMed: 10681404]
29. van Dijk KW, van Vlijmen BJ, de Winther MP, van 't Hof B, van der Zee A, van der Boom H, Havekes LM, Hofker MH. Hyperlipidemia of ApoE2(Arg(158)-Cys) and ApoE3-Leiden transgenic mice is modulated predominantly by LDL receptor expression. Arterioscler Thromb Vasc Biol. 1999; 19:2945–2951. [PubMed: 10591674]
30. Mahley RW, Huang Y. Atherogenic remnant lipoproteins: role for proteoglycans in trapping, transferring, and internalizing. J Clin Invest. 2007; 117:94–98. [PubMed: 17200713]
31. Steinmetz A, Jakobs C, Motzny S, Kaffarnik H. Differential distribution of apolipoprotein E isoforms in human plasma lipoproteins. Arteriosclerosis. 1989; 9:405–411. [PubMed: 2719600]
32. Weisgraber KH. Apolipoprotein E distribution among human plasma lipoproteins: role of the cysteine-arginine interchange at residue 112. J Lipid Res. 1990; 31:1503–1511. [PubMed: 2280190]
33. Pitas RE, Innerarity TL, Arnold KS, Mahley RW. Rate and equilibrium constants for binding of apo-E HDLc (a cholesterol-induced lipoprotein) and low density lipoproteins to human fibroblasts: evidence for multiple receptor binding of apo-E HDLc. Proc Natl Acad Sci U S A. 1979; 76:2311–2315. [PubMed: 221921]
34. Cain WJ, Millar JS, Himebauch AS, Tietge UJF, Maugeais C, Usher D, Rader DJ. Lipoprotein [a] is cleared from the plasma primarily by the liver in a process mediated by apolipoprotein [a]. J Lipid Res. 2005; 46:2681–2691. [PubMed: 16150825]
35. Yeang C, Hung MY, Byun YS, Clopton P, Yang X, Witztum JL, Tsimikas S. Effect of therapeutic interventions on oxidized phospholipids on apolipoprotein B100 and lipoprotein(a). J Clin Lipidol. 2016; 10:594–603. [PubMed: 27206947]

36. Hofmann SL, Eaton DL, Brown MS, McConathy WJ, Goldstein JL, Hammer RE. Overexpression of human low density lipoprotein receptors leads to accelerated catabolism of Lp(a) lipoprotein in transgenic mice. J Clin Invest. 1990; 85:1542–1547. [PubMed: 2139667]

37. Villard EF, Thedrez A, Blankenstein J, Croyal M, Tran TTT, Poirier B, Le Bail JC, Illiano S, Nobécourt E, Krempf M, Blom DJ, Marais AD, Janiak P, Muslin AJ, Guillot E, Lambert G. PCSK9 Modulates the Secretion But Not the Cellular Uptake of Lipoprotein(a) Ex Vivo. JACC: Basic to Translational Science. 2016; 1:419.

38. Jenner JL, Seman LJ, Millar JS, Lamon-Fava S, Welty FK, Dolnikowski GG, Marcovina SM, Lichtenstein AH, Barrett PH, deLuca C, Schaefer EJ. The metabolism of apolipoproteins (a) and B-100 within plasma lipoprotein (a) in human beings. Metabolism. 2005; 54:361–369. [PubMed: 15736114]

39. Kesaniemi YA, Witztum JL, Steinbrecher UP. Receptor-mediated catabolism of low density lipoprotein in man. Quantitation using glucosylated low density lipoprotein. J Clin Invest. 1983; 71:950–959. [PubMed: 6300194]

40. Viney NJ, van Capelleveen JC, Geary RS, Xia S, Tami JA, Yu RZ, Marcovina SM, Hughes SG, Graham MJ, Crooke RM, Crooke ST, Witztum JL, Stroes ES, Tsimikas S. Antisense oligonucleotides targeting apolipoprotein(a) in people with raised lipoprotein(a): two randomised, double-blind, placebo-controlled, dose-ranging trials. Lancet. 2016; 388:2239–2253. [PubMed: 27665230]

41. Graham MJ, Viney N, Crooke RM, Tsimikas S. Antisense inhibition of apolipoprotein (a) to lower plasma lipoprotein (a) levels in humans. J Lipid Res. 2016; 57:340–351. [PubMed: 26538546]

