Sex-Specific Associations of Genetically-Predicted Circulating Lipoprotein(a) and Hepatic *LPA* Gene Expression Levels with Cardiovascular Outcomes: Mendelian Randomization and Observational Analyses

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Abstract:

Background - Elevated Lipoprotein(a) (Lp[a]) levels are associated with coronary artery disease (CAD), ischemic stroke (IS) and calcific aortic valve stenosis (CAVS). Studies investigating the association between Lp(a) levels and these diseases in women have yielded inconsistent results.
Methods - To investigate the association of Lp(a) with sex-specific cardiovascular outcomes, we determined the association between genetically-predicted Lp(a) levels (using 27 single nucleotide polymorphisms (SNPs) at the *LPA* locus) and hepatic *LPA* expression (using 80 SNPs at the *LPA* locus associated with *LPA* mRNA expression in liver samples from the Genotype-Tissue Expression dataset) on CAD, IS and CAVS using individual participant data from the UK Biobank: 408,403 participants of European ancestry (37,102, 4283 and 2574 with prevalent CAD, IS and CAVS, respectively). The long-term association between Lp(a) levels and incident CAD, IS and CAVS in was also investigated in EPIC-Norfolk: 18,721 participants (3964, 846 and 424 with incident CAD, IS and CAVS, respectively).
Results - Genetically-predicted and plasma Lp(a) levels were positively and similarly associated with prevalent and incident CAD and CAVS in men and women. Genetically-predicted and plasma Lp(a) levels was associated with prevalent and incident IS when we studied men and women pooled together, and in men only. Genetically-predicted *LPA* expression levels was associated with prevalent CAD and CAVS in men and women, but not with IS.
Conclusions - Genetically-predicted blood Lp(a) and hepatic *LPA* gene expression as well as serum Lp(a) levels predict the risk of CAD and CAVS in men and in women. Whether RNA interference therapies aiming at lowering Lp(a) levels could be useful in reducing cardiovascular disease risk in both men and women with high Lp(a) levels needs to be determined in large-scale cardiovascular outcomes trials.

Key words: lipoprotein; Mendelian randomization; coronary artery disease; aortic valve stenosis; stroke

Nonstandard abbreviations and acronyms:

CAD: Coronary Artery Disease

CAVS: Calcific Aortic Valve Stenosis

CVD: Cardiovascular Diseases

EPIC: European Prospective Investigation into Cancer and Nutrition

GTEx: Genotype Tissue Expression

HR: Hazard ratio

ICD10: International Classification of Diseases version-10

IS: Ischemic Stroke

LD: linkage disequilibrium

LDL: Low-density Lipoprotein

Lp(a): Lipoprotein(a)

MAF: Minor Allele Frequency

MR: Mendelian Randomization

OR: Odds ratio

OPCS: Office of Population Censuses and Surveys Classification

OxPL: Oxidized Phospholipids

SNP: Single Nucleotide Polymorphism

wGRS: Weighted Genetic Risk Score

Introduction

Plasma levels of lipoprotein(a) [Lp(a)] are associated with a higher risk of several cardiovascular diseases (CVD) such as coronary artery disease (CAD), ischemic stroke (IS) and calcific aortic valve stenosis (CAVS) as well as all-cause mortality.[1-9] Lp(a)-lowering therapies are currently being developed and an antisense oligonucleotide against *LPA* called AKCEA-APO[a]-L_{rx} has recently proved effective in reducing plasma Lp(a) levels in patients with CVD.[10] A large phase

three randomized clinical trial testing the impact of this therapy on cardiovascular outcomes in patients with CVD and elevated Lp(a) is currently ongoing.

In addition to symptoms, clinical manifestations and age of onset, increasing evidence suggest that the basic mechanisms and the risk factors for CVD may be different in men and women.[11-13] Since the overwhelming majority of studies linking Lp(a) levels to CVD risk have been performed in cohorts or post-hoc analyses from trials with an overrepresentation of men or have not specifically addressed potential sex-specific associations, the evidence linking Lp(a) to CVD risk in women remains far from conclusive. For instance, in three cohorts of women (JUPITER trial, Womens' Health Initiative and Womens' Health Study), women with high Lp(a) levels were not at higher CVD risk if they were characterized by optimal LDL cholesterol levels.[14] In the GERA study, genetically-elevated Lp(a) predicted CAVS in men, but not in women.[15] In the Cardiovascular Health Study, which included 2375 women and 1597 men aged over 65 years and followed for 7.3 years, a higher Lp(a) level was associated with the risk of stroke, death from vascular diseases and all-cause mortality in men, but not in women.[16] We also recently observed in the EPIC-Norfolk study a statistically significant risk of all-cause mortality associated with high Lp(a) levels in men but not women.[9] Whether or not high Lp(a) levels predicts CVD in women is a question of particular relevance to better understand the basic mechanisms as well as the risk factors associated with CVD in order to provide optimal care to increasing number women at high risk of CVD.

