Genome-wide association study of the postprandial triglyceride response yields common genetic variation in LIPC (hepatic lipase)

Ibi, D.; Noordam, R.; Klinken, J.B. van; Li-Gao, R.F.; Mutsert, R. de; Trompet, S.; ... ; Dijk, K.W. van

Circulation: Genomic and Precision Medicine

ORIGINAL ARTICLE

Genome-Wide Association Study of the Postprandial Triglyceride Response Yields Common Genetic Variation in LIPC (Hepatic Lipase)

Dorina Ibi, MSc; Raymond Noordam, PhD; Jan Bert van Klinken, PhD; Ruifang Li-Gao, PhD; Renée de Mutsert, PhD; Stella Trompet, PhD; Tim Christen, MSc; Lisanne L. Blauw, PhD; Diana van Heemst, PhD; Dennis O. Mook-Kanamori, MD, PhD; Frits R. Rosendaal, MD, PhD; J. Wouter Jukema, MD, PhD; Martijn E.T. Dollé, PhD; Patrick C.N. Rensen, PhD; Ko Willems van Dijk, PhD

BACKGROUND: The increase in serum triglyceride (TG) concentrations in response to a meal is considered a risk factor for cardiovascular disease. We aimed to elucidate the genetics of the postprandial TG response through genome-wide association studies (GWAS).

METHODS: Participants of the NEO (Netherlands Epidemiology of Obesity) study (n=5630) consumed a liquid mixed meal after an overnight fast. GWAS of fasting and postprandial serum TG at 150 minutes were performed. To identify genetic variation of postprandial TG independent of fasting TG, we calculated the TG response at 150 minutes by the residuals of a nonlinear regression that predicted TG at 150 minutes as a function of fasting TG. Association analyses were adjusted for age, sex, and principal components in a linear regression model. Next, using the identified variants as determinants, we performed linear regression analyses on the residuals of the postprandial response of 149 nuclear magnetic resonance–based metabolite measures.

RESULTS: GWAS of fasting TG and postprandial serum TG at 150 minutes resulted in completely overlapping loci, replicating previous GWAS. From GWAS of the TG response, we identified rs7350789-A (allele frequency=0.36), mapping to hepatic lipase (*LIPC*), to be associated with a smaller increase in TG concentrations at 150 minutes (β=–0.11; *P*-value=5.1×10^{-8}). Rs7350789-A was associated with responses of 33 metabolite measures (*P*-value $<1.34\times10^{-3}$), mainly smaller increases of the TG-component in almost all HDL (high-density lipoprotein) subparticles (HDL-TG), a smaller decrease of HDL diameter and smaller increases of most components of VLDL (very low density lipoprotein) subparticles.

CONCLUSIONS: GWAS of the TG response identified a variant near *LIPC* as a main contributor to postprandial TG metabolism independent of fasting TG concentrations, resulting in smaller increases of HDL-TG and VLDL subparticles.

Key Words: cardiovascular diseases ■ Genome-wide association studies ■ hepatic lipase ■ lipoprotein metabolism ■ metabolites ■ postprandial triglyceride response

It is becoming increasingly clear that both fasting and postprandial (nonfasting) serum triglyceride (TG) concentrations are independent risk factors for developing cardiovascular disease (CVD).[1–4] While clinical TG measures are commonly assessed after an overnight fast, it should be acknowledged that individuals spend most of the day in a postprandial state because of frequent food intake.[5] The typical eating pattern in Western society consists of 3 relatively regularly scheduled meals per day interspersed with more variable eating events. This eating pattern prevents serum TG to reach fasting levels for most of the day.[6] Therefore, postprandial serum TG

Correspondence to: Dorina Ibi, MSc, Department of Human Genetics, Leiden University Medical Center, S4-P, Postbus 9600, 2300 RC Leiden, the Netherlands. Email d.ibi@lumc.nl

The Data Supplement is available at https://www.ahajournals.org/doi/suppl/10.1161/CIRCGEN.119.002s693.

For Sources of Funding and Disclosures, see page 296.

Circulation: Genomic and Precision Medicine is available at www.ahajournals.org/journal/circgen

Nonstandard Abbreviations and Acronyms

CVD	cardiovascular disease
GWAS	genome-wide association studies
HDL	high-density lipoprotein
HL	hepatic lipase
IDL	intermediate-density lipoprotein
LD	linkage disequilibrium
LDL	low-density lipoprotein
NEO	Netherlands Epidemiology of Obesity
NMR	nuclear magnetic resonance
TRL	TG-rich lipoprotein
VLDL	very low-density lipoprotein

concentrations may provide additional insight in CVD risk and the metabolic capacity of an individual to deal with dietary stimuli.[7–9]

The postprandial state is a dynamic, nonsteady-state condition that is characterized by increased serum levels of TG and TRLs (triglyceride-rich lipoproteins), which mainly include chylomicrons, but also VLDLs (very low-density lipoproteins), and their remnants. Cell culture studies and epidemiological data indicate that TRLs and particularly their remnants exert adverse effects on endothelial cells and may penetrate into the subendothelial space to initiate atherosclerotic plaques.[6,10] Large prospective studies have shown that delayed clearance of TRLs and their remnants from the blood stream is associated with atherosclerosis progression and therefore is a risk factor for CVD.[1–3,11] For example, the Copenhagen City Heart Study reported a significant association between higher nonfasting TG concentrations and higher myocardial infarction risk over 31 years of follow-up.[12]

In cohort studies, the postprandial TG response has been shown to be highly variable.[5,6] This interindividual variability is at least partly genetic, although dietary pattern, physical activity, alcohol consumption, age, sex, and insulin sensitivity are more than likely involved.[6,13,14] One cross-sectional study reported that healthy sons of men with established coronary artery disease have higher postprandial serum TG concentrations after consumption of a high-fat meal compared with sons of control subjects without CVD,[14] which suggested that prolonged postprandial hypertriglyceridemia is associated with familial risk for CVD. Multiple genetic variants in the *APOA1/C3/A4/A5* gene cluster have been associated with postprandial lipemia in relatively small studies.[15–17] Thus far, a limited number of genome-wide association studies (GWAS) on postprandial TG concentrations have been performed. These include the HAPI (Heredity and Phenotype Intervention) Heart study in Old Order Amish individuals (n=809), identifying a rare null mutation in *APOC3,* likely due to a founder effect.[18] In addition, a study embedded in the Genetics of Lipid-Lowering Drugs and Diet Network (GOLDN), consisting of 1715 participants, yielded evidence for association of a variant (rs964184) in the vicinity of the APOA1/C3/A4/A5 gene cluster. However, this association was attenuated upon additional statistical adjustment for fasting TG.[19]

In the present study, we set out to elucidate the genetics and underlying biology of the postprandial TG response to a liquid mixed meal. To this end, we performed a GWAS and a subsequent in-depth assessment of nuclear magnetic resonance (NMR)–based metabolite responses of the identified genomic loci in 5630 middle-aged individuals of the NEO (Netherlands Epidemiology of Obesity) study.

