Lifestyle Intervention for Weight Loss and Cardiometabolic Changes in the Setting of Glucokinase Regulatory Protein Inhibition

Glucokinase Regulatory Protein-Leu446Pro Variant in Look AHEAD

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Background—Glucokinase regulatory protein (GCKR) inhibitors offer a novel treatment approach for glucose control in diabetes mellitus; however, their cardiometabolic effects, particularly in relation to increased triglycerides and C-reactive protein (CRP) levels, are of concern. GCKR Leu446Pro is a common variant associated with reduced GCKR function, increased triglycerides, and CRP.

Methods and Results—We investigated whether a 1-year intensive lifestyle intervention (ILI) for weight loss would avert the unfavorable cardiometabolic effects associated with GCKR Leu446Pro when compared with a diabetes mellitus support and education arm in overweight/obese individuals with type 2 diabetes mellitus with triglyceride (n=3214) and CRP (n=1411) data participating in a randomized lifestyle intervention study for weight loss, Action for Health in Diabetes Mellitus (Look AHEAD). Once demographics, medication use and baseline adiposity, and fitness were accounted for, ILI did not modify the baseline association of GCKR-Leu446Pro with elevated triglycerides (β±SE=0.067±0.013, P=1.5×10⁻⁷ and β±SE=0.052±0.015, P=5×10⁻⁴) or with elevated CRP (β±SE=0.136±0.034, P=5.1×10⁻³ and β±SE=0.903±0.038, P=0.015) in the overall sample and Non-Hispanic Whites, respectively. The lack of a protective effect from ILI at 1 year when compared with diabetes mellitus support and education (ILI versus diabetes mellitus support and education interaction for triglyceride and CRP change, respectively: P=0.64 and 0.37 in the overall sample; P=0.27 and 0.05 in Non-Hispanic Whites) persisted after additional adjustment for changes in adiposity and fitness.

Conclusions—Moderate improvements in adiposity and fitness with ILI did not mitigate the adverse cardiometabolic effects of GCKR inhibition in overweight/obese individuals with diabetes mellitus. (Circ Cardiovasc Genet. 2016;9:71-78. DOI: 10.1161/CIRCGENETICS.115.001192.)

Key Words: behavioral intervention ▪ C-reactive protein ▪ diabetes mellitus ▪ GCKR ▪ lifestyle ▪ obesity ▪ triglycerides

GCKR is found abundantly in liver, mainly in the nucleus of hepatocytes, where it plays a major role in the post-translational regulation of glucokinase (hexokinase IV). Glucokinase phosphorylates glucose, a fundamental step in the uptake of glucose by the liver for glycolysis, glycogen formation, and lipogenesis.4 When glucose levels are low, GCKR binds hepatic glucokinase in the nucleus, rendering the enzyme inactive and keeping it sequestered until glucose levels increase.5 The liver sees inhibition of glucokinase activity as a state of glucose depletion; glycogen levels decrease and gluconeogenesis increases, raising circulating glucose levels.6 Glucokinase is also present in pancreatic islet cells, where it functions as a glucose sensor.
that modulates insulin secretion, explaining the increased risk of hypoglycemia observed with activating glucokinase mutations or with systemic pharmacological glucokinase activation.8–9

Recent experiments in animals have shown that small molecule disruptors of GCKR promote lowering of blood glucose in hyperglycemic, but not in normoglycemic, animals,10,11 highlighting the advantage of this pharmacological approach over that of direct glucokinase activation for the treatment of individuals with diabetes mellitus. However, worsening of hypertriglyceridemia and subclinical inflammation, resulting from GCKR inhibition,12 may, as in the case of glucokinase activation,9,13 offset the beneficial glucose-lowering effect of this therapeutic approach. Identifying potential strategies to mitigate the adverse metabolic effects that occur with disruption of GCKR function are needed if glucokinase-related pharmacological strategies are to be used for the treatment of diabetes mellitus.