Over the past few years, several Mendelian randomization (MR) studies have documented the association between genetically-predicted circulating Lp(a) levels and cardiovascular diseases.[3, 8] Results of these studies suggested that the reduction of circulating Lp(a) levels could be associated with cardiovascular benefits. Current strategies to lower Lp(a)

levels mostly rely on hepatic *LPA* gene expression silencing.[17] To our knowledge, the association between the levels of hepatic *LPA* gene expression and cardiovascular disease has not been established using MR. Providing a causal association between hepatic *LPA* gene expression levels and cardiovascular outcomes would strengthen the case of this therapeutic strategy for CVD prevention. MR is an investigation tool in modern epidemiology that determines the association between genetic variants robustly associated with an exposure (i.e. anthropometric traits, behaviors, circulating metabolites or gene expression) and assess whether these variants are associated with outcomes (i.e. risk of chronic diseases or mortality). Akin to a randomized clinical trial, MR studies offer the possibility of investigating the association between exposures and outcomes in a "natural" randomized trial (as genetic variants are randomly acquired at meiosis) in such a way that these associations are not influenced by reverse causality and unlikely to be influenced by confounding (providing that genetic instruments predict the outcome only via their association with the exposure).

Altogether, as cardiovascular outcomes trials testing the Lp(a)-lowering hypothesis via hepatic *LPA* gene expression inhibition are expected to enroll women, the association between circulating Lp(a) and hepatic *LPA* gene expression with CVD in both men and women needs to be firmly established to inform on the potential of these therapies to decrease CVD risk in patients with high Lp(a) levels.

Here, we used a 2-sample Mendelian randomization (2SMR) study design to determine whether genetically-predicted Lp(a) levels and hepatic *LPA* gene expression levels are causally associated with CAD, IS and CAVS in men and women of the UK Biobank. We also investigated the association between plasma Lp(a) levels and long-term CAD, IS and CAVS in

men and women in the European Prospective Investigation into Cancer and Nutrition (EPIC)-Norfolk study.

Methods

Full methods used in the study are presented in the data supplement. UK Biobank has approval from the North West Multi-centre Research Ethics Committee (MREC). The Norwich District Health Authority Ethics Committee approved the study, and all participants gave signed informed consent. The data that supports the findings of this study are available from the corresponding author upon reasonable request.

Results

Genetically-predicted Lipoprotein(a) and cardiovascular outcomes in men and women of the UK Biobank

The UK Biobank study sample included a total of 408,403 participants (187,610 men and 220,793 women). Of them 37,102 had CAD (25,152 men and 11,950 women), 4283 had IS (2723 men and 1560 women) and 2574 had CAVS (1659 men and 915 women). We separated the study population into LPA-wGRS tertiles. Supplementary Table I presents median Lp(a) levels across the LPA-wGRS tertiles. Plasma Lp(a) levels were markedly elevated in the top tertile compared to the bottom and middle tertiles. The ORs for prevalent CAD, IS and CAVS in all study participants as well as in men and women separately are shown in Figure 1. Participants in the top LPA-wGRS tertile were at higher risk of CAD, IS and CAVS compared to participants in the bottom LPA-wGRS tertiles. Slightly higher associated with CAD were observed in men (OR for CAD = 1.32 [95% CI 1.27-1.36] in men and 1.20 [95% CI 1.15-1.25] in women).

Although the measures of association of the LPA-wGRS with IS were comparable in men and women, these associations only reached the level of statistical significance in men (OR for IS = 1.11 [95% CI 1.02-1.22] in men and 1.12 [95% CI 0.99-1.27] in women). Similar associations were observed in men and women with respect to CAVS (OR for CAVS = 1.43 [95% CI 1.27-1.60] in men and 1.45 [95% CI 1.24-1.70] in women).

Genetically-predicted hepatic LPA gene expression levels and cardiovascular outcomes in men and women of the UK Biobank

The hepatic *LPA* gene expression model included 80 SNPs and explained 25.0% of the variance in *LPA* gene expression ($p=8.68x10^{-13}$). We separated the UK Biobank population into tertiles of hepatic *LPA* gene expression levels. Supplementary Table II presents median Lp(a) levels across the hepatic *LPA* gene expression levels tertiles. The ORs for prevalent CAD, IS and CAVS in all study participants as well as in men and women separately are shown in Figure 2. Participants in the top tertile were at higher risk of CAD, IS and CAVS compared to participants in the bottom tertile. Similar associations were observed in men and women with respect to CAD (OR for CAD = 1.14 [95% CI 1.10-1.18] in men and 1.11 [95% CI 1.06-1.16] in women). No associations were found between hepatic *LPA* gene expression levels and IS in men and women (OR for IS = 1.01 [95%CI 0.92-1.11] in men and 1.06 [95%CI 0.94-1.20] in women). For CAVS, similar associations were observed in men and women (OR for CAVS = 1.27 [95% CI 1.13-1.43] in men and 1.22 [95% CI 1.04-1.44] in women).

Genetically-predicted Lipoprotein(a) and low-density lipoprotein cholesterol and cardiovascular outcomes in men and women of the UK Biobank

Because of the skewed distribution of Lp(a) levels, and of our results in Figure 1 showing similar association in participants of the bottom and middle LPA-wGRS tertiles, we pooled the first two

tertiles to investigate the association between the LPA-wGRS in two LDL cholesterol level categories (those below the median versus equal or above the median of a LDL-C-wGRS). Supplementary Table III presents median Lp(a) and LDL cholesterol levels in these subgroups. Results presented in Supplementary Figure I suggest that participants in the top LPA-wGRS and with a LDL-C-wGRS equal or above the median were at the highest risk of CAD (with men at slightly higher risk than women in these subcatergories). Men in the top LPA-wGRS were at higher IS risk, regardless of LDL-C-wGRS categories while women in the top LPA-wGRS were at higher IS risk, but only in those with a genetically higher LDL-C-wGRS. Both men and women in the top LPA-wGRS and with a LDL-C-wGRS equal or above the median were at the highest risk of CAVS.

