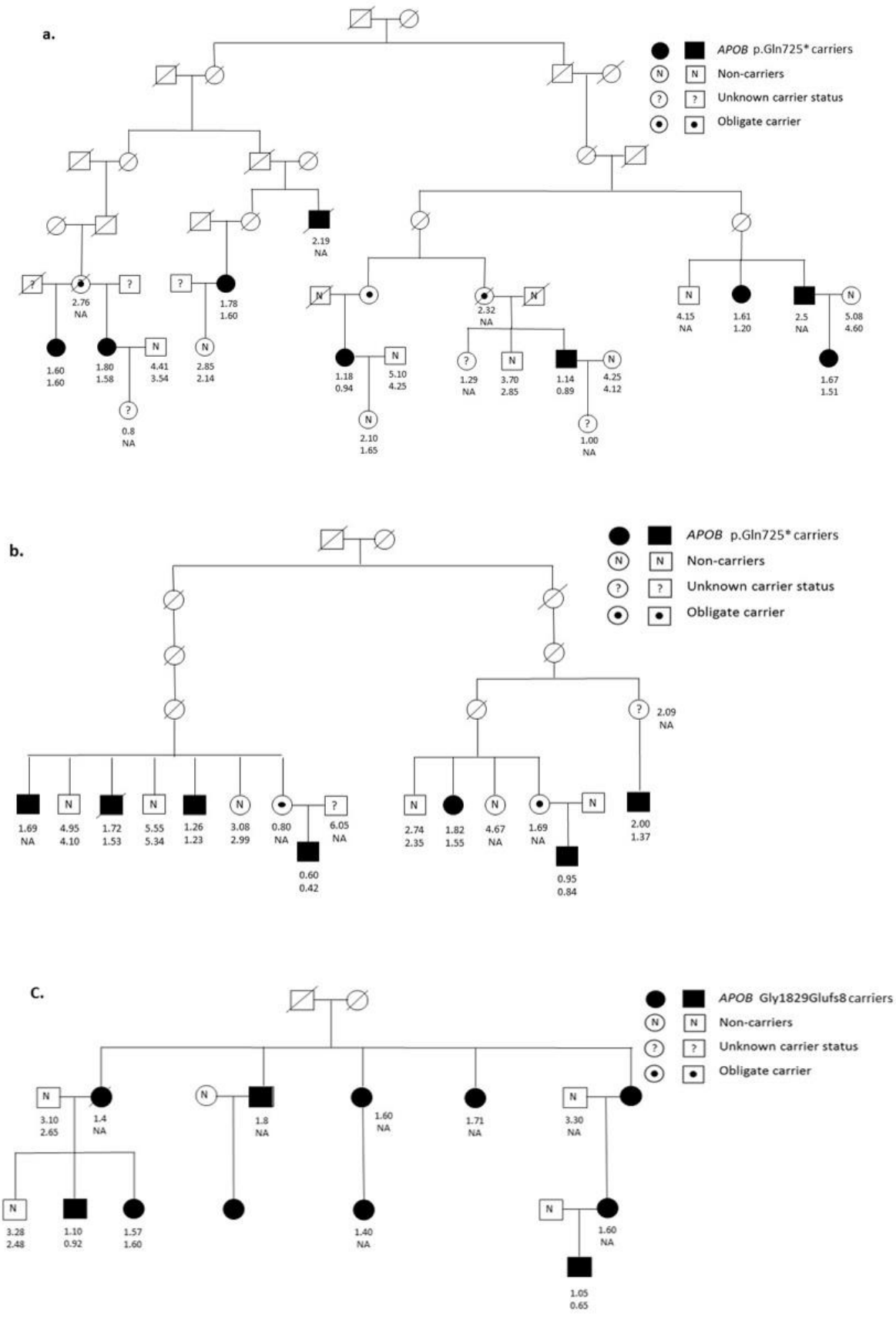
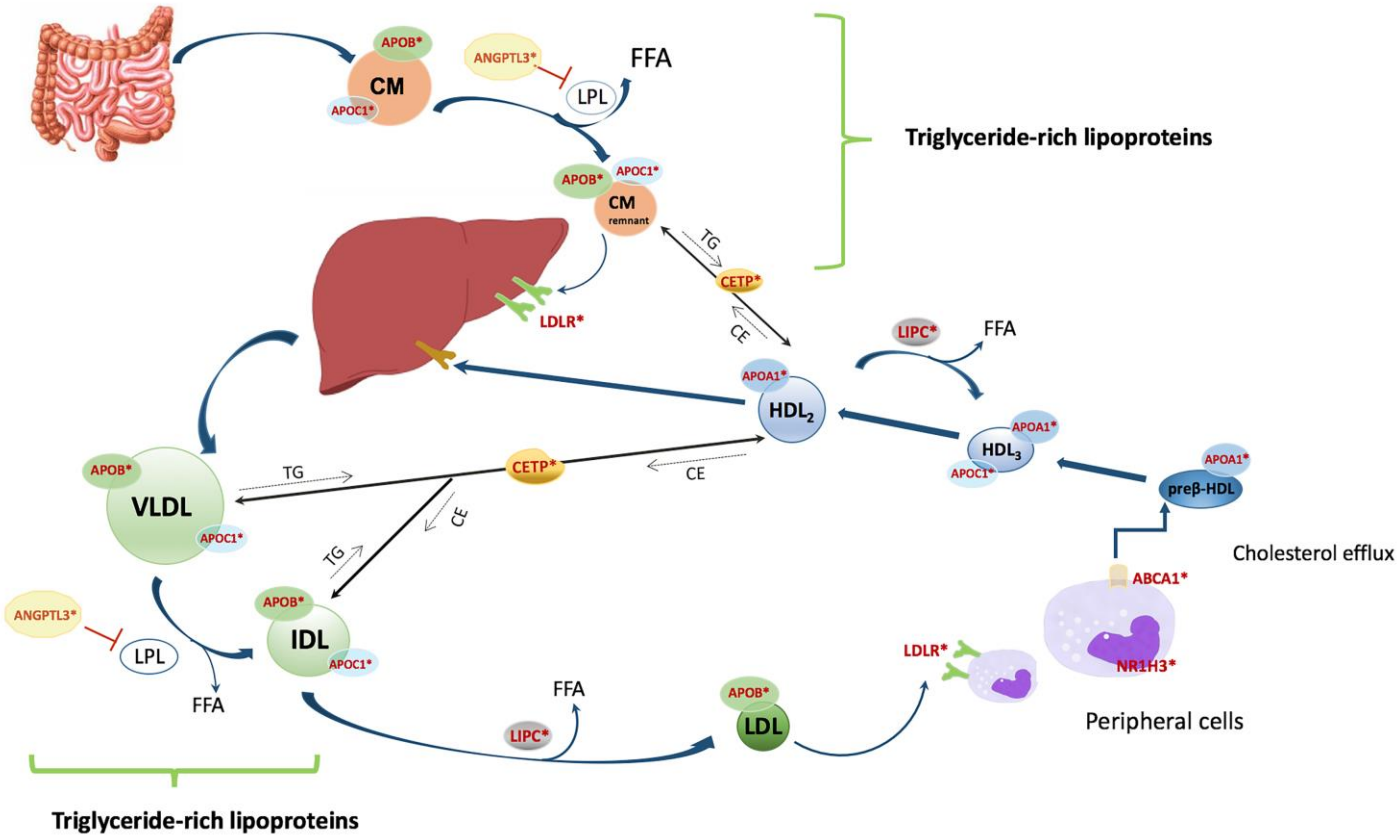