Understanding the effects of genetics on disease development and response to therapy has revolutionized the treatment of cardiometabolic disease: Sulfonylureas are now used instead of insulin to effectively treat individuals with neonatal diabetes mellitus expressing ATP-sensitive potassium channel subunit Kir6.2-activating mutations14; monoclonal antibody therapies that inhibit proprotein convertase subtilisin-kexin type 9 are now available as potent therapeutic alternatives for individuals at high risk of direct glucokinase activation for the treatment of individuals with diabetes mellitus. However, worsening of hypertriglyceridemia and subclinical inflammation, resulting from GCKR inhibition,11,12 may, as in the case of glucokinase activation,9,13 offset the beneficial glucose-lowering effect of this therapeutic approach. Identifying potential strategies to mitigate the adverse metabolic effects that occur with disruption of GCKR function are needed if glucokinase-related pharmacological strategies are to be used for the treatment of diabetes mellitus.

In this substudy of The Action for Health in Diabetes (Look AHEAD), a randomized lifestyle intervention trial in overweight/obese individuals with type 2 diabetes mellitus (T2DM), we examined the effects of an intensive lifestyle intervention (ILI) aimed at producing weight loss on changes in triglyceride and CRP levels in the setting of genetic GCKR inhibition. We hypothesized that in the setting of diabetes mellitus, a 1-year of ILI would favorably alter the association of GCKR-Leu446Pro with dyslipidemia and high CRP levels, when compared with a control arm of diabetes mellitus support and education (DSE), and that the favorable effects of ILI would depend on the resulting improvement of overall adiposity and fitness. The liver plays an important role in the synthesis of CRP and in the production of circulating triglyceride-rich lipoproteins. We have observed in Look AHEAD that ILI resulted in a reduction in triglyceride and CRP levels,22,23 changes that could lead to an attenuation of the proinflammatory and lipogenic effects of GCKR-Leu446Pro in the liver. Our hypothesis is supported by findings from the Diabetes Prevention Program (DPP) in nondiabetic individuals showing that lifestyle intervention for weight loss was able to reduce the effects of the P466 allele on hypertriglyceridemia.24

Methods

Study Design and Participants

Look AHEAD study is a multicenter trial that randomly assigned participants with T2DM who were overweight or obese to ILI, with the goal of producing 7% weight loss through calorie restriction and physical activity or to DSE with no weight loss or physical activity goals.20 The intervention was modeled after that of DPP, but intensified given the greater severity of obesity and the use of insulin in our participants. During the first year, ILI participants attended 3 group sessions and 1 individual monthly encounter (initial 6 months), followed by 2 group and 1 individual monthly appointments thereafter, aimed at supporting behavioral change to increase physical activity to 175 weekly minutes of moderate-intensity exercise and to reduce caloric intake. The activity program relied on at-home exercise, mostly brisk walking. Participants were also asked to increase lifestyle forms of physical activity. The energy intake goal was 1200 to 1500 kcal/day if body weight was <114 kg and 1500 to 1800 kcal/day if weight was ≥114 kg. DSE participants received 3 group health information sessions during the year. All participants were required to pass a test of behavioral adherence before enrollment26 and to continue care with a primary provider during trial participation. ILI treatment session attendance at 1 year was excellent, regardless of underlying weight category.17 Although the randomized study ended early without a significant difference in the primary cardiovascular end point between treatment arms, the behavioral intervention was successful as participants in the ILI arm lost significantly greater amounts of weight and showed greater improvement in fitness, waist circumference, and indices of diabetes mellitus control, including a reduction in diabetes mellitus medication use, hemoglobin A1c, and fasting glucose, compared with DSE participants.22,23 The maximal benefits of ILI on weight loss and on fitness were evident during the first year of the trial when the intervention was most intense. In addition, and in relation to this study, participants in the ILI group showed greater improvements in triglycerides and CRP levels at year 1 when compared with the DSE group.21,22

Of 4047 Look AHEAD participants with available DNA, 276 were excluded: 1 subject had missing data on the marker of interest (rs1260326), 256 subjects had missing lipid medication information, 240 subjects were taking niacin or fibrates at both measurement times and were excluded because of the known medication effects on triglycerides, and 19 subjects were excluded because they were missing triglycerides and CRP levels at both time points. If participants initiated niacin or fibrates use after enrollment, the 1-year time point was dropped from the analyses, but the subjects themselves were retained in the data set because they still contributed to the estimation of baseline outcomes. This yielded an effective sample size of 3771 participants with both phenotypic and genotypic data, of which 2607 were non-Hispanic Whites (NHW).