METHODS

Complete methods are available in the Data Supplement. The NEO study design was approved by the medical ethics committee of the Leiden University Medical Center, and all participants gave their written informed consent. Because of consent issues, we cannot make the individual data of NEO study participants available to other researchers for purposes of reproducing the results or replicating the procedure. However, a file including GWAS summary statistics can be requested via k.willems_van_dijk@lumc.nl.

RESULTS

Population Characteristics

Characteristics of the NEO study population as well as for the separate Leiden and Leiderdorp subcohorts are summarized in Table 1. The mean age in the total population was 56 years, and 48.5% of the population were men. Compared with the Leiderdorp subcohort, participants in the Leiden subcohort were more frequently men (49.9% versus 44.3%), had a higher mean body mass index (31.3 versus 26.3 kg/m^2), used more frequently lipid-lowering drugs (17.6% versus 10.4%), had higher fasting and postprandial serum TG concentrations (1.34 versus 1.00 mmol/L, respectively, for fasting TG; and 2.01 versus 1.63 mmol/L, respectively, for TG at 150 minutes). Compared with the Leiderdorp subcohort, the median untransformed TG response at 150 minutes was somewhat lower in the Leiden subcohort (0.06 [IQR, −0.18 to 0.30] and −0.03 [IQR, −0.26 to 0.22] mmol/L, respectively, as calculated using the nonlinear prediction model between fasting and postprandial TG levels) but showed overlapping and wide interquartile ranges.

Genome-Wide Association Analysis

The results for the fasting and postprandial TG GWAS analyses in the total cohort are shown in Figure 1. Genetic

Table 1. Characteristics of the Total, Leiden, and Leiderdorp Cohorts From the NEO Study

Characteristics	Total Cohort (N=5630)	Leiden Subcohort (N=4192)	Leiderdorp Subcohort (N=1438)
Number of participants	5630	4192	1438
Age, y	56.0 (5.9)	55.9 (5.9)	56.1 (6.0)
Men	48.5%	49.9%	44.3%
BMI, kg/m^2	30.0 (4.8)	31.2 (4.2)	26.2 (4.5)
Lipid-lowering drug users	15.8%	17.7%	10.4%
Fasting TG, mmol/L	1.25 (0.88–1.77)	1.34 (0.95–1.87)	1.00 (0.71–1.45)
Postprandial TG at 30 min, mmol/L Postprandial TG_at 150 min, mmol/L	1.43 (1.04–1.95) 1.91 (1.37–2.64)	1.51 (1.12–2.06) 2.01 (1.47–2.72)	1.20 (0.89–1.67) 1.63 (1.14–2.30)
TG response, mmol/L	−0.008 (−0.24–0.24)	−0.03 (−0.26–0.22)	0.06 (−0.18–0.30)

Results are presented as median (inter quartile range) for not normally distributed data, mean (SD), or number (percentage). TG response: residuals at 150 min. BMI indicates body mass index; NEO, Netherlands Epidemiology of Obesity; and TG, triglyceride.

variants mapping to the known TG loci near *APOA1, LPL, APOE, GCKR,* and *CILP2* showed genome-wide significant signals in both fasting and postprandial TG concentrations. Interestingly, the signals in the postprandial analyses completely mirrored the signals of the fasting analyses, although there is some variation in the *P*-values that were reached in the separate analyses. Locus *MLXIPL* on chromosome 7 was borderline significant in the fasting TG GWAS, but achieved a *P*-value of 1.2×10^{-10} in the postprandial TG analyses. However, both effect sizes for rs71556736-T (the top hit in *MLXIPL* locus) were similar (fasting, β [SE]=−0.14 [0.03] SD and postprandial, β [SE]=−0.17 [0.03] SD). The full list with the lead hits for both fasting and postprandial TG is given in Table I in the Data Supplement.

Figure 2 displays the Manhattan plot for the TG response residuals at 150 minutes in the total NEO cohort. On chromosome 15, rs7350789 reached a suggestive *P*-value of 5.1×10^{-8}. In addition to rs7350789, there were 6 independent variants with a suggestive *P*-value *(P*<5.0×10^{-6}, Table II in the Data Supplement), but here we pursued the variant with the strongest evidence for association, based on the high number of associated variants in strong linkage disequilibrium (LD). None of the loci identified for either fasting and postprandial TG levels showed evidence for association (*P*-values >0.05; Table III in the Data Supplement). The regional association plot is shown in Figure 3A, showing that rs7350789 is 23 kb upstream of the *LIPC*, which encodes HL (hepatic lipase). Rs7350789-A had a per-allele decrease (SE) in TG response residual of −0.11 (0.02) SD (Table 2, Figure 3B). Because the 2 subcohorts from the NEO study had different study characteristics at fasting, analyses on the rs7350789 variant were stratified accordingly. Given the differences in the median postprandial TG response residuals (Table 1), rs7350789-A in the *LIPC* locus still showed an effect in the same direction, although the magnitude of the effect was somewhat smaller in the Leiden subcohort compared with the Leiderdorp subcohort (β [SE]=−0.09 [0.02] SD and β [SE]=−0.15 [0.04] SD, respectively). Adjustment for body mass index and lipid-lowering medication did not change the results (β=−0.11 [0.019] SD and β=−0.11 [0.020] SD), respectively (Table IV in the