42. Frischmann ME, Ikewaki K, Trenkwalder E, Lamina C, Dieplinger B, Soufi M, Schweer H, Schaefer JR, Konig P, Kronenberg F, Dieplinger H. In vivo stable-isotope kinetic study suggests intracellular assembly of lipoprotein(a). Atherosclerosis. 2012; 225:322–327. [PubMed: 23099120]

43. Tsimikas S, Viney NJ, Hughes SG, Singleton W, Graham MJ, Baker BF, Burkey JL, Yang Q, Marcovina SM, Geary RS, Crooke RM, Witztum JL. Antisense therapy targeting apolipoprotein(a): a randomised, double-blind, placebo-controlled phase 1 study. Lancet. 2015; 386:1472–1483. [PubMed: 26210642]

Abbreviations

LDLR	LDL receptor
LRP-1	LDL related protein-1
CVD	cardiovascular disease
CAVS	calcific aortic valve stenosis
ApoE	Apolipoprotein E
SDC1	syndecan-1
hsCRP	high-sensitivity C-reactive protein

Highlights

- Lp(a) levels were 65% higher in ε4/ε4 compared to ε2/ε2 genotypes.
- LDL-C, apoB, directly measured Lp(a)-C, LDL-C corrected for Lp(a)-C content, LDL particle number and small dense LDL also had similar patterns.
- Triglyceride and hsCRP levels were highest in ε2/ε2 and lowest in ε4/ε4 genotype.
- These data suggest that circulating Lp(a) levels are not only genetically determined by hepatocyte apolipoprotein(a) synthesis, but can also be influenced by the *APOE* genotype possibly due to apoE isoform differences in lipoprotein clearance pathways.

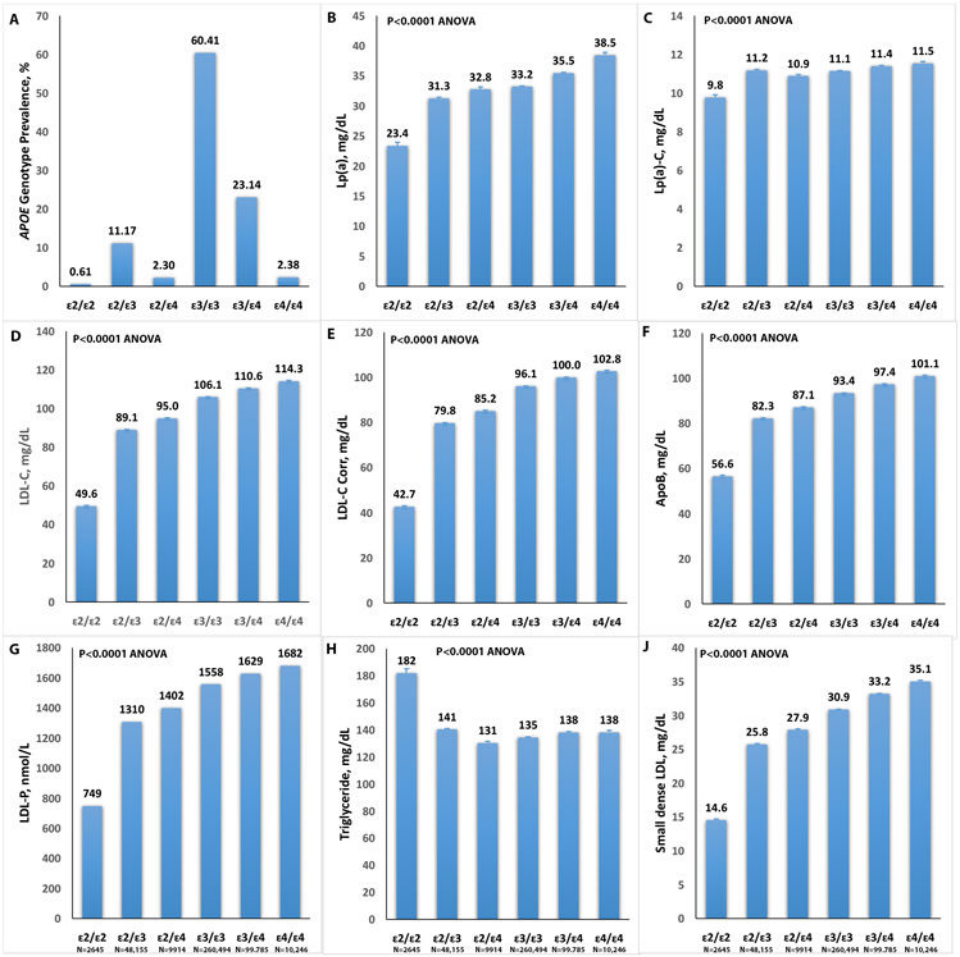

Figure 1.