In NHW-specific analyses, all 4 subjects from the smallest site were excluded because they had all been assigned to the ILI arm, and therefore, this site could not contribute to the estimation of ILI-DSE differences. Of note, measurements for CRP were obtained in approximately the first half of Look AHEAD participants at each study site, 1523 of which had also consented to genetic testing. Analysis on the effect of the intervention on the association of the GCKR variant with biomarker change was performed in 3214 participants with triglyceride levels at baseline and year 1 (2232 NHW) and in 1411 (1085 NHW) with available CRP data at both time points. All included participants signed informed consent for participation in Look AHEAD, including genetic analyses, with Institutional Review Board approval by their local institutions. Genetic analyses were approved by the Miriam Hospital and Tufts Medical Center Institutional Review Boards and measurement of CRP by the Baylor College of Medicine Institutional Review Board. Genotyping was performed using the Metabochip (Illumina, San Diego, CA).

Anthropometry and Fitness

Procedures for anthropometric measures, including body mass index (BMI) and waist circumference, have been previously described.19
Fitness was defined as the estimated level of metabolic equivalents of task (1 metabolic equivalents of task = 3.5 mL/kg/min of oxygen uptake) achieved on a treadmill workload (speed and grade) at 80% of maximal heart rate (submaximal) or at a rating of 16 on the Rating of Perceived Exertion scale for participants on β-blockers, as described previously.29

**Biomarker Measurements**

Fasting triglyceride levels were measured enzymatically using methods standardized to the Centers for Disease Control and Prevention reference methods.23 Hs-CRP was measured using a latex particle–enhanced immunoturbidometric assay (Equal Diagnostics/Genzyme) as previously described.22

**Statistical Analysis**

Continuous variables were summarized using mean/SD if symmetrical. Variables that were log-transformed to reduce skewness, including CRP and triglyceride levels, were displayed in both the original scale using median/interquartile-range and in the logarithmic scale using mean/SD in descriptive analysis.

Baseline (Y0) and Year 1 (Y1) measurements of interest were modeled jointly across time as multivariate normal variables with an unstructured covariance matrix. Associations with baseline values of the outcome of interest (Y0) and with change in the outcome from baseline to year-1 (Y1-Y0) were then evaluated by a linear transformation. Because both CRP and triglyceride levels were log-transformed before being entered into the regression model (Y=ln Z), regression coefficients (β) measure the effect per minor allele copy (T) on the scale in which each particular outcome was analyzed, that is, they represent differences in log CRP and log triglyceride levels between the CC and TC genotypic groups. Differences between CC and TT genotypic groups can be estimated by doubling the marker effects shown on these tables. Of note, absolute differences in the logarithmic scale become relative differences, that is, percentage changes, when expressed in the original scale of the data. Hence, marker effects in the CRP and triglyceride scale can be obtained by exponentiating the corresponding linear regression coefficients and associated 95% confidence interval end points.

Three-way interaction models of individual single-nucleotide polymorphism (SNP) markers with measurement time (1 year versus baseline) and study arm (ILI versus DSE) were estimated for each outcome in Splus 8.2 (TIBCO Software, Seattle, WA) using restricted maximum likelihood. An additive genetic model was used, with genotype coded by the number of minor alleles (0/1/2 copies). Three distinct types of SNP effects were estimated, which can be interpreted as the effect of 1 additional copy of the corresponding minor allele on (1) baseline triglyceride and CRP levels for ILI and DSE participants combined; (2) ILI versus DSE differences in baseline triglyceride and CRP levels as a randomization check; (3) ILI versus DSE differences in 1 year change in triglycerides and CRP levels (SNP×treatment×time interaction). All our results are based on full 3-way hierarchical interaction models, that is, one in which all 2-way interactions is also included in the model, with no additional model simplification. To aid with the interpretation of interactions, we report marker effects on 1-year change separately for ILI and DSE.