a.
APOB p.Gln725* carriers
Non-carriers
Unknown carrier status
Obligate carrier
2.19
NA
2.76
NA
1.78
1.60
2.32
NA
4.15
NA
1.61
1.20
2.5
NA
5.08
4.60
1.60
1.60
1.80
1.58
4.41
3.54
2.85
2.14
0.8
NA
1.18
0.94
5.10
4.25
2.10
1.65
1.29
NA
3.70
2.85
1.14
0.89
4.25
4.12
1.00
NA
1.67
1.51
b.
APOB p.Gln725* carriers
Non-carriers
Unknown carrier status
Obligate carrier
2.09
NA
1.69
NA
4.95
4.10
1.72
1.53
5.55
5.34
1.26
1.23
3.08
2.99
0.80
NA
6.05
NA
0.60
0.42
2.74
2.35
1.82
1.55
4.67
NA
1.69
NA
2.00
1.37
0.95
0.84
C.
APOB Gly1829Glufs8 carriers
Non-carriers
Unknown carrier status
Obligate carrier
3.10
2.65
1.4
NA
1.8
NA
1.60
NA
1.71
NA
3.30
NA
3.28
2.48
1.10
0.92
1.57
1.60
1.40
NA
1.60
NA
1.05
0.65

Supplementary Figure 1

Pedigrees of families with *APOB* p.Gln725* mutation and *APOB* p.Gly1829Glufs8 mutation

(**a**,**b**) Pedigrees of families with *APOB* p.Gln725* mutation. (**c**) Pedigree of family with *APOB* p.Gly1829Glufs8 mutation. Squares denote male family members, circles denote female family members, and symbols with slashes denote deceased family members. Values below symbols are non-HDL cholesterol (above) and LDL cholesterol (below) in mmol/L for family members with available lipid information. The pedigrees have been simplified to maintain confidentiality.