Lipoprotein(a) levels and cardiovascular outcomes in men and women of the EPIC-Norfolk study

The baseline characteristics of EPIC-Norfolk study participants by Lp(a) levels have been recently published.[9] The average follow-up of the EPIC-Norfolk study was 20 years. The distribution of Lp(a) was comparable in men and women (Supplementary Figure II). Compared to participants with Lp(a) levels <50 mg/dL, those with Lp(a) levels ≥50 mg/dL had a higher hazard ratio (HR) of CAD, IS and CAVS (Supplementary Table IV). In sex-specific analyses, high Lp(a) levels predicted higher risk of CAD and CAVS in both men and women while the association of high Lp(a) levels with IS was not statistically significant when the data was analyzed in men and women separately.

In order to identify the level of Lp(a) at which Lp(a) may be associated with cardiovascular outcomes, we investigated the effect of Lp(a) on CAD, IS and CAVS incidence in participants above the 50th percentile of the Lp(a) distribution according to pre-established

percentiles of men and women pooled together. Results presented in Table 1 suggest that, in the total population, a higher CAD risk was observed in participants above the median and the strength of the Lp(a)-CAD association increased up to the 90th percentile value of the Lp(a) distribution. Similar observations were observed in men only. In women, however, only those with an Lp(a) level equal or above the 90th Lp(a) percentile were at increased CAD risk. Table 2 and Table 3 present the effect of Lp(a) on IS and CAVS incidence in participants above the 50th percentile of the Lp(a) distribution, respectively. We could not establish a cutoff value at which Lp(a) was associated with an increase IS risk, in all study participants or in men and women separately. In the total study population, participants above the 90th Lp(a) percentile had a higher CAVS risk. In men, the risk of CAVS appeared to be statistically significant in those with Lp(a) equal or above the 95th percentile of the Lp(a) distribution. In women, CAVS risk was increased in those with Lp(a) equal or above the 90th percentile of the Lp(a) distribution, suggesting that higher Lp(a) percentiles may be associated with some cardiovascular outcomes in a sex-specific manner.

Discussion

In the absence of robust evidence linking high Lp(a) levels with cardiovascular outcomes in women, we used a genetic association study akin to Mendelian randomization study to determine the sex-specific association of lifelong exposure to elevated Lp(a) levels or elevated hepatic *LPA* gene expression level and the risk of CAD, IS and CAVS in men and women separately. Although measures of association of Lp(a) metrics and CVD were sometimes higher in men, our results suggest that the association between Lp(a) levels and the risk of CAD, IS and CAVS is comparable in men and women, and with the exception of IS, independent of genetically-

predicted LDL cholesterol levels. Here, we used two study designs: a Mendelian randomization and observational analyses. The advantage of Mendelian randomization is that our exposure is genetically estimated and less likely to be influenced by confounders or random measurement error. This study design is particularly useful here as some confounders might influence our exposure (Lp[a]) levels) in a sex-specific manner. For instance, plasma Lp(a) levels could be influenced by sex, menopausal status and hormone replacement therapy use.[18-19] Plasma Lp(a) levels could also be influenced by lipid-lowering therapies such as statins, which are prescribed to men in a higher proportion.[20] Interestingly however, we replicated these results in a large observational long-term follow-up study of >18,000 British men and women and found very similar associations. Observational analyses performed in the EPIC-Norfolk study identified sex-specific cutpoints at which Lp(a) levels might be associated with cardiovascular outcomes.

Lp(a) is an important risk factor for a broad range of CVD such as CAD, IS and CAVS. The increasing evidence suggesting that genetically-elevated Lp(a) levels is linked with these outcomes also supports the causal relationship between Lp(a) and CVD. Many pathobiological mechanisms have been proposed to explain the detrimental impact of Lp(a) on the onset of CVD. First, Lp(a) has a unique "pro-inflammatory" proteome and lipidome; it is an important carrier of oxidized phospholipids (OxPL) in the bloodstream.[1] OxPLs promote tissue necrosis as well as macrophage chemotaxis and oxLDL uptake within the arterial wall, thereby contributing to plaque inflammation.[21] Recent studies also suggest that Lp(a) might have pro-calcifying properties, which could be attributed to their OxPL cargo as well as lysophosphatidic acid (lysp-PA).[22] Lyso-PA may also activate pro-inflammatory mechanisms in valvular interstitial cells.[23]

Two studies recently estimated the magnitude of Lp(a) reductions that will be required to generate cardiovascular benefits comparable to what can be obtained from LDL-C lowering

therapies.[24, 28] These estimates range between 65 and 100 mg/dL. Although our study provides evidence that the relative risk of CVD may be comparable in men and women, since men are at higher absolute risk for CVD, we believe that additional studies will be required to determine whether the extent of Lp(a)-lowering required to derive significant and cost-effective cardiovascular benefits is comparable in men and women. The genetic variants at the *LPA* locus that were used to identify SNPs associated with high Lp(a) levels included SNPs that may be associated with the expression and/or the function of nearby genes (including but not limited to *PLG*, *SLC22A3*, and *IGF2R*) and may influence CVD risk via Lp(a)-independent mechanism, a limitation of MR known as horizontal pleiotropy. Given that our results suggest that variants influencing hepatic *LPA* genetic expression are also associated with CVD and that other reports using sequence variants and intronic SNPs in the *LPA* gene also reported an effect of these variants on the risk of CVD in the UK Biobank and other cohorts [3,4]), the possibility of horizontal pleiotropy driving the reported associations is unlikely.