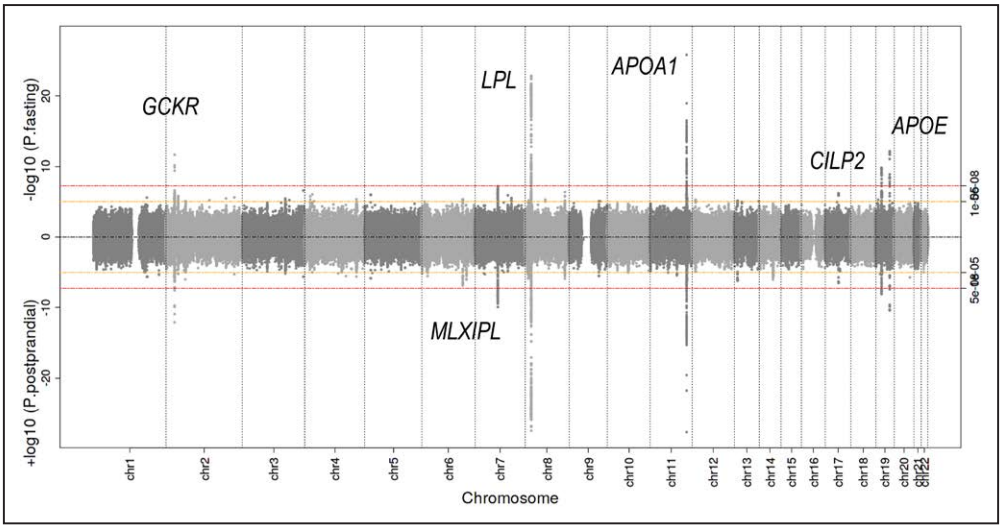

Figure 1. Miami plot showing P-values of the single-nucleotide polymorphism (SNP) associations with fasting TG and postprandial TG.

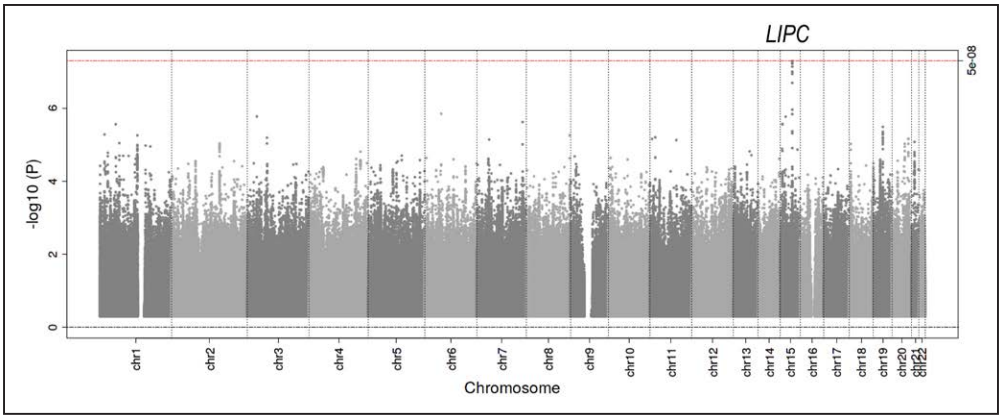

Figure 2. Manhattan plot of the GWAS of postprandial TG residuals at 150 min in the NEO Study.
x axis shows chromosomal positions. *y* axis shows -log10 *P-values. The red line indicates the* genome-wide significance threshold (P=5.0×10^{-8}).

Data Supplement). Furthermore, effect sizes were similar in men and women (β=−0.11 [0.030] SD and β=−0.10 [0.026] SD, respectively). Rs7350789-A, mapping to *LIPC,* was not associated with either fasting TG levels (β [SE]=0.031 [0.019] SD, *P*-value=0.1) or postprandial TG levels (β [SE]=−0.010 [0.019] SD, *P*-value=0.6).

NMR-Based Metabolomics

Based on the single signal of the postprandial residual TG GWAS in the full cohort, we conducted association analyses between rs7350789-A in *LIPC* and residuals of the postprandial response of 149 NMR-based metabolite measures at T=150 (Figure 4, Table V in the Data Supplement). Rs7350789-A was significantly associated with postprandial responses of 33 residuals of metabolite measures (*P*-value <1.34×10^{-3}). Overall, the rs7350789-A allele was associated with a smaller increase of the TG-component in almost all HDL (high-density lipoprotein) subparticles (largest effect on HDL-TG, *P*-value=4.5×10^{-7}), a smaller decrease in HDL diameter (*P*-value=2.8×10^{-4}), and a smaller increase of most components of VLDL subparticles (largest effect on VLDL-cholesterol, *P*-value=2.7×10^{-6}). In addition, this variation in the *LIPC* locus was significantly associated with a smaller increase in plasma monounsaturated fatty acids and total fatty acids and a larger decrease of beta-hydroxybutyrate.

To gain additional insight in the role of rs7350789-A, we performed association analyses of this SNP with fasting and postprandial NMR-based metabolite levels (Figure IIA and IIB and Table VI in the Data Supplement). In the fasting state, rs7350789-A was associated with higher metabolite measures of many components of the larger HDL, LDL (low-density lipoprotein), IDL (intermediate-density lipoprotein), and very small VLDL particles. The strongest associations were observed for the TG-component in LDL and HDL, which were both significantly increased (*P*-value=3.39×10^{-57} and *P*-value=1.35×10^{-67}, respectively). In addition, both HDL and LDL diameter were significantly larger (*P*-value=3.21×10^{-36} and *P*-value=1.59×10^{-26}, respectively), whereas VLDL diameter was significantly smaller (*P*-value=9.49×10^{-09}). Association analyses of rs7350789-A with postprandial NMR-based metabolite levels were remarkably similar to the associations of rs7350789-A with fasting levels.

