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Perspective

Leveraging Human Genetics to Understand the Relation of LDL Cholesterol with Type 2 Diabetes

Erik Ingelsson[1,2*] and Joshua W. Knowles[1,2]

In a recent issue of the *Journal of the American Medical Association*, Lotta and colleagues used human genetic data to address the link of LDL cholesterol (LDL-C)[3] with type 2 diabetes (T2D) *(1)*. They set out to investigate whether LDL-C–lowering alleles in or near genes encoding targets of 3 different lipid-lowering therapies (statins, PCSK9 inhibitors, and ezetimibe) were associated with risk of T2D. Human genetic studies have proven to be useful in predicting efficacy and adverse effects of perturbation of drug targets, such as cholesteryl ester transfer protein (CETP) and lipoprotein-associated phospholipase A2 (Lp-PLA2) inhibitors.

Previous studies have shown that LDL-C–lowering alleles in 3-hydroxy-3-methylglutaryl-CoA reductase (*HMGCR*),[4] the gene encoding the target of statins, are associated with lower risk of coronary heart disease (CHD), but also an increased risk of T2D. These observations are consistent with metaanalyses of randomized clinical trials of statins, which show that while statins decrease LDL-C concentrations and risk of cardiovascular events, they are also associated with a slightly increased risk of T2D. In contrast, individuals with familial hypercholesterolemia (FH) caused by mutations in low density lipoprotein receptor (*LDLR*) seem to be protected against T2D. Similarly, studies of variation in proprotein convertase subtilisin/kexin type 9 (*PCSK9*) have consistently predicted an effect of PCSK9 inhibitors on LDL-C–lowering and decreased risk of CHD, and a study published soon after the *JAMA* paper by Lotta et al. showed that *PCSK9* variants associated with lower LDL-C also were associated with higher fasting glucose, body weight, and waist-to-hip ratio, and an increased risk of T2D *(2)*. This observation contrasts to the initial (small) clinical trial data on PCSK9 inhibitors that have not shown significant effects on T2D risk. The observations of LDL-C–lowering alleles in both *HMGCR* and *PCSK9* being associated with T2D risk is especially intriguing in the light of our study published in 2015. In this study we used a larger set of LDL-C–increasing alleles (beyond those in or near known drug target genes) to investigate the associations of genetically determined lipid fractions with glucose and insulin metabolism and T2D risk and reported that lower genetically determined LDL-C was associated with higher fasting glucose and T2D risk *(3)*. These findings taken together indicate that there is a causal link between LDL-C concentrations per se and T2D, and that this is unlikely to be a statin-specific adverse effect as has been suggested in some prior literature.

However, while LDL-C–lowering alleles in or near NPC1 like intracellular cholesterol transporter 1 (*NPC1L1*), the molecular target of ezetimibe representing the third major mechanism by which LDL-C–lowering treatment acts, have been shown to be associated with lower risk of CHD, Lotta et al. were the first to establish associations of these alleles with risk of T2D. They did this by investigating associations of LDL-C–lowering genetic variants with T2D and CHD in large-scale metaanalyses including 50 775 individuals with T2D and 270 269 controls, and 60 801 individuals with CHD and 123 504 controls. Lotta et al. used data from EPIC (European Prospective Investigation into Cancer and Nutrition)-InterAct, UK Biobank, and DIAGRAM (DIAbetes Genetics Replication And Meta-analysis) for the T2D associations, and data from CARDIoGRAMplusC4D (Coronary ARtery DIsease Genome wide Replication and Meta-analysis plus The Coronary Artery Disease C4D), MAGIC (the Meta-Analyses of Glucose and Insulin-related traits Consortium), and GIANT (Genetic Investigation of ANthropometric Traits) for associations with CHD and other traits reflecting glucose metabolism and anthropometry. Apart from investigating LDL-C–lowering alleles in the *HMGCR*, *PCSK9*, and *NPC1L1* loci, they also assessed such alleles in 2 other LDL-C–related loci, specifically *LDLR* and ATP Binding Cassette Subfamily G Member 5 (*ABCG5*)/ATP Binding Cassette Subfamily G Member 8 (*ABCG8*).

Apart from establishing that LDL-C–lowering alleles in the *NPC1L1* locus are associated with T2D risk, we believe that the most important contribution of Lotta et al. is the comparison of effects across the different

[1] Department of Medicine, Division of Cardiovascular Medicine, [2] Cardiovascular Institute, Stanford University School of Medicine, Stanford, CA.
* Address correspondence to this author at: Stanford University, Cardiovascular Medicine, 300 Pasteur Dr., mail code 5773, Stanford, CA, 94305. E-mail eriking@stanford.edu.
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[3] Nonstandard abbreviations: LDL-C, LDL cholesterol; T2D, type 2 diabetes; CETP, cholesteryl ester transfer protein; Lp-PLA2, lipoprotein-associated phospholipase A2; CHD, coronary heart disease; FH, familial hypercholesterolemia.
[4] Human genes: *HMGCR,* 3-hydroxy-3-methylglutaryl-CoA reductase; *LDLR,* low density lipoprotein receptor; *PCSK9,* proprotein convertase subtilisin/kexin type 9; *NPC1L1,* NPC1 like intracellular cholesterol transporter 1; *ABCG5,* ATP Binding Cassette Subfamily G Member 5; *ABCG8,* ATP Binding Cassette Subfamily G Member 8.