The evidence supporting Lp(a) as a potential therapeutic target for residual cardiovascular risk emerged from a wide range of studies with different designs, from basic and translational studies, to long-term observation studies, as well as Mendelian randomization studies. However, the ultimate proof of causality of Lp(a) in the setting of CVD would be a trial demonstrating that reducing Lp(a) levels would impact long-term health outcomes (Figure 3). Gene silencing therapies are currently under investigation for their capacity to influence both Lp(a) levels and CVD risk. The antisense oligonucleotide AKCEA-APO[a]-L_{rx} has recently been shown to be efficient in reducing plasma Lp(a) levels in patients with documented cardiovascular diseases.[10] An effect of this investigative therapy on pro-inflammatory gene expression of blood monocytes and the reduction in transendothelial migration capacity of monocytes has also recently been

suggested.[25] Although the sex-specific impact of this therapy was not reported, under the assumption of a comparable Lp(a) reduction for men and women, our study results suggest that the impact of Lp(a)-lowering therapies might reduce relative CVD risk in men and women. Our study also provides evidence that the inhibition of hepatic *LPA* gene expression may also be associated with cardiovascular benefits. As most strategies currently under development to target Lp(a) are based on drugs that interfere with *LPA* messenger RNA (such as antisense oligonucleotides or siRNAs), this finding suggests that the mechanism of action of RNA-interfering therapy may represent an effect way to reduce Lp(a) levels and influence long-term CVD risk. However, in both cases, only a long-term clinical trial of Lp(a)-lowering with investigative therapies will inform on change in risk or health trajectories of men and women with high Lp(a).

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study; collection, management, analysis, and interpretation of the data; preparation, review, or approval of the manuscript; and decision to submit the manuscript for publication.

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Supplemental Material
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References [26-35]

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Table 1. Sex-specific risk of coronary artery disease associated with very high lipoprotein(a) levels. Event rates and hazard ratios for coronary artery disease in participants of the EPIC-Norfolk study with very high lipoprotein(a) levels for the entire cohort are presented, as well as in men and women separately. Model 1 is adjusted for age and sex (in the total study population). Model 2 is adjusted for age, sex (in the total study population), smoking, body mass index, systolic blood pressure, diabetes mellitus and estimated glomerular filtration rate.

Lp(a) percentiles	≤50	>50-80	>80-90	>90-95	>95-100
Lp(a) range, mg/dL	≤11.4	>11.4 to 35.0	>35.0 to 53.3	>53.3 to 69.7	>69.7
All					
Cases/total (event rate, %)	1751/9366	1240/5611	442/1872	267/936	264/936
	(18.7)	(22.1)	(23.6)	(28.5)	(28.2)
Model 1	1.00	1.11 (1.03-1.2)	1.3 (1.17-1.44)	1.65 (1.45-1.88)	1.58 (1.39-1.79)
Model 2	1.00	1.12 (1.04-1.2)	1.31 (1.18-1.46)	1.66 (1.46-1.9)	1.61 (1.42-1.84)
Men					
Cases/total (event rate, %)	1077/4288	732/2476	292/865	144/384	150/371
	(25.1)	(29.6)	(33.8)	(37.5)	(40.4)
Model 1	1.00	1.12 (1.02-1.23)	1.42 (1.25-1.62)	1.68 (1.41-2)	1.73 (1.46-2.05)
Model 2	1.00	1.13 (1.03-1.25)	1.41 (1.24-1.61)	1.72 (1.44-2.05)	1.84 (1.55-2.19)
Women					
Cases/total (event rate, %)	674/5078	508/3135	150/1007	123/552	114/565
	(13.3)	(16.2)	(14.9)	(22.3)	(20.2)
Model 1	1.00	1.09 (0.97-1.23)	1.1 (0.92-1.31)	1.58 (1.3-1.91)	1.38 (1.13-1.68)
Model 2	1.00	1.09 (0.97-1.22)	1.16 (0.97-1.38)	1.57 (1.29-1.91)	1.34 (1.1-1.65)

Table 2. Sex-specific risk of ischemic stroke associated with very high lipoprotein(a) levels. Event rates and hazard ratios for ischemic stroke in participants of the EPIC-Norfolk study with very high lipoprotein(a) levels for the entire cohort are presented, as well as in men and women separately. Model 1 is adjusted for age and sex (in the total study population). Model 2 is adjusted for age, sex (in the total study population), smoking, body mass index, systolic blood pressure, diabetes mellitus and estimated glomerular filtration rate.