DISCUSSION

Our study aimed to identify genes independent of fasting TG concentrations that influence the postprandial TG response. To this end, we performed a GWAS of residuals of the TG response after taking into account the strong nonlinear correlation between fasting and postprandial TG concentrations. We identified rs7350789, mapping to *LIPC* that encodes for HL, to be associated with the residuals of TG response. Association analyses of postprandial metabolite response residuals and rs73509789-A showed that this variation in the *LIPC* locus is significantly associated with 33 metabolite measures: mainly with a smaller increase of the TG-component in almost all HDL subparticles, a smaller decrease of HDL diameter, and a smaller increase of most components of VLDL subparticles at 150 minutes after the meal.

The GWAS of fasting and postprandial TG concentrations in the NEO study revealed overlapping genetic loci, including the previously identified loci near *APOA1, LPL, APOE, GCKR, and CILP2.* However, the signals found in the postprandial TG level GWAS resulted in somewhat different *P*-values for the various loci as compared with the fasting TG GWAS. Locus *MLXIPL* on chromosome 7 was borderline significant in the fasting TG GWAS but achieved a *P*-value of 1.2×10^{-10} in the postprandial TG GWAS. Given the similarity of the effect sizes, the difference in *P*-values is likely because of power. In conclusion, the results indicate that the major loci that affect postprandial TG concentrations are the same as

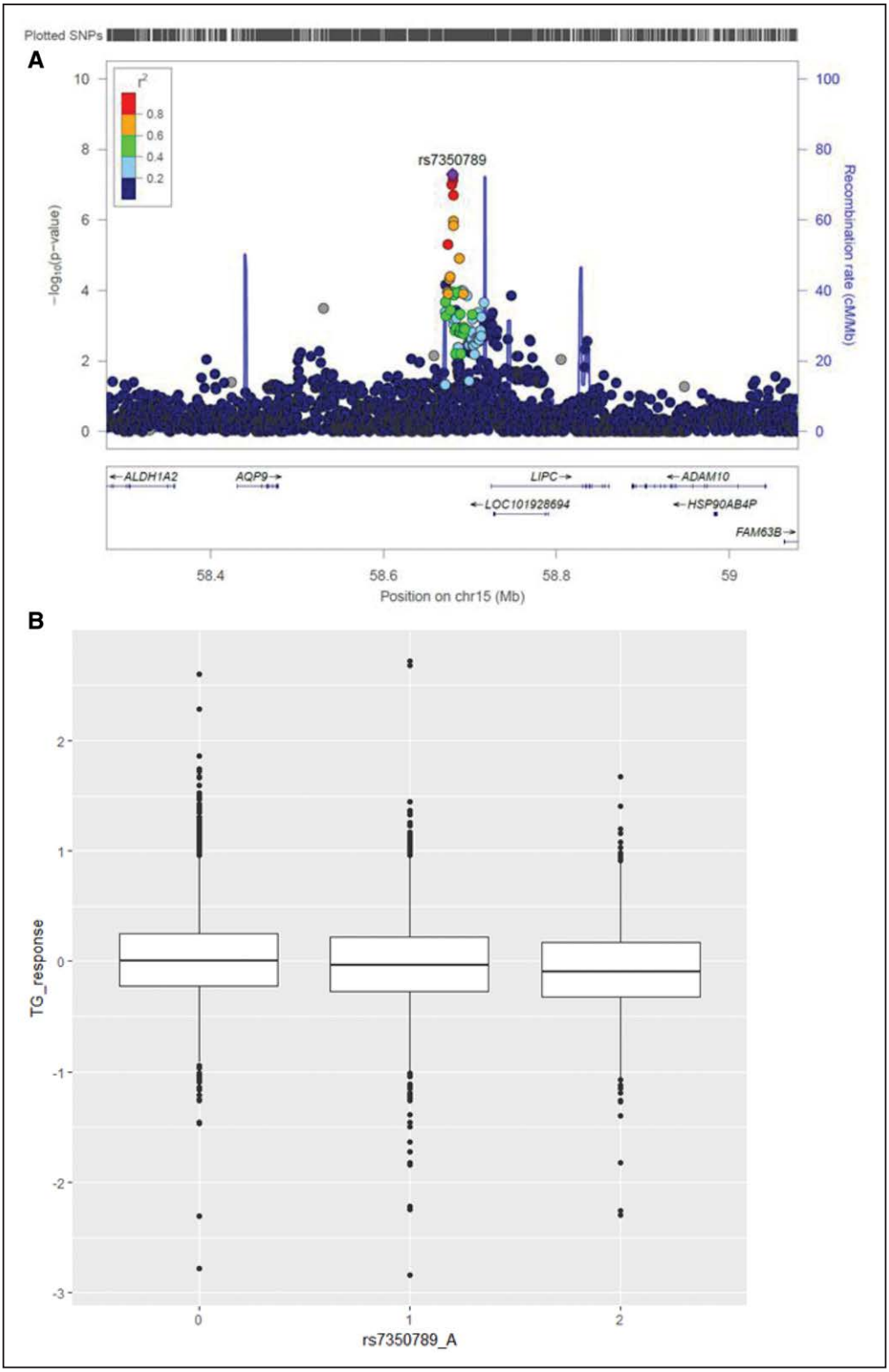

Figure 3. *LIPC* and TG response.
A, Regional association plot for *LIPC* on chromosome 15. The purple diamond represents the lead SNP for the locus. **B**, Rs7350789-A genotype specific effects on TG response measured as residuals at 150 min.

those that play a major role in determining fasting TG concentrations.

In previous large-scale GWAS meta-analyses performed by the Global Lipids Genetics Consortium, rs1532085 near the *LIPC* locus was associated with TG concentrations.[21] Interestingly, this SNP is 3.7 kb upstream from the residual TG response SNP rs7350789 and the LD of rs1532085 and rs7350789 is high

Table 2. Summary Statistics of the Association of the Suggestive (Borderline) *LIPC* SNP and TG Response in Response to a Mixed Meal in NEO

							Total Cohort		Leiden Cohort		Leiderdorp Cohort	
CHR	SNP	Position	Gene	EA/ NEA	EAF	Imputation Quality	β* (SE)	*P*-Value	β* (SE)	*P*-Value	β* (SE)	*P*-Value
Suggestive SNP												
15	rs7350789	58679668	LIPC	A/G	0.36	1	−0.11 (0.02)	5.1×10^{-8}	−0.09 (0.02)	9.2×10^{-5}	−0.15 (0.04)	5.2×10^{-5}

Threshold for genome-wide significance is 5×10^{-8}. Threshold for the suggestive signals is 1×10^{-6}. CHR indicates chomosome; EAF, effect allele frequency; (N)EA, (non) effect allele; and SNP, single nucleotide polymorphism.

