METABOLISM CLINICAL AND EXPERIMENTAL 64 (2015) 1359–1371

Available online at www.sciencedirect.com

Metabolism

www.metabolismjournal.com

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)

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ARTICLE INFO

Article history:
Received 4 December 2014
Accepted 1 July 2015

Keywords:
Triglyceride
Postprandial lipemia
Gwas
Genetics

ABSTRACT

Objective. The triglyceride (TG) response to a high-fat meal (postprandial lipemia, PPL) affects cardiovascular disease risk and is influenced by genes and environment. Genes involved in lipid metabolism have dominated genetic studies of PPL TG response. We sought to elucidate common genetic variants through a genome-wide association (GWA) study in the Genetics of Lipid Lowering Drugs and Diet Network (GOLDN).

Methods. The GOLDN GWAS discovery sample consisted of 872 participants within families of European ancestry. Genotypes for 2,543,887 variants were measured or imputed from HapMap. Replication of our top results was performed in the Heredity and Phenotype Intervention (HAPI) Heart Study (n = 843). PPL TG response phenotypes were constructed from plasma TG measured at baseline (fasting, 0 hour), 3.5 and 6 hours after a high-fat meal, using a random coefficient regression model. Association analyses were adjusted for

Abbreviations: GOLDN, Genetics of Lipid Lowering Drugs and Diet Network; TG, triglyceride; PPL, postprandial lipemia; GWA, genome-wide association; HAPI, Heredity and Phenotype Intervention; SNP, single nucleotide polymorphism; HDL, high density lipoprotein; CVD, cardiovascular disease; CHD, coronary heart disease; FamHS, Family Heart Study; AUC, area under the curve; AUI, area under the increase; PC, principal component; QQ, quartile-quartile; LD, linkage disequilibrium; EA, effect allele; EAF, effect allele frequency; Chr, chromosome; SE, standard error; SD, standard deviation.

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http://dx.doi.org/10.1016/j.metabol.2015.07.001
0026-0495/

covariates and principal components, as necessary, in a linear mixed model using the kinship matrix; additional models further adjusted for fasting TG were also performed. Meta-analysis of the discovery and replication studies (n = 1715) was performed on the top SNPs from GOLDN.

Results. GOLDN revealed 111 suggestive (p < 1E–05) associations, with two SNPs meeting GWA significance level (p < 5E–08). Of the two significant SNPs, rs964184 demonstrated evidence of replication (p = 1.20E–03) in the HAPI Heart Study and in a joint analysis, was GWA significant (p = 1.26E–09). Rs964184 has been associated with fasting lipids (TG and HDL) and is near *ZPR1* (formerly *ZNF259*), close to the *APOA1/C3/A4/A5* cluster. This association was attenuated upon additional adjustment for fasting TG.

Conclusion. This is the first report of a genome-wide significant association with replication for a novel phenotype, namely PPL TG response. Future investigation into response phenotypes is warranted using pathway analyses, or newer genetic technologies such as metabolomics.

1. Introduction

Fasting and postprandial plasma triglyceride (TG) levels are known risk factors for cardiovascular disease (CVD) [1–4]. Common genetic variants associated with fasting TG, assessed through single nucleotide polymorphisms (SNPs), have been studied extensively [5–10]. The results of a meta-analysis of lipid genome wide association (GWA) studies by Teslovich et al. [5] implicated 24 loci associated with fasting TG, with the most significant single nucleotide polymorphism (SNP), rs964184. While fasting TG remains the gold standard of TG measurement by physicians and is most studied in relation to CVD risk [11], humans spend a majority of their waking hours in the postprandial state [1,3,4,11].

Postprandial lipemia (PPL) encompasses the changes in plasma TG and lipid profile as well as the duration of these changes due to the ingestion of a high-fat meal [11]. An elevated or elongated PPL leads to the production of atherogenic TG-rich lipoproteins and activation of thrombotic processes [11]. Atherogenic TG-rich lipoproteins that remain in the circulation for extended periods of time are independently related to progression of coronary heart disease (CHD) and thus are a risk factor for CVD [3,11–15].

Meal challenges that induce PPL produce large and highly variable changes in circulating TG's and TG rich lipoproteins. Within a population the PPL TG response is more variable than fasting TG levels and is believed to be modulated by both genes and environment [15]. To date, there has been one genome-wide association study (GWAS) of PPL in an extended pedigree which found a rare mutation [16], while the majority of genetic PPL studies have investigated candidate genes with known involvement in lipid metabolism (i.e. *APOA1/C3/A4/A5* cluster, *ABCA1*, *CETP*, *GCKR*, *IL6*, *LPL*, *PLIN1*, *TCF7L2*, etc.) [17,18]. Some of these studies demonstrated genetic variation associated with PPL, however they suffer from small sample sizes, differing meal challenges, and limited replication [17].

We sought to elucidate the genetic determinants of PPL TG response to a standardized high-fat meal by performing a GWAS among participants from the Genetics of Lipid Lowering Drugs and Diet Network (GOLDN). We used the Hereditary and Phenotype Intervention (HAPI) Heart Study for replication of our top findings, and also performed a joint meta-analysis (discovery and replication results) for the top discovery findings from GOLDN.

2. Materials and Methods

2.1. Study Design

The GOLDN Study was designed to characterize the genetic basis of TG response to two environmental contexts: one to raise triglycerides (consumption of a high-fat meal, PPL); and one to lower TG (a 3-week treatment with 160 g/day of fenofibrate). The specific methodology of GOLDN is reported elsewhere [19]. The study population consisted of 189 families, recruited from 3-generational pedigrees at two genetically homogeneous European ancestry field centers of the NHLBI Family Heart Study (FamHS: Minneapolis, MN, and Salt Lake City, UT) [20]. Inclusion criteria were: ≥18 years of age, fasting triglycerides (TGs) <1500 mg/dL, willingness to participate in the study and attend the scheduled clinic exams, being part of a family with at least 2 members in a sibship, AST and ALT tests within normal range, and creatinine ≤2.0. Only subjects not using lipid-lowering agents (pharmaceuticals or nutraceuticals) for at least 4 weeks after the screening visit were eligible. The Institutional Review Boards at the University of Alabama at Birmingham, the University of Minnesota, the University of Utah, and Tufts University approved the study protocol. Informed consent was obtained on all participants.

This analysis focused on plasma TG concentration in response to the standardized high fat challenge (PPL; n = 872), which followed the protocol of Patsch et al. [21]. The caloric intake of the intervention meal was determined by body surface area, containing 700 kilocalories per m^2 of body surface area (2.93 MJ/m^2 body surface area). The meal composition was 83% of calories from fat, 14% from carbohydrates, and 3% from protein. The meal was formulated to have a cholesterol content of 240 mg and a polyunsaturated:saturated fat ratio of 0.06. Based on these guidelines, the average individual ingested 175 mL of heavy whipping cream (39.5% fat) combined with 7.5 mL powdered, instant, non-fat, dry milk, and blended with ice. To increase palatability of the drink, 15 mL of chocolate or