Lp(a) percentiles	≤50	>50-80	>80-90	>90-95	>95-100
Lp(a) range, mg/dL	≤11.4	>11.4 to 35.0	>35.0 to 53.3	>53.3 to 69.7	>69.7
All					
Cases/total (event rate, %)	412/9367	256/5613	73/1872	52/936	53/937
	(4.4)	(4.6)	(3.9)	(5.6)	(5.7)
Model 1	1.00	0.92 (0.79-1.08)	0.87 (0.68-1.11)	1.23 (0.92-1.64)	1.22 (0.91-1.62)
Model 2	1.00	0.90 (0.77-1.05)	0.87 (0.67-1.12)	1.21 (0.9-1.63)	1.2 (0.9-1.61)
Men					
Cases/total (event rate, %)	198/4289	133/2477			
	(4.6)	(5.4)	37/865 (4.3)	25/384 (6.5)	18/371 (4.9)
Model 1	1.00	1.06 (0.85-1.32)	0.92 (0.65-1.31)	1.53 (1.01-2.31)	1.06 (0.66-1.72)
Model 2	1.00	1.06 (0.85-1.32)	0.92 (0.65-1.32)	1.56 (1.03-2.37)	1.15 (0.71-1.87)
Women					
Cases/total (event rate, %)	214/5078	123/3136	36/1007	27/552	35/566
	(4.2)	(3.9)	(3.6)	(4.9)	(6.2)
Model 1	1.00	0.81 (0.65-1.01)	0.81 (0.57-1.15)	1.01 (0.68-1.51)	1.28 (0.9-1.83)
Model 2	1.00	0.76 (0.61-0.96)	0.81 (0.56-1.17)	0.96 (0.64-1.46)	1.2 (0.83-1.74)

Table 3. Sex-specific risk of calcific aortic valve stenosis associated with very high lipoprotein(a) levels. Event rates and hazard ratios for calcific aortic valve stenosis in participants of the EPIC-Norfolk study with very high lipoprotein(a) levels for the entire cohort are presented, as well as in men and women separately. Model 1 is adjusted for age and sex (in the total study population). Model 2 is adjusted for age, sex (in the total study population), smoking, body mass index, systolic blood pressure, diabetes mellitus and estimated glomerular filtration rate.

Lp(a) percentiles	≤50	>50-80	>80-90	>90-95	>95-100
Lp(a) range, mg/dL	≤11.4	>11.4 to 35.0	>35.0 to 53.3	>53.3 to 69.7	>69.7
All					
Cases/controls (event rate, %)	178/9367	124/5613	46/1872	33/936	43/937
	(1.9)	(2.2)	(2.5)	(3.5)	(4.6)
Model 1	1.00	1.05 (0.83-1.32)	1.27 (0.92-1.76)	1.83 (1.26-2.65)	2.36 (1.69-3.3)
Model 2	1.00	1.03 (0.82-1.3)	1.31 (0.95-1.82)	1.90 (1.31-2.75)	2.44 (1.74-3.42)
Men					
Cases/controls (event rate, %)	94/4289	64/2477	27/865	13/384	23/371
	(2.2)	(2.6)	(3.1)	(3.4)	(6.2)
Model 1	1.00	1.07 (0.78-1.47)	1.45 (0.94-2.22)	1.63 (0.92-2.92)	2.93 (1.86-4.63)
Model 2	1.00	1.06 (0.77-1.47)	1.49 (0.97-2.29)	1.69 (0.94-3.02)	3.12 (1.97-4.94)
Women					
Cases/controls (event rate, %)	84/5078 (1.7)	60/3136 (1.9)	19/1007 (1.9)	20/552 (3.6)	20/566 (3.5)
Model 1	1.00	1.02 (0.73-1.42)	1.08 (0.66-1.78)	1.96 (1.2-3.19)	1.90 (1.17-3.1)
Model 2	1.00	0.99 (0.71-1.39)	1.12 (0.68-1.85)	1.99 (1.22-3.24)	1.84 (1.12-3.03)

Figure Legends:

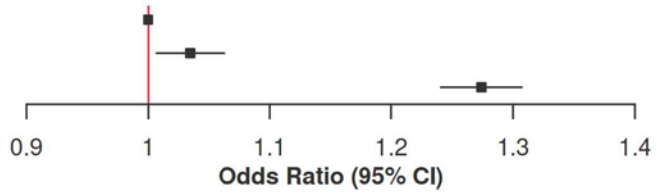
Figure 1. Sex-specific association between a weighted genetic risk score of lipoprotein(a) levels and cardiovascular disease risk. Sex-specific association between genetically-predicted lipoprotein(a) levels and the risk of coronary artery disease (A) ischemic stroke (B) and calcific aortic valve stenosis (C) in the UK Biobank. To estimate genetically-predicted lipoprotein(a) levels, study participants were separated into tertiles according to a weighted genetic-risk score of 26 independent single-nucleotide polymorphisms in the *LPA* region weighted for its impact on lipoprotein(a) levels and the odds ratios for cardiovascular diseases were obtained after adjusting for age, sex (in the non-sex-specific analyses) for the 10 main ancestry-based principal components.

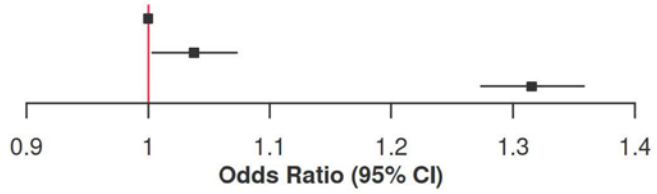
Figure 2. Sex-specific association between genetically-predicted hepatic *LPA* gene expression levels and cardiovascular disease risk. Sex-specific association between genetically-predicted hepatic *LPA* gene expression levels and the risk of coronary artery disease (A) ischemic stroke (B) and calcific aortic valve stenosis (C) in the UK Biobank. To estimated genetically-predicted hepatic *LPA* gene expression levels, study participants were separated into tertiles according to their genetically-predicted level of hepatic *LPA* gene expression and the odds ratios for cardiovascular diseases were obtained after adjusting for age, sex (in the non-sex-specific analyses) and the 10 main ancestry-based principal components.