*Beta coefficient expressed in SD units.

(R^2=0.81). However, in our GWAS of fasting and postprandial TG, this *LIPC* SNP was not a significant hit. This is likely because of power problems, because the Global Lipids Genetics Consortium meta-analysis consisted of many tens of thousands of individuals. The effect size of rs7350789-A in the total NEO cohort was −0.11 SD per additional risk allele, indicating a smaller increase of serum TG after a meal in carriers of this allele. Sensitivity analyses stratified by NEO subcohort (Leiden and Leiderdorp) and additional adjustment for body mass index and lipid-lowering medication did not materially change this result. Furthermore, although studies have shown differences in activity of HL in men and women,[22,23] we did not observe differences in the effect size of rs7359789-A on TG response residuals in men and women. These data indicate that although the *P*-value did not reach the genome wide threshold of 5.0×10^{-8}, the association of rs7350789 with the TG response residuals is robust.

As rs7350789 is not in the coding region of a gene, we assessed whether this SNP was in an expression quantitative trait locus for liver gene expression levels. A recent mega-analysis of liver expression quantitative trait locus identified rs11853674-G as an expression quantitative trait locus for *LIPC* with a negative beta (β [SE]=−0.51 [0.11]).[24] Rs7350789-A is in moderate LD with rs11853674-G (R^2=0.46) and thus associated with decreased expression of *LIPC* and likely reduced hepatic production of HL. In addition, recent genetic association analyses of HL activity showed that rs10468017-T near the *LIPC* gene is associated with decreased HL activity.[25] Rs10468017-T is in strong LD with rs7350789-A (R2=0.74), and this further corroborates that rs7350789-A reduces hepatic production of HL.

The *LIPC* gene is expressed in the liver, and the majority of the secreted HL is associated with cell surface proteoglycans, which keep HL in an enzymatically inactive form. Cell surface bound HL is exchanged with circulating HDL and under postprandial conditions the HDL-associated HL is presumably activated by changes in the composition of the HDL. The activated HL is thought to hydrolyze TG from chylomicron remnants, IDL and HDL.[26,27] The results from the NMR-analyses showed that in the fasting state, rs7350789-A was not associated with total TG levels, but with the redistribution of TG over the different lipoprotein fractions, mainly from VLDL particles to the larger HDL, LDL, IDL, and very small VLDL particles (Figure IIA in the Data Supplement), which is concordant with a previous report.[28] This redistribution is nearly identical in the postprandial state (Figure IIB in the Data Supplement), and this likely reflects the lagging effect of the associations in the fasting state. These data indicate that, apparently in the fasting state, lower *LIPC* gene expression is associated with higher TG-content in both HDL and LDL sized particles. Moreover, these data indicate that HL is functional in the fasting state.

As rs7350789-A is also associated with higher plasma levels of HDL particles, this HDL could function as an increased circulating reservoir of liver-derived HL. This increased reservoir of HDL-associated HL is activated after the meal and could explain the smaller than expected increase of TG in HDL particles and smaller than expected increase of the larger VLDL particles. Our results from the NMR-analysis support this role of HL in hydrolyzing TG in HDL particles after a meal, since rs7350789-A is associated with a smaller increase of the postprandial TG concentration in HDL subparticles. The meal thus specifically seems to activate the HDL-TG hydrolysis activity of HDL-associated HL.

In addition, rs7350789-A is associated with a smaller increase of most VLDL subparticles, but not specifically VLDL-TG or IDL-TG. Moreover, no effect was found in the responses of any of the LDL subparticles. These data indicate that in addition to a role for HL in decreasing HDL-TG, HL plays a role in lipoprotein metabolism that is independent of its lipolytic function. In vitro as well in vivo experiments have suggested that HL can act as a bridge between lipoproteins and the cell surface, promoting cellular uptake of lipoproteins and HDL.[29,30] Our NMR data indicate that, independent of lipolytic activity, HL indeed seems to play a role in the clearance of VLDL subparticles irrespective of the particle size. The meal thus seems to uncover a role for HL in whole particle VLDL clearance. Our data indicate that HL has different roles in lipid and lipoprotein metabolism in the fasting versus the postprandial state.

In parallel, rs7350789-A was associated with a smaller than expected decrease of the HDL diameter. Although this effect is very small, apparently, TG-content in HDL particles plays a role in their