Relationship of *APOE* genotypes (prevalence shown in panel A) to Lp(a) mass (B), Lp(a)-C (C), LDL-C (D), LDL-C corr (E), apoB (F), LDL-particle number (LDL-P) (G), triglycerides (H) and small dense LDL (J).

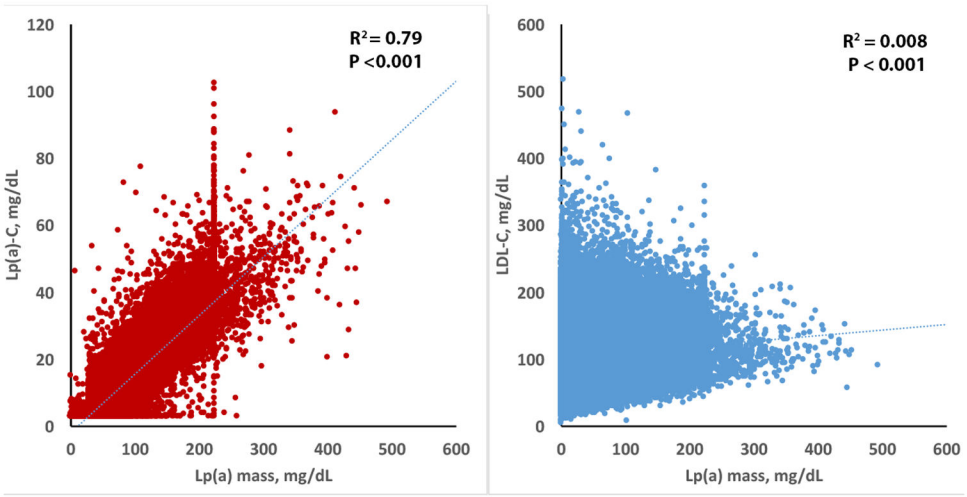

Figure 2.
Relationship of Lp(a) mass to Lp(a)-C and LDL-C.

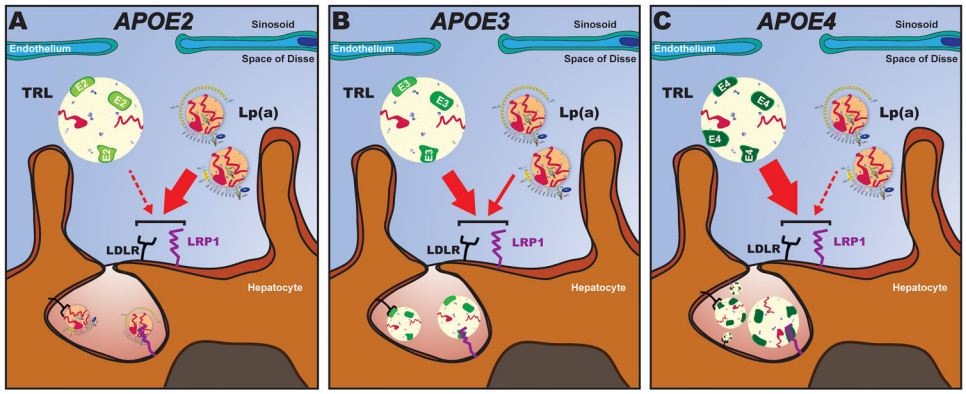

Figure 3.

Conceptual rendition of the potential role of apoE, LDLR, LRP1 in the catabolsim of Lp(a). ApoE on triglyceride-rich lipoproteins (TRLs) compete with Lp(a) for binding to LDLR and LRP1 on hepatocytes. (A) ApoE2 is the lower affinity isoform for LDLR and LRP1, which allow LDL and Lp(a) particles to clear faster. (B-C) Relatively high affinity isoforms, such as apoE4 and apoE3 favor hepatic clearance of TRLs via LDLR and LRP1 over LDL and Lp(a). This results in elevated LDL-C, apoB and Lp(a) levels. (C) ApoE4 partitions preferentially on TRLs and thus generates a bigger mass of lipoproteins to outcompete Lp(a) binding.