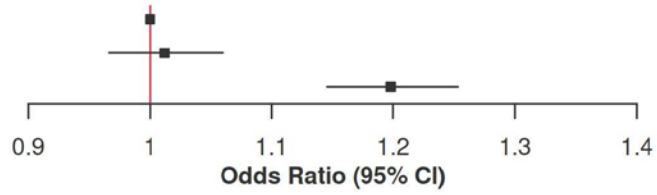
Figure 3. Mendelian randomization analysis provides evidence that genetically-predicted lipoprotein(a) levels and genetically-predicted *LPA* hepatic *LPA* expression levels may be causally linked to cardiovascular outcomes in the UK Biobank.

A

	Tertile	Odds Ratio for Coronary Artery Disease (CI 95%)	P-value	Cases/Controls
All	T1	1.00 (1.00 - 1.00)	NA	11,322 / 124,813
	T2	1.03 (1.01 - 1.06)	0.014	11,680 / 124,454
	T3	1.27 (1.24 - 1.31)	<0.001	14,100 / 122,034
Men	T1	1.00 (1.00 - 1.00)	NA	7,623 / 54,914
	T2	1.04 (1.00 - 1.07)	0.032	7,874 / 54,662
	T3	1.32 (1.27 - 1.36)	<0.001	9,655 / 52,882
Women	T1	1.00 (1.00 - 1.00)	NA	3,737 / 69,861
	T2	1.01 (0.97 - 1.06)	0.62	3,780 / 69,817
	T3	1.20 (1.15 - 1.25)	<0.001	4,433 / 69,165

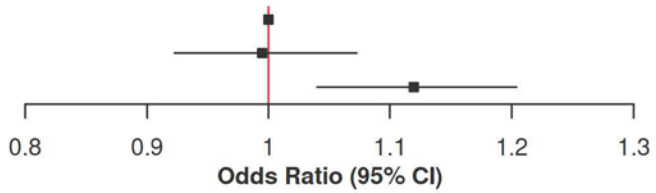


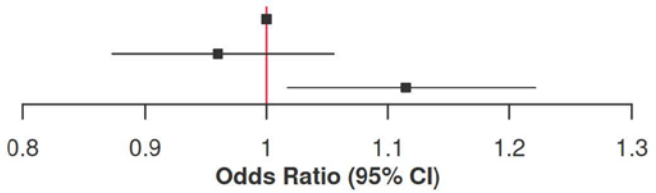


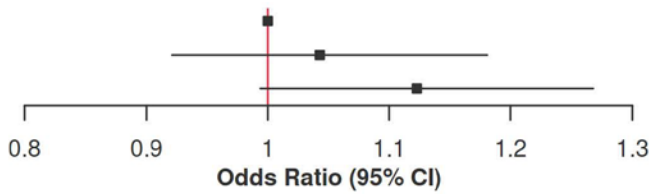


B

	Tertile	Odds Ratio for Ischemic Stroke (CI 95%)	P-value	Cases/Controls
All	T1	1.00 (1.00 - 1.00)	NA	1,376 / 134,759
	T2	0.99 (0.92 - 1.07)	0.91	1,369 / 134,765
	T3	1.12 (1.04 - 1.20)	0.0027	1,538 / 134,596
Men	T1	1.00 (1.00 - 1.00)	NA	886 / 61,651
	T2	0.96 (0.87 - 1.06)	0.41	851 / 61,685
	T3	1.11 (1.02 - 1.22)	0.021	986 / 61,551
Women	T1	1.00 (1.00 - 1.00)	NA	493 / 73,105
	T2	1.04 (0.92 - 1.18)	0.53	514 / 73,083
	T3	1.12 (0.99 - 1.27)	0.067	553 / 73,045

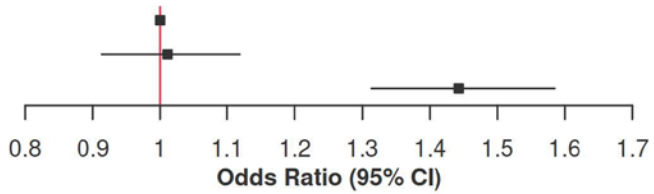


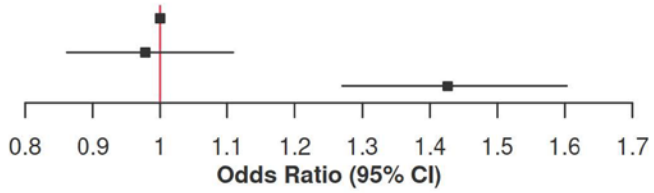


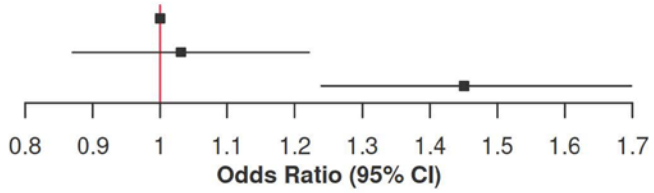


C

	Tertile	Odds Ratio for Calcific Aortic Valve Stenosis (CI 95%)	P-value	Cases/Controls
All	T1	1.00 (1.00 - 1.00)	NA	746 / 135,389
	T2	1.01 (0.91 - 1.12)	0.86	754 / 135,380
	T3	1.44 (1.31 - 1.58)	<0.001	1,074 / 135,060
Men	T1	1.00 (1.00 - 1.00)	NA	488 / 62,049
	T2	0.98 (0.86 - 1.11)	0.75	477 / 62,059
	T3	1.43 (1.27 - 1.60)	<0.001	694 / 61,843
Women	T1	1.00 (1.00 - 1.00)	NA	263 / 73,335
	T2	1.03 (0.87 - 1.22)	0.76	271 / 73,326
	T3	1.45 (1.24 - 1.70)	<0.001	381 / 73,217