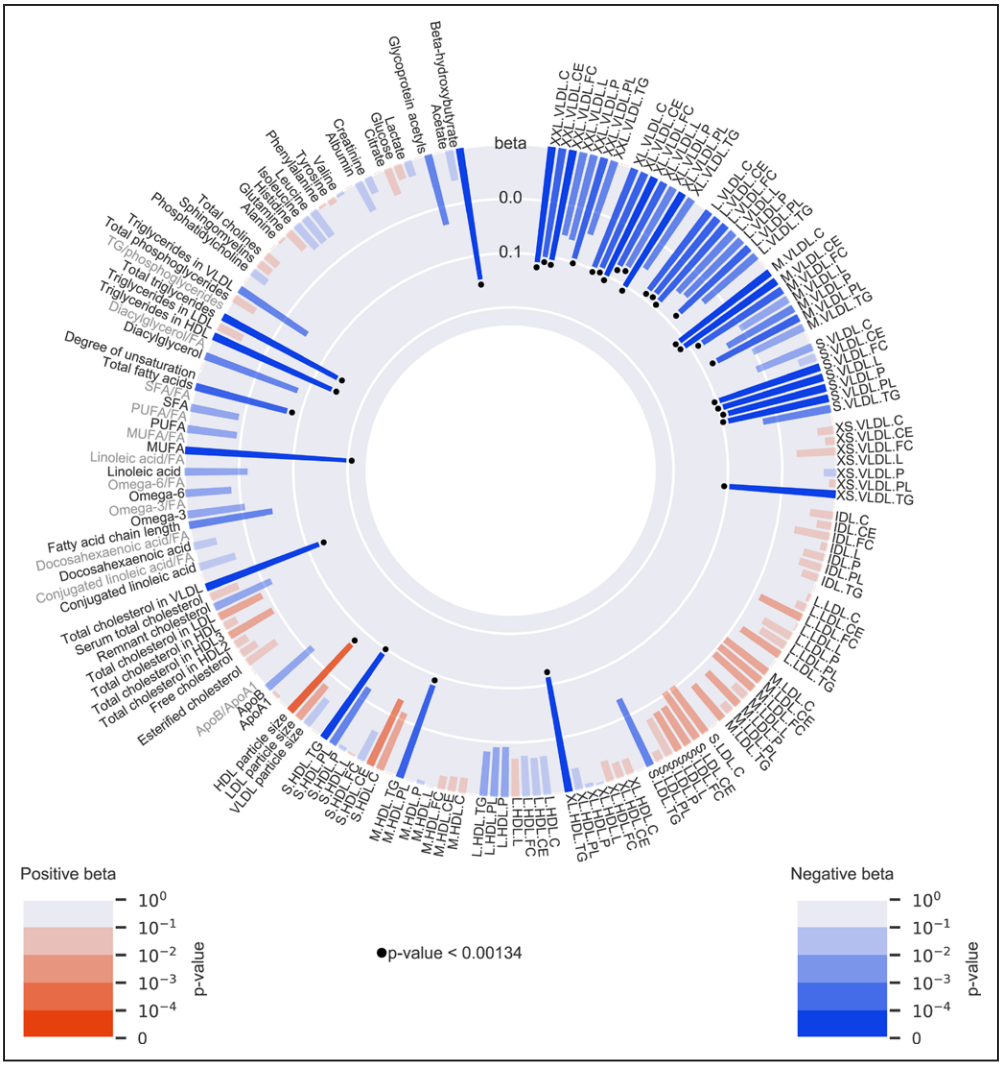

Figure 4. The associations of rs7350789-A and residuals of postprandial NMR-based metabolite measures.
Bar heights represent the magnitude of the beta coefficient from linear regression, which is expressed in SD (SD) units. Red bars indicate positive betas and blue bars indicate negative betas. The transparency of the bars indicates the level of statistical significance. A *P-value* <0.00134 is regarded statistical significant, as represented by the black dots. Full names for the NMR-based metabolite measures are reported in Blauw et al.[20]

size distribution. Whether this is a direct effect due to changes in HL activity or an indirect effect due to changes in substrate specificity of the HDL particles themselves remains to be determined.

Surprisingly, the association of rs7350789-A with NMR metabolite residuals showed a larger decrease of the postprandial beta-hydroxybutyrate concentration. This ketone body is connected to the oxidation of fatty acids.[31] Possibly, either as a result of the smaller increase of serum HDL-TG or the increased clearance of VLDL-particles, fatty acid oxidation is decreased. The pathophysiological mechanism underlying the association of variation in the *LIPC* locus and beta-hydroxybutyrate remains to be investigated. Other findings of interest for follow-up studies are the associations between the *LIPC* locus and the smaller increase of postprandial monounsaturated fatty acids and total fatty acids levels.

Our results show that carriers of the rs7350789-A allele near *LIPC* have a smaller than expected increase of TG after a meal. However, in the fasting state, the same variant is associated with redistribution of TG from VLDL to HDL and LDL. Since these associations represent both pro- and anti-atherogenic effects, it is difficult to predict the CVD risk in carriers of the rs7350789-A genetic variation. A lookup of rs7350789-A in relation to coronary artery disease in publicly available data of the CARDIoGRAMplusC4D consortium[32] did not reveal evidence for an association (β [SE]=–0.015 [0.009], *P*-value=0.12). Given the small absolute effect of rs7350789-A on the postprandial TG response, this may not be surprising. Although our variant has not been associated with CVD risk, recent Mendelian Randomization analyses showed that the LDL TG raising alleles rs1800588-T and rs10468017-T near *LIPC* were

associated with lower HL activity and increased CVD risk in CARDIoGRAMplusC4D.[25] As indicated above, rs10468017 is in LD with our *LIPC* variant rs7350789. Decomposing the Mendelian Randomization analyses revealed that the major contribution of the *LIPC* locus to coronary artery disease risk was contributed by rs1800588, which was not in LD with rs7350789 and thus explains the lack of association of rs7350789 with coronary artery disease (rs10468017-T: β [SE]=–0.015 [0.011], *P*-value=0.15; rs1800588-T: β [SE]=–0.038 [0.011], *P*-value=0.0005).

One of the strengths of the NEO study is the mixed meal-response design that allows for standardized assessment of postprandial meal responses. Many studies that include nonfasting measures have no information on the timing and composition of the last meal before the blood draw. It should be noted, however, that a limitation of the study is that the last postprandial measurement was performed 150 minutes after the meal intake, which may not be the peak of the postprandial TG excursion. Previous research indicated that serum TG may peak up to 3 to 4 hours after meal ingestion.[3,33]

In conclusion, we performed a GWAS to identify loci involved in the TG response following a mixed-meal and to provide insight into postprandial lipoprotein metabolism by a metabolomics analysis, using the identified loci. We identified a variant near *LIPC* as a contributor to postprandial TG and lipoprotein metabolism independent of fasting TG concentrations, mainly affecting HDL and VLDL subparticles. In addition, we showed that the same loci that affect fasting TG concentrations also play a major role in determining postprandial TG concentrations.

ARTICLE INFORMATION

Received July 19, 2019; accepted May 26, 2020.

Affiliations

Department of Human Genetics (D.I., J.B.v.K., K.W.v.D.), Division of Gerontology and Geriatrics, Department of Internal Medicine (R.N., D.v.H.), Department of Clinical Epidemiology (R.L.-G., R.d.M., D.O.M.-K., F.R.R.), Department of Cardiology (S.T., J.W.J.), Department of Public Health & Primary Care (D.O.M.-K.), Division of Endocrinology, Department of Internal Medicine (J.B.v.K., L.L.B., P.C.N.R., K.W.v.D.), and Einthoven Laboratory for Experimental Vascular Medicine, Leiden University Medical Center (P.C.N.R., K.W.v.D.). National Institute for Public Health and the Environment (RIVM), Bilthoven, the Netherlands (D.I., M.E.T.D.).