Table 1

Baseline variables expressed by APOE isoform status

Variable	ε2/ε2	ε2/ε3	ε2/ε4	ε3/ε3	ε3/ε4	ε4/ε4	P-trend
ApoE genotype, Number	2,645	48,155	9,914	262,337	100,388	10,307	
ApoE genotype, %	0.61	11.2	2.3	60.8	23.3	2.4	<0.001
Age, years	55.4 (15.1)	56.1 (15.0)	55.4 (14.6)	55.6 (15.0)	55.0 (14.7)	54.0 (14.3)	<0.001
Sex, % Female	53.0	52.0	52.3	52.1	52.7	52.9	<0.001
BMI	30.1 (6.9)	29.9 (6.8)	30.1 (7.2)	29.6 (6.7)	29.7 (6.8)	29.8 (7.0)	<0.001
Lp(a) mass, mean, mg/dL	23.4 (29.2)	31.3 (38.0)	32.8 (38.5)	33.2 (39.1)	35.5 (41.6)	38.5 (44.1)	<0.001
Lp(a) mass, median, mg/dL	11 (5-30)	15 (6-43)	16 (7-46)	17 (7-46)	18 (7-50)	20 (8-56)	<0.001
Lp(a)-C, mean, mg/dL	9.8 (7.7)	11.2 (8.0)	10.9 (7.6)	11.1 (8.1)	11.4 (8.4)	11.5 (8.8)	<0.001
Lp(a)-C, median, mg/dL	7.7 (5.1-12.0)	9.1 (5.5-14.5)	8.8 (5.3-14.5)	9.0 (5.2-14.7)	9.1 (5.3-15.0)	9.0 (5.1-15.4)	<0.001
TC, mg/dL	176.5 (62.6)	177.4 (1.5)	181.9 (42.8)	187.5 (43.0)	191.6 (44.4)	195.8 (46.1)	<0.001
LDL-C, mg/dl	49.6 (18.5)	89.1 (28.8)	95.0 (30.5)	106.1 (33.6)	110.6 (35.2)	114.3 (36.8)	<0.001
LDL-C, corr, mg/dL	42.7 (18.4)	79.8 (29.7)	85.2 (31.4)	96.1 (34.5)	100.0 (36.2)	102.8 (37.8)	<0.001
ApoB, mg/dL	56.6 (19.2)	82.3 (23.5)	87.1 (24.7)	93.4 (25.6)	97.45 (26.8)	101.1 (27.7)	<0.001
LDL particle #, nmol/L	749 (361)	1309 (476)	1402 (507)	1558 (537)	1629 (560)	1682 (578)	<0.001
Small dense LDL, mg/dL	14.6 (7.3)	25.8 (12.1)	27.9 (12.8)	30.9 (14.3)	33.2 (15.1)	35.1 (15.6)	<0.001
non-HDL-C, mg/dL	122.3 (64.3)	122.9 (39.5)	127.8 (40.6)	133.7 (39.8)	138.5 (41.2)	143.1 (42.6)	<0.001
HDL-C, mg/dL	54.2 (16.8)	54.6 (16.4)	54.1 (16.5)	52.9 (15.6)	53.1 (15.4)	53.8 (15.5)	<0.001
ApoA-I	152.1 (31.6)	149.6 (30.4)	147.4 (30.8)	146.8 (29.8)	144.5 (29.6)	143.5 (29.6)	<0.001
TG, mg/dL	140 (96-218)	115 (80-167)	112 (79-166)	109 (77-155)	109 (77-159)	109 (77-161)	<0.001
hsCRP, mg/L	2.3 (1.1-5.1)	2.2 (1.0-5.1)	1.8 (0.8-4.3)	2.0 (0.9-4.6)	1.5 (0.67-3.6)	1.1 (0.5-2.7)	<0.001

The data is provided as mean (SD) or median (IQR). LDL-C corr is LDL-C corrected for Lp(a)-C content and is derived as LDL-C minus (Lp(a) mass*0.3)

Table 2

Baseline variables expressed by *APOE* isoform status and LDL-C cutoffs of < or ≥190 mg/dL and < or ≥250 mg/dL