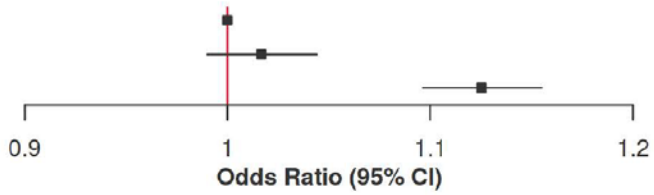


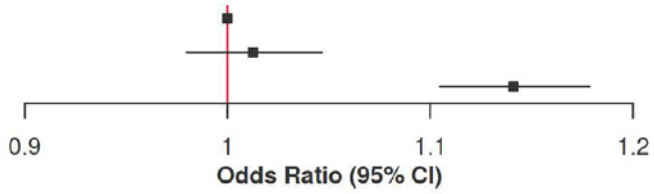


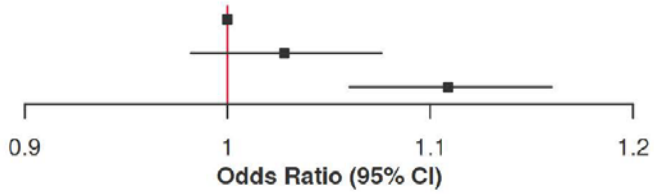


A

	Tertile	Odds Ratio for Coronary Artery Disease (CI 95%)	P-value	Cases/Controls
All	T1	1.00 (1.00 - 1.00)	NA	11,860 / 124,275
	T2	1.02 (0.99 - 1.04)	0.23	12,040 / 124,094
	T3	1.13 (1.10 - 1.15)	<0.001	13,202 / 122,932
Men	T1	1.00 (1.00 - 1.00)	NA	8,031 / 54,506
	T2	1.01 (0.98 - 1.05)	0.46	8,120 / 54,416
	T3	1.14 (1.10 - 1.18)	<0.001	9,001 / 53,536
Women	T1	1.00 (1.00 - 1.00)	NA	3,819 / 69,779
	T2	1.03 (0.98 - 1.08)	0.24	3,920 / 69,677
	T3	1.11 (1.06 - 1.16)	<0.001	4,211 / 69,387

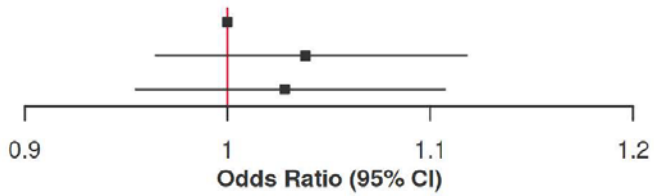


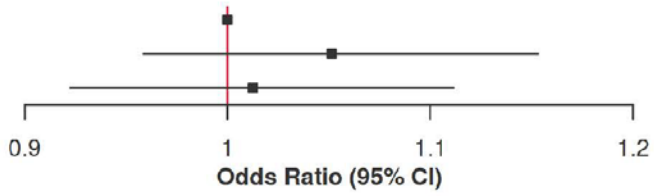


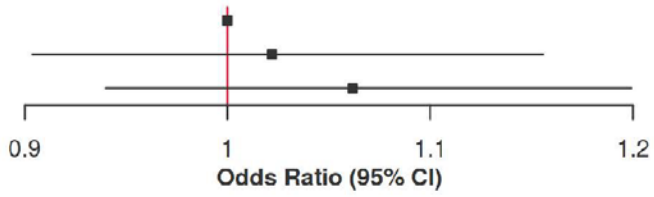


B

	Tertile	Odds Ratio for Ischemic Stroke (CI 95%)	P-value	Cases/Controls
All	T1	1.00 (1.00 - 1.00)	NA	1,397 / 134,738
	T2	1.04 (0.96 - 1.12)	0.33	1,450 / 134,684
	T3	1.03 (0.95 - 1.11)	0.47	1,436 / 134,698
Men	T1	1.00 (1.00 - 1.00)	NA	889 / 61,648
	T2	1.05 (0.96 - 1.15)	0.3	934 / 61,602
	T3	1.01 (0.92 - 1.11)	0.81	900 / 61,637
Women	T1	1.00 (1.00 - 1.00)	NA	506 / 73,092
	T2	1.02 (0.90 - 1.16)	0.75	517 / 73,080
	T3	1.06 (0.94 - 1.20)	0.35	537 / 73,061