Acknowledgments

We express our gratitude to all individuals who participate in the study. We are grateful to all participating general practitioners for inviting eligible participants. We furthermore thank P. van Beelen and all research nurses for collecting the data and P. Noordijk and her team for sample handling and storage and I. de Jonge, MSc for data management of the NEO study. In addition, we thank Alexander Blauw for writing the Python script to design the circular figure.

Sources of Funding

The NEO study is supported by the participating Departments, the Division and the Board of Directors of the Leiden University Medical Centre, and by the Leiden University, Research Profile Area "Vascular and Regenerative Medicine." Dr van Heemst was supported by the European Commission funded project HUMAN (Health-2013-INNOVATION-1-602757). The authors acknowledge the support from The Netherlands Cardiovascular Research Initiative: an initiative with support of the Dutch Heart Foundation (CVON2014-02 ENERGISE).

Disclosures

None.

REFERENCES

1. Nordestgaard BG, Benn M, Schnohr P, Tybjaerg-Hansen A. Nonfasting triglycerides and risk of myocardial infarction, ischemic heart disease, and death in men and women. *JAMA.* 2007;298:299–308. doi: 10.1001/jama.298.3.299
2. Bansal S, Buring JE, Rifai N, Mora S, Sacks FM, Ridker PM. Fasting compared with nonfasting triglycerides and risk of cardiovascular events in women. *JAMA.* 2007;298:309–316. doi: 10.1001/jama.298.3.309
3. Pirillo A, Norata GD, Catapano AL. Postprandial lipemia as a cardiometabolic risk factor. *Curr Med Res Opin.* 2014;30:1489–1503. doi: 10.1185/03007995.2014.909394
4. Alcala-Diaz JF, Delgado-Lista J, Perez-Martinez P, Garcia-Rios A, Marin C, Quintana-Navarro GM, Gomez-Luna P, Camargo A, Almaden Y, Caballero J, et al. Hypertriglyceridemia influences the degree of postprandial lipemic response in patients with metabolic syndrome and coronary artery disease: from the CORDIOPREV study. *PLoS One.* 2014;9:e96297. doi: 10.1371/journal.pone.0096297
5. Rosenson RS, Davidson MH, Hirsh BJ, Kathiresan S, Gaudet D. Genetics and causality of triglyceride-rich lipoproteins in atherosclerotic cardiovascular disease. *J Am Coll Cardiol.* 2014;64:2525–2540. doi: 10.1016/j.jacc.2014.09.042
6. Lopez-Miranda J, Williams C, Lairon D. Dietary, physiological, genetic and pathological influences on postprandial lipid metabolism. *Br J Nutr.* 2007;98:458–473. doi: 10.1017/S000711450774268X
7. Nakamura A, Monma Y, Kajitani S, Noda K, Nakajima S, Endo H, Takahashi T, Nozaki E. Effect of glycemic state on postprandial hyperlipidemia and hyperinsulinemia in patients with coronary artery disease. *Heart Vessels.* 2016;31:1446–1455. doi: 10.1007/s00380-015-0757-y
8. Boquist S, Ruotolo G, Tang R, Björkegren J, Bond MG, de Faire U, Karpe F, Hamsten A. Alimentary lipemia, postprandial triglyceride-rich lipoproteins, and common carotid intima-media thickness in healthy, middle-aged men. *Circulation.* 1999;100:723–728. doi: 10.1161/01.cir.100.7.723
9. Sharrett AR, Chambless LE, Heiss G, Paton CC, Patsch W. Association of postprandial triglyceride and retinyl palmitate responses with asymptomatic carotid artery atherosclerosis in middle-aged men and women. The Atherosclerosis Risk in Communities (ARIC) Study. *Arterioscler Thromb Vasc Biol.* 1995;15:2122–2129. doi: 10.1161/01.atv.15.12.2122
10. Hyson D, Rutledge JC, Berglund L. Postprandial lipemia and cardiovascular disease. *Curr Atheroscler Rep.* 2003;5:437–444. doi: 10.1007/s11883-003-0033-y
11. Stampfer MJ, Krauss RM, Ma J, Blanche PJ, Holl LG, Sacks FM, Hennekens CH. A prospective study of triglyceride level, low-density lipoprotein particle diameter, and risk of myocardial infarction. *JAMA.* 1996;276:882–888.
12. Langsted A, Freiberg JJ, Tybjaerg-Hansen A, Schnohr P, Jensen GB, Nordestgaard BG. Nonfasting cholesterol and triglycerides and association with risk of myocardial infarction and total mortality: the Copenhagen City Heart Study with 31 years of follow-up. *J Intern Med.* 2011;270:65–75. doi: 10.1111/j.1365-2796.2010.02333.x
13. Tiret L, Gerdes C, Murphy MJ, Dallongeville J, Nicaud V, O'Reilly DS, Beisiegel U, De Backer G. Postprandial response to a fat tolerance test in young adults with a paternal history of premature coronary heart disease - the EARS II study (European Atherosclerosis Research Study). *Eur J Clin Invest.* 2000;30:578–585. doi: 10.1046/j.1365-2362.2000.00674.x
14. Uiterwaal CS, Grobbee DE, Witteman JC, van Stiphout WA, Krauss XH, Havekes LM, de Bruijn AM, van Tol A, Hofman A. Postprandial triglyceride response in young adult men and familial risk for coronary atherosclerosis. *Ann Intern Med.* 1994;121:576–583. doi: 10.7326/0003-4819-121-8-199410150-00004
15. Ordovas JM, Cassidy DK, Civeira F, Bisgaier CL, Schaefer EJ. Familial apolipoprotein A-I, C-III, and A-IV deficiency and premature atherosclerosis due to deletion of a gene complex on chromosome 11. *J Biol Chem.* 1989;264:16339–16342.
16. Marín C, López-Miranda J, Gómez P, Paz E, Pérez-Martínez P, Fuentes F, Jiménez-Perepérez JA, Ordovás JM, Pérez-Jiménez F. Effects of the human apolipoprotein A-I promoter G-A mutation on postprandial lipoprotein metabolism. *Am J Clin Nutr.* 2002;76:319–325. doi: 10.1093/ajcn/76.2.319
17. Calabresi L, Cassinotti M, Gianfranceschi G, Safa O, Murakami T, Sirtori CR, Franceschini G. Increased postprandial lipemia in Apo A-IMilano carriers. *Arterioscler Thromb.* 1993;13:521–528. doi: 10.1161/01.atv.13.4.521