Variable	ε2/ε2	ε2/ε2		ε2/ε2	ε2/ε2	
	LDL-C <190	LDL-C ≥190	p-value	LDL-C <250	LDL-C ≥250	p-value
Number	2644	1		2645	0	
Lp(a) mass, mean, mg/dL	23.4 (29.2)	3.0	0.49	23.4 (29.2)	-	-
Lp(a) mass, median, mg/dL	11.0 (5.0-30.0)		0.49	11.0 (5.0-30.0)	-	-
Lp(a)-C*, mg/dL	7.7 (5.1-12.0)	-	-	7.7 (5.1-12.0)	-	-
LDL-C, mg/dl	49.5 (18.1)	244	<0.001	49.5 (18.1)	-	-
LDL-Ccorr, mg/dl	42.7 (18.1)	243	<0.001	42.7 (18.1)	-	-
non-HDL-C, mg/dL	122.3 (64.3)	228	0.10	122.3 (64.3)	-	-
ApoB, mg/dL	56.6 (54.0)	199	<0.001	56.6 (54.0)	-	-
TG, mg/dL	140 (96-218)	108	0.65	140 (96-218)	-	-
	ε2/ε3	ε2/ε3		ε2/ε3	ε2/ε3	
	LDL-C <190	LDL-C ≥190	p-value	LDL-C <250	LDL-C ≥250	p-value
Number	48,011	144		48,138	17	
Lp(a) mass, mean, mg/dL	31.2 (37.9)	63.3 (57.5)	<0.001	31.3 (38.0)	67.3 (66.4)	<0.001
Lp(a) mass, median, mg/dL	15.0 (6.0-43.0)	46.0 (16.0-99.8)	<0.001	15.0 (6.0-43.0)	44.0 (13.5-109.5)	<0.001
Lp(a)-C*, mg/dL	9.1 (5.5-14.4)	15.2 (7.5-23.2)	<0.001	9.1 (5.5-14.5)	15.7 (10.1-30.3)	<0.001
LDL-C, mg/dl	88.7 (27.9)	217.5 (32.9)	<0.001	89.0 (28.5)	282.2 (53.1)	<0.001
LDL-Ccorr, mg/dl	79.4 (28.8)	198.5 (35.9)	<0.001	79.7 (29.4)	2612.0 (50.7)	<0.001
non-HDL-C, mg/dL	122.4 (38.6)	269.3 (51.5)	<0.001	122.8 (39.3)	342.9 (67.0)	<0.001
ApoB, mg/dL	82.1 (22.9)	173.9 (27.8)	<0.001	82.3 (23.3)	215.1 (29.9)	<0.001
TG, mg/dL	115 (80-167)	168 (109-235)	<0.001	115 (80-167)	162.5 (89.3)	0.45
	ε2/ε4	ε2/ε4		ε2/ε4	ε2/ε4	
	LDL-C <190	LDL-C ≥190	p-value	LDL-C <250	LDL-C ≥250	
Number	9,853	62		9906	8	
Lp(a) mass, mean, mg/dL	32.7 (38.4)	51.8 (51.8)	<0.001	32.7 (38.5)	77.0 (61.0)	0.001
Lp(a) mass, median, mg/dL	16.0 (7.0-46.0)	26.0 (15.3-86.3)	<0.001	16.0 (7.0-46.0)	91 (18-101.5)	0.30