alleles. While the effect on CHD was similar across the different alleles, with about 40% lower risk per 1 mmol/L predicted decrease in LDL-C, the effects of these alleles on T2D risk were more heterogeneous, with odds ratios ranging from 1.13 to 2.42. That said, it should be noted that the LDL-C–lowering alleles in all 3 loci with genes encoding drug targets currently used to treat hypercholesterolemia (*HMGCR*, *PCSK9*, *NPC1L1*) were significantly associated with higher risk of T2D. Furthermore, the effects were similar (odds ratios from 1.13 to 1.39 per 1 mmol/L predicted decrease in LDL-C) for all investigated alleles, except those in *NPC1L1* (the gene encoding the drug target of ezetimibe) that showed a larger effect size—which, however, could be a technical artifact due to the normalization procedure where a small technical or biological variation can be amplified. This is also consistent with another recent report that came out after the publication of Lotta et al. showing variants in *PCSK9* and *HMGCR* having about the same effect on the risk of cardiovascular events and T2D per unit decrease of LDL-C *(4)*. This, taken together with other evidence including our study across a larger number of LDL-C loci *(3)*, indicates that there is a more general inverse association between LDL-C and T2D risk, not only restricted to specific pathways. A consequence of this is that it is unlikely that the risk of T2D is restricted to certain LDL-C–lowering drug therapies, but that we should be prepared to accept that the lowering of LDL-C is associated with a slight increase in T2D risk, regardless of the means.

The results of Lotta et al. show that the LDL-C–lowering alleles in or near *NPC1L1* were associated with larger increases of T2D risk than the other alleles for a similar decrease in LDL-C, and although there certainly is a possibility that this difference could have arisen by chance, it raises the question whether ezetimibe is likely to have more prominent glucometabolic side effects than statins and PCSK9 inhibitors. It would certainly be useful to redo these genetic analyses when even larger datasets become available in a not too distant future to get more precise estimates, but ultimately, proof of the relationships between different mechanisms of LDL-C reduction and T2D risk is most likely to emerge from a combination of evidence from the ongoing large PCSK9 inhibitor outcomes trials, as well long-term follow-up of IMPROVE-IT (Improved Reduction of Outcomes: Vytorin Efficacy International Trial) and postmarketing surveillance data. It is also important to notice that ezetimibe is not as potent in LDL-C–lowering (on average LDL-C is only reduced by about 15%, or approximately 0.4 mmol/L), and because of this more modest LDL-C–lowering, the signal for T2D may be small. There are reasonably good arguments for why a similar reduction in LDL-C using different drugs acting on different pathways would also be associated with different risk of T2D. For example, while statins and PCSK9 inhibitors act primarily via effects on cholesterol metabolism in the liver, ezetimibe acts on cholesterol absorption in the intestine, thereby exerting effects on the enterohepatic circulation, which is increasingly recognized as a regulator of plasma LDL-C concentrations.

So, what should the practicing clinician make of this, and do these findings impact clinical management of hypercholesterolemia? For statins, the benefits from lowering the risk of CHD by far outweigh the adverse effects on T2D risk—even in individuals that develop diabetes. In a posthoc analysis of the JUPITER (Justification for the Use of Statins in Prevention: an Intervention Trial Evaluating Rosuvastatin) trial, statins accelerated the average time to diagnosis of diabetes by about 5 weeks for rosuvastatin as compared to placebo for those that developed T2D. Further, we know that the risk of incident T2D associated with statins is not the same for all individuals. Those having a healthy fasting glucose, triglycerides, and body weight have a very low baseline risk of incident T2D that is barely affected by the use of statins whereas those with increased fasting glucose, triglycerides, or body weight have a moderate to high risk of T2D that is markedly increased by statin use *(5)*. However, it remains unclear whether the increased risk of T2D associated with statin use is due to decreased insulin secretion, decreased insulin sensitivity, or both, and additional studies are needed to clarify this important issue.

At this point, we believe that the influence of these studies on clinical guidelines should be relatively modest, if any. For statins, the benefits from lowering the risk of CHD by far outweigh the adverse effects on T2D risk—even in individuals who develop diabetes. Whether this holds true also for other LDL-C–lowering therapies remains to be addressed. Although some may argue that the study by Lotta et al. indicates that ezetimibe may be particularly prone to adverse effects on glucose metabolism and T2D risk, we want to point out that you cannot extrapolate the effect size of an *NPC1L1* allele seen in one study to the actual risk of events in real clinical situations. Also, there are other things influencing clinical management, such as other side effects, cost efficiency, and need for LDL-C–lowering, especially for high-risk populations. Even if the ultimate proof of effects of drugs needs to come from randomized clinical trials, these studies taken together do provide rather convincing evidence that lowering of LDL-C is associated with a modestly increased risk of T2D, and that this risk increase is unlikely to be restricted to one drug class. As a consequence, we would argue that it is a good idea for the practicing clinician to pay attention to metabolic side effects in patients being treated with lipid-lowering therapy, particularly in individuals who are at a higher risk of developing T2D owing to risk factors such as obesity, signs of insulin

resistance, family history of T2D, or an unhealthy lifestyle. However, apart from that, we do not believe that there is sufficient evidence to withhold lipid-lowering therapy from individuals otherwise meeting the treatment criteria.

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