Outcomes at baseline and year 1 were adjusted in successively elaborated models for (1) study site, age, sex, ancestry (based on the first 3 Ancestry Informative Marker principal components), concurrent use of lipid-lowering medications (over 97% statins), and thiazolidinediones (with pioglitazone and rosiglitazone effects modeled separately), as well as baseline hormonal replacement therapy in women; (2) all model (1) covariates plus BMI and fitness at baseline; (3) all Model (1) covariates plus BMI and fitness as time-varying covariates. Of note, Model 3 accounts for both baseline values and 1-year change in BMI and fitness on 1-year change in outcomes. With the exception of study site, covariates were fully interacted with time, treatment, and time by treatment, so as to allow their effects to vary across study arm and time point. Finally, to compare our findings with those from DPP assessing the effects of ILI on the association of triglyceride and CRP changes with GCKR-Leu446Pro in individuals without diabetes mellitus,24 we examined the associations of interest in models that adjusted for covariates in model (1) with the addition of baseline BMI and waist circumference, instead of medication use.

**Results**

**Participant Characteristics at Baseline and Year 1**

Sample characteristics are shown in Table 1 for the cohort of Look AHEAD participants with genetic and biomarker data. Sample sizes are shown for each outcome, both in the overall cohort and in NHW and mainly reflect biomarker availability because study dropout in Look AHEAD at year 1 was low (≤3%).23 Participants were obese, with mean BMI of 36.2 kg/m². Triglyceride levels were mildly elevated at baseline, more so in NHW than in the overall group. CRP levels were high in both the overall sample and in NHW as previously described.22 Use of statin and thiazolidinediones were similar between groups. The frequency of the minor allele (T) for the GCKR-Leu446Pro variant (rs1260326) was 35% in the overall sample and 40% in NHW, without differences in prevalence by treatment arm.

Similar to the overall Look AHEAD cohort,23 ILI participants in this study, in both the overall and the NHW samples, showed lower BMI, waist circumference, triglycerides, and CRP and improved fitness at 1 year, as compared with participants in the DSE group (all P<0.001; Table 1). Differences in lipid-lowering medication and thiazolidinedione use at 1 year were observed between study arms, with greater medication use among DSE participants.

**Effects of GCKR-Leu446Pro Variant on Baseline Triglyceride and CRP Levels**

Marker effects for the full sample and for NHW at baseline are shown in Table 2 for triglycerides and in Table 3 for CRP. Given that marker effects on triglyceride and CRP levels showed no between-arm differences by genotypic group in either the overall sample or among NHW at baseline, we show baseline results with both treatment groups combined. GCKR-Leu446Pro was significantly associated with increased levels of triglycerides and CRP at baseline. The association of the T allele with elevated log triglycerides in the overall sample (β±SE=0.068±0.013; P=7.3×10⁻⁴) corresponds to a 7% increase in triglycerides per minor allele copy (exp [β=1.07, 95% confidence interval=1.04–1.10]). The association was also significant among NHW alone (β±SE=0.053±0.015; P=3×10⁻⁴), with a 5% increase in triglycerides per minor allele copy (exp [β=1.05, 95% confidence interval=1.02–1.09]). Likewise, the T allele was associated with high log CRP in both the overall sample (β±SE=0.148±0.036; P=4.2×10⁻⁵) and among NHW (β±SE=0.016±0.042; P=0.01). This association corresponds to a 16% increase in CRP per minor allele copy (exp [β=1.16, 95% confidence interval=1.08–1.24) in the overall sample, attenuated to an 11% increase among NHW (exp [β=1.11, 95% confidence interval=1.02–1.21]). Additional adjustment for BMI and cardiorespiratory fitness at baseline did not materially affect these findings.