Variable	**ε2/ε2**	**ε2/ε2**		**ε2/ε2**	**ε2/ε2**	
Lp(a)-C, mg/dL	8.8 (5.3-14.5)	13.6 (7.4-20.3)	0.002	8.8 (5.3-14.5)	14.5 (11.0-19.0)	0.002
LDL-C, mg/dl	94.2 (29.0)	215.4 (27.0)	<0.001	94.8 (30.1)	272.4 (22.1)	<0.001
LDL-Ccorr, mg/dl	84.5 (30.1)	199.8 (27.0)	<0.001	85.0 (31.1)	249.3 (16.7)	<0.001
non-HDL-C, mg/dL	127.0 (39.2)	259.1 (44.0)	<0.001	127.7 (40.1)	336.8 (34.5)	0.32
ApoB, mg/dL	86.6 (23.8)	168.5 (23.2)	<0.001	87.0 (24.4)	211.9 (14.8)	<0.001
TG, mg/dL	112 (79-166)	154 (105-217)	0.025	112 (79-166)	169 (124-229)	0.025
	ε3/ε3	ε3/ε3		ε3/ε3	ε3/ε3	
	LDL-C <190	LDL-C ≥190	p-value	LDL-C <250	LDL-C ≥250	p-value
Number	256,822	3,672		260,253	241	
Lp(a) mass, mean, mg/dL	33.1 (38.8)	46.0 (50.4)	<0.001	33.2 (39.0)	47.7 (52.4)	<0.001
Lp(a) mass, median, mg/dL	17.0 (7.0-46.0)	25.0 (10.0-71.0)	<0.001	17.0 (7.0-46.0)	26.0 (11.0-70.5)	<0.001
Lp(a)-C*, mg/dL	9.0 (5.2-14.6)	12.2 (6.7-19.7)	<0.001	9.0 (5.2-14.7)	13.3 (6.8-22.7)	<0.001
LDL-C, mg/dl	104.6 (31.1)	212.3 (25.4)	<0.001	105.9 (33.1)	284.7 (37.9)	<0.001
LDL-Ccorr, mg/dl	94.7 (32.3)	198.4 (28.9)	<0.001	95.9 (34.1)	269.4 (38.7)	<0.001
non-HDL-C, mg/dL	132.1 (37.3)	247.2 (41.0)	<0.001	133.5 (39.3)	328.2 (78.0)	<0.001
ApoB, mg/dL	92.4 (24.2)	166.7 (25.2)	<0.001	93.3 (25.5)	215.3 (34.4)	<0.001
TG, mg/dL	108 (77-154)	159 (117-217)	<0.001	108 (77-155)	168 (117-235)	<0.001
	ε3/ε4	ε3/ε4		ε3/ε4	ε3/ε4	p-value
Number	LDL-C <190	LDL-C ≥190	p-value	LDL-C <250	LDL-C ≥250	
Lp(a) mass, mean, mg/dL	97,621	2,164		99.641	144	
Lp(a) mass, median, mg/dL	35.2 (41.4)	47.6 (50.9)	<0.001	35.4 (41.6)	47.7 (9.0-80.8)	<0.001
Lp(a)-C, mg/dL	17.0 (7.0-50.0)	26.0 (10.0-73.0)	<0.001	18.0 (7.0-50.0)	26.0 (9.0-80.8)	<0.001
LDL-C, mg/dl	9.1 (5.3-14.7)	11.9 (6.7-19.4)	<0.001	9.1 (5.3-14.9)	15.3 (9.5-24.5)	<0.001
LDL-Ccorr, mg/dl	108.3 (31.8)	212.9 (26.9)	<0.001	110.3 (34.5)	288.8 (42.0)	<0.001
non-HDL-C, mg/dL	97.8 (33.2)	198.6 (30.6)	<0.001	99.8 (35.6)	274.5 (46.5)	<0.001
ApoB, mg/dL	136.2 (38.0)	245.0 (38.7)	<0.001	138.3 (40.6)	325.8 (61.6)	<0.001
TG, mg/dL	95.9 (24.7)	166.9 (25.0)	<0.001	97.3 (26.4)	215.8 (34.1)	<0.001
	108 (77-157)	153 (112-209)	<0.001	109 (77-159)	163.5 (117-218)	<0.001
	ε4/ε4	ε4/ε4		ε4/ε4	ε4/ε4	

Variable	ε2/ε2	ε2/ε2		ε2/ε2	ε2/ε2	
	LDL-C <190	LDL-C ≥190	p-value	LDL-C <250	LDL-C ≥250	p-value
Number	9,926	320		10,225	21	
Lp(a) mass, mean, mg/dL	38.1 (43.8)	50.2 (52.1)	<0.001	38.4 (44.1)	54.8 (71.6)	0.089
Lp(a) mass, median, mg/dL	20.0 (8.0-55.0)	27.0 (11.0-85.3)	<0.001	20.0 (8.0-56.0)	21.0 (7.0-92.0)	0.089
Lp(a)-C, mg/dL	8.9 (5.1-15.2)	12.1 (6.0-85.3)	<0.001	9.0 (5.1-15.4)	15.9 (4.5-23.4)	0.10
LDL-C, mg/dl	111.0 (32.3)	214.5 (24.4)	<0.001	113.9 (36.0)	280.2 (30.9)	<0.001
LDL-Ccorr, mg/dl	99.7 (33.7)	199.4 (29.5)	<0.001	103.5 (37.1)	263.8 (42.8)	<0.001
non-HDL-C, mg/dL	139.8 (38.7)	244.4 (168.8)	<0.001	142.7 (41.9)	312.8 (35.6)	<0.001
ApoB, mg/dL	98.9 (24.9)	168.8 (24.9)	<0.001	100.9 (27.2)	212.2 (29.4)	<0.001
TG, mg/dL	108 (76-159)	143 (99-199)	0.001	109 (77-161)	121 (113-178)	0.41

The data is presented as mean (SD) or median (IQR).
For Lp(a)-C there were 667 patients in e2/e2, 84 in e2/e3, 62 in e2/e4, 86,853 in e3/e3, 34,995 in e3/e4, and 142 in e4/e4 groups.