Supplementary Figure 2

A schematic illustration of the lipid metabolism genes harboring mutations described in this study.
Cholesterol and triglycerides (TG) are transferred together by triglyceride-rich lipoproteins, including very low-density lipoprotein (VLDL), intermediate-density lipoproteins (IDL), chylomicrons, and chylomicron remnants. Apolipoprotein B, encoded by *APOB*, is an essential structural component of VLDL, IDL, LDL, and chylomicrons. The *APOA1* gene encodes apolipoprotein A-I, the major protein component of HDL particles that have a key role in reverse cholesterol transport. Apolipoprotein C-I, encoded by *APOC1*, is associated with both HDL and triglyceride-rich lipoproteins, inhibits cholesteryl ester transfer protein (*CETP*), and hinders hepatic clearance of triglyceride-rich lipoproteins. ATP-binding cassette transporter (*ABCA1*) mediates the efflux of cholesterol and phospholipids to lipid-poor apolipoprotein A-1 to form nascent pre-β-HDL. Triglycerides from triglyceride-rich lipoproteins are hydrolyzed into free fatty acids by lipoprotein lipase (LPL). Angiopoietin-like 3 (encoded by the *ANGPTL3* gene) inhibits LPL. CETP and hepatic lipase (encoded by *LIPC*) are central in lipoprotein remodeling. CETP facilitates transfer of cholesteryl ester (CE) from HDL to triglyceride-rich lipoproteins in exchange for triglycerides. Hepatic lipase (LIPC) remodels large lipoprotein particles into smaller and denser particles. IDL is converted to LDL by the action of hepatic lipase (*LIPC*). LDL is taken up by liver and other tissues in an endocytotic process that involves the LDL receptor (*LDLR*). Liver X receptor, encoded by the *NR1H3* gene, is a cholesterol sensor that induces transcription of multiple genes that protect cells from cholesterol overload.

Supplementary Note

The Egger regression estimate proposed by Bowden *et al.*[1] using the effect estimates for the allele that has a positive effect on the predictor, can be extended to include multiple covariates for performing weighted multiple regression. When we apply multiple Egger regression to the data of Do *et al.*[2], the triglyceride effect, with high density lipoprotein cholesterol (HDL-C) and low density lipoprotein cholesterol (LDL-C) levels included in the analysis, is 0.15 (P=0.028), lower than the reported 0.37 (P=1.3e-9) based on residual regression (See Table). Since non-HDL-C encompasses cholesterol from triglyceride-rich lipoproteins (including very low density lipoprotein cholesterol (VLDL-C)) and has a stronger correlation with the risk of coronary artery disease (CAD) than LDL-C[3], we are also interested in whether triglycerides associate with the risk of CAD after accounting for non-HDL-C instead of LDL-C levels. Although the effect estimates of Do *et al.* do not include non-HDL-C effects, these can be estimated from the LDL-C and triglyceride effects. The multiple Egger regression estimates this effect as insignificant 0.09 (P=0.33), while the linear regression estimates it as 0.22 (P=0.00011).

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Predictor	Covariates	Residual regression effect (P)	Multiple Egger regression effect (P)
LDL-C	–	0.41 (9.7e-21)	0.62 (1.5e-14)
LDL-C	TG, HDL-C	0.39 (4.8e-23)	0.56 (3.6e-15)
HDL-C	–	-0.18 (0.00066)	-0.29 (0.0050)
HDL-C	LDL-C, TG	-0.03 (0.37)	-0.14 (0.11)
TG	–	0.44 (1.7e-8)	0.39 (1.2e-5)
TG	LDL-C, HDL-C	0.37 (1.3e-9)	0.15 (0.028)
TG	Non-HDL-C, HDL-C	0.22 (0.00011)	0.09 (0.33)
Non-HDL-C	–	0.50 (9.6e-27)	0.65 (4.9e-16)
Non-HDL-C	TG, HDL-C	0.42 (2.2e-21)	0.56 (3.5e-12)

Table. Mendelian randomization analysis of the relationship between lipid levels and coronary artery disease (CAD) risk. The lipid levels are for low density lipoprotein cholesterol (LDL-C), high density lipoprotein cholesterol (HDL-C), triglycerides (TG) and non-HDL cholesterol (non-HDL-C). The non-HDL-C effects were estimated based on the relationship between LDL-C and TG effects and non-HDL-C effects in Iceland.

References

1. Bowden, J., Davey Smith, G. & Burgess, S. Mendelian randomization with invalid instruments: effect estimation and bias detection through Egger regression. Int J Epidemiol 44, 512-25 (2015).

2. Do, R. et al. Common variants associated with plasma triglycerides and risk for coronary artery disease. Nat Genet 45, 1345-52 (2013).

3. Varbo, A. et al. Remnant cholesterol as a causal risk factor for ischemic heart disease. J Am Coll Cardiol 61, 427-36 (2013).