**Associations of GCKR-Leu446Pro With 1-Year Change in Triglyceride and CRP Levels by Treatment Arm**

Marker effects from baseline to follow-up are shown in Table 4 for triglycerides and in Table 5 for CRP. Estimates are presented...
separately for ILI and DSE and then compared across study arms. As seen from Table 4, no evidence emerged of ILI versus DSE differences in marker effects on 1-year change in triglycerides, whether one adjusted or not for baseline BMI and fitness or their 1-year change (P value ≥ 0.56 in the overall sample and ≥ 0.26 among NHW). Comparable results were obtained...
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Table 2. Baseline Associations of GCKR-Leu446Pro With Triglycerides in the Overall Sample and in Non-Hispanic Whites

| Model | Overall | Non-Hispanic Whites |
|-------|---------|---------------------|
|       | Beta    | SE      | P Value | Beta    | SE      | P Value |
| 1     | 0.068   | 0.013   | 7.3E-08 | 0.053   | 0.015   | 0.0003  |
| 2     | 0.067   | 0.013   | 1.5E-07 | 0.052   | 0.015   | 0.0005  |

Model 1 adjusts for age, sex, ancestry (PCAs 1–3), clinic site, hormonal replacement therapy, use of lipid-lowering medication, and thiazolidinediones (pioglitazone and rosiglitazone modeled separately). Model 2 adds baseline BMI and fitness to Model 1. Allele T (the minor allele) was used as the coded allele. BMI indicates body mass index; GCKR, glucokinase regulatory protein; and PCAs 1–3, first 3 Ancestry Informative Marker principal components.

Discussion

To our knowledge, this is the first study to evaluate the interaction of GCKR-Leu446Pro status with a randomly assigned behavioral lifestyle intervention for weight loss on CRP and triglyceride levels in the setting of diabetes mellitus. Our study follows promising findings from the DPP23 in individuals without diabetes mellitus and takes advantage of the common occurrence of this genetic variant associated with decreased GCKR function. Our studies in a large sample of overweight/obese adults with T2DM show that, in contrast to what was observed with triglycerides in the less obese non-diabetic DPP cohort, randomization to ILI did not protect GCKR-Leu446Pro carriers against the increase in triglycerides and CRP levels observed at baseline, despite significant improvements in adiposity and fitness. Our results suggest that moderate weight loss and improved fitness are insufficient to mitigate the unfavorable effects of GCKR inhibition on hypertriglyceridemia and on subclinical inflammation, as measured by CRP, in individuals with T2DM and point to the need of developing other strategies to mitigate the adverse cardiometabolic effects of GCKR inhibitors for the treatment of hyperglycemia in diabetes mellitus.

As observed in studies involving mostly/only individuals without diabetes mellitus,11,21,24 we found in this cohort of overweight/obese individuals with diabetes mellitus that GCKR-Leu446Pro is associated with a median 7% and 15% increase in baseline triglyceride and CRP levels, respectively. GCKR, highly expressed in hepatocytes, blocks glucokinase activity when glucose levels are low by sequestering the enzyme in the nucleus.5,6 Decreased GCKR function results in increased gluconeogenesis and glycogenolysis and may, particularly if chronically stimulated, increase lipid synthesis, resulting in a rise in very low-density lipoprotein secretion and triglyceride deposition in the liver.4 Hepatic fat deposition activates mechanisms that result in local inflammation and increase the secretion of CRP.25

In DPP, the only other study to date examining the effects of a randomized weight loss intervention in GCKR carriers,33 a 1-year weight loss intervention was able to weaken the effects of GCKR-Leu446Pro on baseline triglyceride levels. The lack of an effect of ILI on the triglyceride effects of the variant in our study of diabetic patients, when compared with DPP (a study in adults without diabetes mellitus), is not the result of sample size differences, race/ethnicity, or a lack of significant weight loss because sample size was similar for this outcome between studies and change in weight loss was greater in Look AHEAD than in DPP. Furthermore, both studies included a diverse population. Rather, our results suggest that among overweight/obese diabetic patients, the underlying metabolic abnormalities may be such that ILI is not able to avert the deleterious effects of GCKR inhibition on triglyceride levels, a phenomenon that may in part be as a result of peculiarities of the effects of GCKR inhibition in individuals with chronic hyperglycemia, to the more advanced age and longer duration of dysmetabolism in our diabetic cohort, and to the use of medications that are not routinely prescribed in nondiabetic adults, including statins and thiazolidinediones. In addition, our results on the association of GCKR-Leu446Pro on CRP did not differ materially from those for CRP change in all models under examination in the overall sample (P≥0.20), although a borderline significant interaction effect was detected among NHW alone (P=0.05), after regression adjustment for the full set of variables under consideration.