18. Pollin TI, Damcott CM, Shen H, Ott SH, Shelton J, Horenstein RB, Post W, McLenithan JC, Bielak LF, Peyser PA, et al. A null mutation in human APOC3 confers a favorable plasma lipid profile and apparent cardioprotection. *Science.* 2008;322:1702–1705. doi: 10.1126/science.1161524
19. Wojczynski MK, Parnell LD, Pollin TI, Lai CQ, Feitosa MF, O'Connell JR, Frazier-Wood AC, Gibson Q, Aslibekyan S, Ryan KA, et al. Genome-wide association study of triglyceride response to a high-fat meal among participants of the NHLBI Genetics of Lipid Lowering Drugs and Diet Network (GOLDN). *Metabolism.* 2015;64:1359–1371. doi: 10.1016/j.metabol.2015.07.001
20. Blauw LL, Noordam R, Soidinsalo S, Blauw CA, Li-Gao R, de Mutsert R, Berbée JFP, Wang Y, van Heemst D, Rosendaal FR, et al. Mendelian randomization reveals unexpected effects of CETP on the lipoprotein profile. *Eur J Hum Genet.* 2019;27:422–431. doi: 10.1038/s41431-018-0301-5
21. Willer CJ, Schmidt EM, Sengupta S, Peloso GM, Gustafsson S, Kanoni S, Ganna A, Chen J, Buchkovich ML, Mora S, et al; Global Lipids Genetics Consortium. Discovery and refinement of loci associated with lipid levels. *Nat Genet.* 2013;45:1274–1283. doi: 10.1038/ng.2797
22. Sorva R, Kuusi T, Dunkel L, Taskinen MR. Effects of endogenous sex steroids on serum lipoproteins and postheparin plasma lipolytic enzymes. *J Clin Endocrinol Metab.* 1988;66:408–13.
23. Feitosa MF, Myers RH, Pankow JS, Province MA, Borecki IB. LIPC variants in the promoter and intron 1 modify HDL-C levels in a sex-specific fashion. *Atherosclerosis.* 2009;204:171–177. doi: 10.1016/j.atherosclerosis.2008.09.007
24. Strunz T, Grassmann F, Gayán J, Nahkuri S, Souza-Costa D, Maugeais C, Fauser S, Nogoceke E, Weber BHF. A mega-analysis of expression quantitative trait loci (eQTL) provides insight into the regulatory architecture of gene expression variation in liver. *Sci Rep.* 2018;8:5865. doi: 10.1038/s41598-018-24219-z
25. Silbernagel G, Scharnagl H, Kleber ME, Delgado G, Stojakovic T, Laaksonen R, Erdmann J, Rankinen T, Bouchard C, Landmesser U, et al. LDL triglycerides, hepatic lipase activity, and coronary artery disease: an epidemiologic and Mendelian randomization study. *Atherosclerosis.* 2019;282:37–44. doi: 10.1016/j.atherosclerosis.2018.12.024
26. Deeb SS, Zambon A, Carr MC, Ayyobi AF, Brunzell JD. Hepatic lipase and dyslipidemia: interactions between genetic variants, obesity, gender and diet running title: hepatic lipase regulation and dyslipidemia. *JLR Papers In Press.* 2003;44:1279–1286.
27. Santamarina-Fojo S, González-Navarro H, Freeman L, Wagner E, Nong Z. Hepatic lipase, lipoprotein metabolism, and atherogenesis. *Arterioscler Thromb Vasc Biol.* 2004;24:1750–1754. doi: 10.1161/01.ATV.0000140818.00570.2d
28. Kettunen J, Demirkan A, Würtz P, Draisma HH, Haller T, Rawal R, Vaarhorst A, Kangas AJ, Lyytikäinen LP, Pirinen M, et al. Genome-wide study for circulating metabolites identifies 62 loci and reveals novel systemic effects of LPA. *Nat Commun.* 2016;7:11122. doi: 10.1038/ncomms11122
29. Zambon A, Deeb SS, Bensadoun A, Foster KE, Brunzell JD. *In vivo* evidence of a role for hepatic lipase in human apoB-containing lipoprotein metabolism, independent of its lipolytic activity. *J Lipid Res.* 2000;41:2094–2099.
30. Freeman L, Amar MJ, Shamburek R, Paigen B, Brewer HB Jr, Santamarina-Fojo S, González-Navarro H. Lipolytic and ligand-binding functions of hepatic lipase protect against atherosclerosis in LDL receptor-deficient mice. *J Lipid Res.* 2007;48:104–113. doi: 10.1194/jlr.M600321-JLR200
31. Benjamin B, Wada Y, Grundy SM, Szuszkiewicz-Garcia M, Vega GL. Fatty acid oxidation in normotriglyceridemic men. *J Clin Lipidol.* 2016;10:283–288. doi: 10.1016/j.jacl.2015.11.018
32. Nikpay M, Goel A, Won HH, Hall LM, Willenborg C, Kanoni S, Saleheen D, Kyriakou T, Nelson CP, Hopewell JC, et al. A comprehensive 1,000 Genomes-based genome-wide association meta-analysis of coronary artery disease. *Nat Genet.* 2015;47:1121–1130. doi: 10.1038/ng.3396
33. Borén J, Matikainen N, Adiels M, Taskinen MR. Postprandial hypertriglyceridemia as a coronary risk factor. *Clin Chim Acta.* 2014;431:131–142. doi: 10.1016/j.cca.2014.01.015