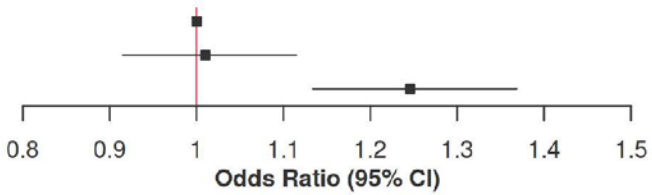


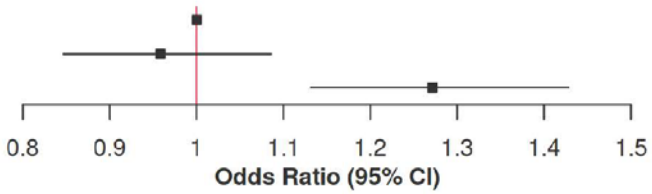


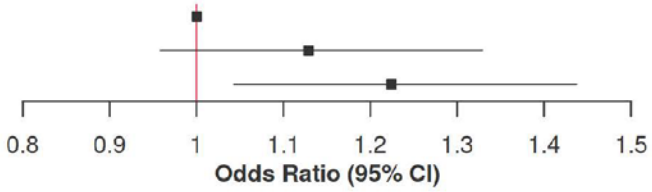


C

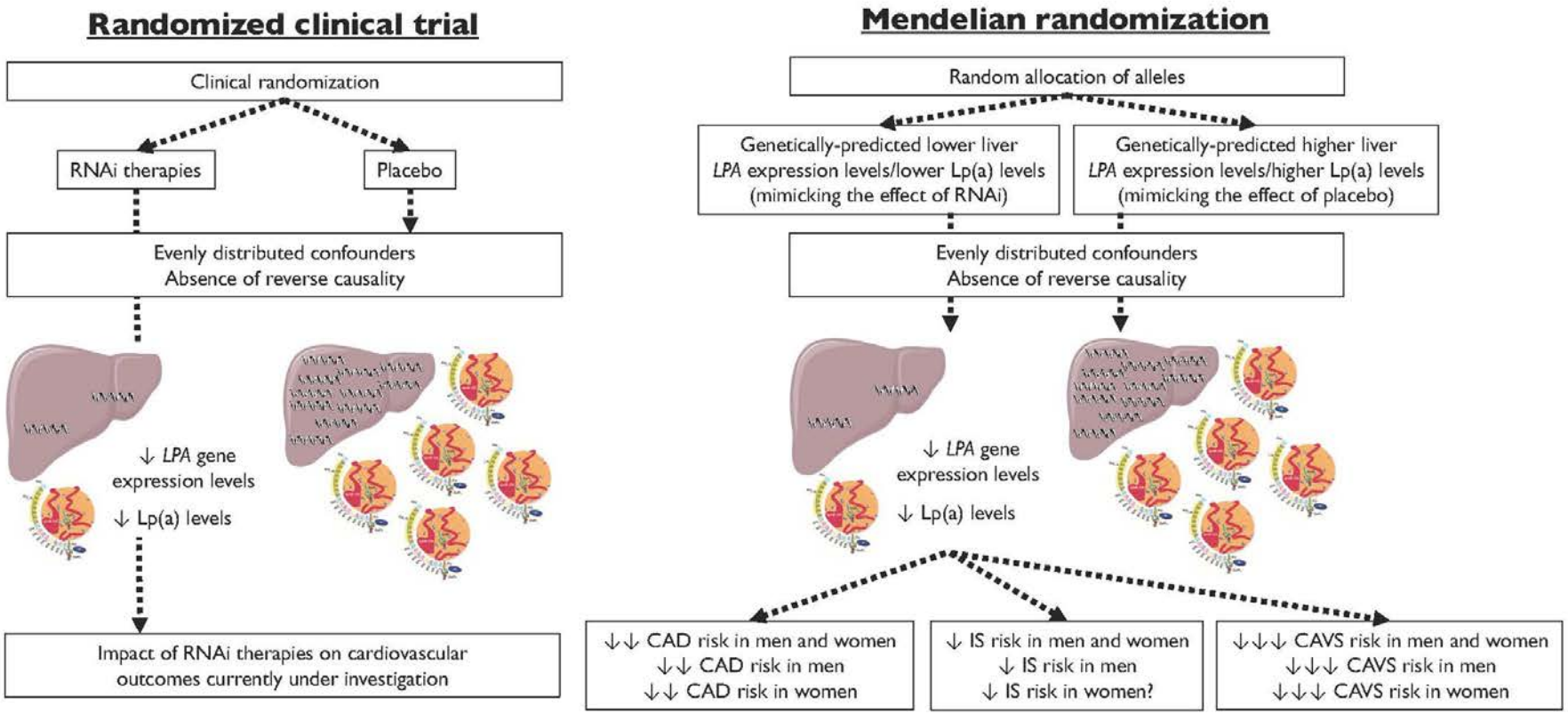
	Tertile	Odds Ratio for Calcific Aortic Valve Stenosis (CI 95%)	P-value	Cases/Controls
All	T1	1.00 (1.00 - 1.00)	NA	791 / 135,344
	T2	1.01 (0.92 - 1.11)	0.86	799 / 135,335
	T3	1.25 (1.13 - 1.37)	<0.001	984 / 135,150
Men	T1	1.00 (1.00 - 1.00)	NA	514 / 62,023
	T2	0.96 (0.85 - 1.09)	0.53	493 / 62,043
	T3	1.27 (1.13 - 1.43)	<0.001	652 / 61,885
Women	T1	1.00 (1.00 - 1.00)	NA	273 / 73,325
	T2	1.13 (0.96 - 1.33)	0.16	308 / 73,289
	T3	1.22 (1.04 - 1.44)	0.015	334 / 73,264







Study objectives: Determine the sex-specific associations of genetically-predicted circulating Lp(a) and hepatic *LPA* gene expression levels with cardiovascular outcomes using Mendelian randomization



Study conclusions: Genetically-predicted circulating Lp(a) and hepatic *LPA* gene expression levels are associated with a lower risk of CAD, IS and CAVS in men and women included in the UK Biobank