Table 3. Baseline Associations of GCKR-Leu446Pro With CRP in the Overall Sample and in Non-Hispanic Whites

| Model | Overall | Non-Hispanic Whites |
|-------|---------|---------------------|
|       | Beta    | SE      | P Value | Beta    | SE      | P Value |
| 1     | 0.148   | 0.036   | 4.2E-05 | 0.106   | 0.042   | 0.011  |
| 2     | 0.136   | 0.034   | 5.1E-05 | 0.093   | 0.038   | 0.015  |

Model 1 adjusts for age, sex, ancestry (PCAs 1–3), clinic site, hormonal replacement therapy, use of lipid-lowering medication, and thiazolidinediones (pioglitazone and rosiglitazone modeled separately). Model 2 adds baseline BMI and fitness to Model 1. Allele T (the minor allele) was used as the coded allele. BMI indicates body mass index; GCKR, glucokinase regulatory protein; and PCAs 1–3, first 3 Ancestry Informative Marker principal components.
of DPP. The CRP by treatment interaction we observed in favor of the DSE arm was limited to the NHW subgroup and was only of borderline significance (P ≤ 0.05 after adjustment for the full set of variables, including baseline BMI, fitness, and medication use). This interaction remained unchanged after accounting for adiposity and fitness changes. Of note, when analyzing data from our sample of overweight/obese individuals with diabetes mellitus using regression models similar to those of Pollin et al24 in the prediabetic sample of DPP, we were unable to find evidence of a protective effect of ILI on triglyceride or CRP change. These models (in the Data Supplement) examined the effects of ILI on the association of GCKR-Leu446Pro variant with changes in triglyceride and CRP levels after adjustments for demographics, baseline BMI, and waist circumference, without controlling for medication use.

We acknowledge that our study has several strengths, as well as limitations. Look AHEAD is the largest randomized lifestyle intervention study for weight loss in individuals with diabetes mellitus to date. It provides a unique opportunity, not available from observational studies, to examine the effects of intentional weight loss, increased physical activity, and improved fitness on the association of genetics with metabolic traits. Our study contributes novel data in the setting of T2DM, suggesting that changes in weight and fitness with ILI do not significantly mitigate the unfavorable lipid and inflammatory effects associated with GCKR inhibition. Our results have translational significance because they relate to a potential approach in the management of the adverse metabolic consequences of glucose-lowering therapies that rely on the disruption of GCKR function.

We recognize that the effects of ILI on GCKR-Leu446Pro observed in this study are not indicative of causality and that

Table 5. Associations of GCKR-Leu446Pro With 1-Year Change in CRP Levels by Treatment Arm in the Overall Sample and in Non-Hispanic Whites

| Study Arm | Model | Overall | Non-Hispanic Whites |
|-----------|-------|---------|---------------------|
|           | Beta  | SE      | P Value             | Beta  | SE      | P Value             |
| ILI       |       |         |                     |       |         |                     |
| 1         | 0.024 | 0.046   | 0.59                | 0.065 | 0.055   | 0.24                |
| 2         | 0.022 | 0.046   | 0.63                | 0.067 | 0.055   | 0.22                |
| 3         | 0.011 | 0.044   | 0.80                | 0.061 | 0.053   | 0.25                |
| DSE       |       |         |                     |       |         |                     |
| 1         | -0.065| 0.048   | 0.18                | -0.086| 0.057   | 0.13                |
| 2         | -0.064| 0.048   | 0.19                | -0.089| 0.057   | 0.12                |
| 3         | -0.060| 0.047   | 0.20                | -0.089| 0.055   | 0.10                |
| ILI vs DSE|       |         |                     |       |         |                     |
| 1         | 0.089 | 0.066   | 0.18                | 0.151 | 0.079   | 0.06                |
| 2         | 0.086 | 0.066   | 0.20                | 0.156 | 0.079   | 0.05                |
| 3         | 0.071 | 0.064   | 0.27                | 0.150 | 0.076   | 0.05                |

Model 1 adjusts for age, sex, ancestry (PCAs 1–3), clinic site, hormonal replacement therapy at baseline, 1-year use of lipid-lowering medication, and thiazolidinediones (pioglitazone and rosiglitazone modeled separately). Model 2 adds baseline BMI and fitness to Model 1. Model 3 adds BMI and fitness at 1 year to Model 2. Allele T (the minor allele) was used as the coded allele. BMI indicates body mass index; DSE, diabetes mellitus support and education arm; GCKR, glucokinase regulatory protein; ILI, intensive lifestyle intervention; and PCAs 1–3, first 3 Ancestry Informative Marker principal components.
the association of the genetic variant with triglycerides and CRP may not be fully comparable to that seen with pharmacological GCKR inhibition. Although Look AHEAD is the largest trial available to date with data in overweight/obese individuals with diabetes mellitus within a randomized lifestyle intervention for weight loss, our findings of genotype-treatment response interaction are limited because SNP×treatment interactions are likely to have smaller effect sizes than SNP main effects. Our power limitations preclude the performance of sensitivity analyses exploring whether certain subsets of diabetic GCKR variant carriers, such as those who achieved the greatest weight loss within the ILI arm in Look AHEAD, may have experienced a mitigation of the variant effect on lipid and inflammatory biomarker levels. Furthermore, it is possible, although highly unlikely given the findings of Pollin et al24 using a similar statistical approach in nondiabetic individuals, that a nonlinear model would have uncovered a potential benefit from ILI on variant carriers. Of note, despite our findings suggesting that ILI does not weaken the unfavorable associations of GCKR-Leu446Pro with elevated triglycerides and CRP, our study also suggests that GCKR inhibition does not interfere with the beneficial effects of ILI on triglyceride and CRP change in overweight/obese individuals with diabetes mellitus. Strategies that directly target triglyceride25 and inflammatory pathways deserve further investigation as potential approaches to enhance the metabolic benefits of ILI in the setting of GCKR inhibition.

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Disclosures

None.

Appendix

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CLINICAL PERSPECTIVE

There is a growing need for treatment strategies that correct hyperglycemia without causing significant hypoglycemia or increasing cardiovascular disease risk in patients who have type-2 diabetes mellitus. Inhibitors of glucokinase regulatory protein (GCKR) have shown promise in controlling blood glucose levels; however, this benefit is associated with worsening of hypertriglyceridemia and subclinical inflammation. To better predict the effects of GCKR inhibition in the setting of type-2 diabetes mellitus, we studied the effects of a common coding variant, GCKR-Leu446Pro (rs1260326), which has reduced GCKR function, in Look Action for Health in Diabetes Mellitus (AHEAD), a randomized lifestyle intervention trial in overweight/obese individuals with type-2 diabetes mellitus. We asked whether an intensive lifestyle intervention aimed at producing weight loss also modified the effects of genetic GCKR inhibition on changes in triglyceride and the inflammatory biomarker C-reactive protein. Our hypothesis was supported by findings from the Diabetes Prevention Program that showed that a behavioral intervention reduced the effects of the P466 allele on hypertriglyceridemia in nonobese individuals. Compared with baseline, the body mass index decreased significantly in the intensive lifestyle intervention group but not in a diet support and education group; however, intensive lifestyle intervention did not protect against the increase in triglyceride and C-reactive protein levels observed with GCKR-Leu446Pro at baseline. Taken together, our results demonstrate that a behavioral intervention designed to produce moderate weight loss does not mitigate the adverse cardiometabolic effects of GCKR inhibition in obese individuals with diabetes mellitus. Additional strategies are required to better control the effects of GCKR inhibition on triglycerides and inflammation.