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A High Intake of Saturated Fatty Acids Strengthens the Association between the Fat Mass and Obesity-Associated Gene and BMI[1–3]

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Abstract

Evidence that physical activity (PA) modulates the association between the fat mass and obesity-associated gene (*FTO*) and BMI is emerging; however, information about dietary factors modulating this association is scarce. We investigated whether fat and carbohydrate intake modified the association of *FTO* gene variation with BMI in two populations, including participants in the Genetics of Lipid Lowering Drugs and Diet Network (GOLDN) study (n = 1069) and in the Boston Puerto Rican Health (BPRHS) study (n = 1094). We assessed energy, nutrient intake, and PA using validated questionnaires. Genetic variability at the *FTO* locus was characterized by polymorphisms rs9939609 (in the GOLDN) and rs1121980 (in the GOLDN and BPRHS). We found significant interactions between PA and *FTO* on BMI in the GOLDN but not in the BPRHS. We found a significant interaction between SFA intake and *FTO* on BMI, which was stronger than that of total fat and was present in both populations (P-interaction = 0.007 in the GOLDN and P-interaction = 0.014 in BPRHS for categorical; and P-interaction = 0.028 in the GOLDN and P-interaction = 0.041 in BPRHS for continuous SFA). Thus, homozygous participants for the *FTO*-risk allele had a higher mean BMI than the other genotypes only when they had a high-SFA intake (above the population mean: 29.7 ± 0.7 vs. 28.1 ± 0.5 kg/m^2; P = 0.037 in the GOLDN and 33.6. ± 0.8 vs. 31.2 ± 0.4 kg/m^2; P = 0.006 in BPRHS). No associations with BMI were found at lower SFA intakes. We found no significant interactions with carbohydrate intake. In conclusion, SFA intake modulates the association between *FTO* and BMI in American populations. J. Nutr. 141: 2219–2225, 2011.

Introduction

Minor alleles at the *FTO* locus (namely, SNP[15] rs9939609 and rs1121980, both in high linkage disequilibrium) have been associated with higher BMI and obesity risk in multiple populations (1–8). Hence, the *FTO* gene has now been considered as one of the most important in common forms of obesity (9,10). However, despite the overall consistency reported, there are a number of studies in which the *FTO* gene has not been associated with BMI or obesity (11–15).

These inconsistencies may be the result of gene-environment interactions between the *FTO* gene and lifestyle variables as suggested by some reports. This indicates that the effects of *FTO* variants on BMI are not unavoidable but, rather, can be considerably modulated by environmental factors. Identifying those environmental interactions will be needed for establishing targeted preventive approaches in individuals with greater genetic susceptibility to obesity. PA has been the environmental factor that is most commonly reported as showing a significant interaction with the effects of variations (both of the SNP rs9939609 and the rs1121980) in the *FTO* (16–21). Accordingly, a lower level of PA would boost the effect of the variants associated with high BMI (called risk-alleles), whereas a higher

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[3] Supplemental Tables 1–4 and Figures 1 and 2 are available from the "Online Supporting Material" link in the online posting of the article and from the same link in the online table of contents at jn.nutrition.org.
[14] These authors contributed equally to this work.
[15] Abbreviations used: BPRHS, Boston Puerto Rican Health Study; GOLDN, Genetics of Lipid Lowering Drugs and Diet Network; IRB, Institutional Review Board; PA, physical activity; SNP, single nucleotide polymorphism.
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level of PA would neutralize the genetic effects of the risk-alleles on BMI. However, not all studies reported support this interaction of the *FTO* polymorphism with PA (22–24). Recently, another relevant interaction of an *FTO* genetic variant (rs9939609) with total fat and carbohydrate intake in determining BMI has been described in middle-aged individuals in Sweden (20). In this population (20), the greater BMI in individuals with the *FTO* risk-allele was restricted to those who reported a high-fat diet, whereas the *FTO* risk-allele was not associated with a higher BMI among participants with lower fat intakes. An inverse interaction was observed with carbohydrate intake. Despite its relevance, the consolidation of this gene-diet interaction has not been pursued in other populations, thus the need to undertake replication studies on this interaction. Moreover, because only total dietary fat was investigated in that study, it is necessary to go deeper into which types of fatty acids are most implicated in that interaction. Based on the results of our previous work in which we found that SFA consistently interact with polymorphisms associated with obesity (25), our hypothesis was that SFA may play an important role in modulating the effects of the *FTO* polymorphisms. Some human studies (mainly carried out in children) have found that participants carrying the *FTO* risk-allele consume more total energy and fat than noncarriers (26,27). Our objectives, therefore, were to: *1*) study the association between *FTO* variants, anthropometric variables, and energy and macronutrient intake in two independent, adult, U.S. populations that differed in demographic and lifestyle characteristics; *2*) determine whether the previously reported interaction between total fat and carbohydrate intake as well as the interaction of PA with the *FTO* variants on BMI could be replicated; and *3*) investigate whether SFA intake has a stronger interaction than total fat intake in these populations.

Participants and Methods

We studied 2163 participants from two independent U.S. populations (the GOLDN and BPRHS) that had been extensively characterized by our group in previous studies (28,29). The IRB of the institutions involved approved the study protocols and all participants provided written informed consent.

The GOLDN Study. About 1200 adult individuals of European ancestry were recruited from two National Heart, Lung and Blood Institute Family Heart Study field centers (Minneapolis, MN and Salt Lake City, UT) as previously reported (28). We included 1069 participants (507 men and 562 women) for whom genetic (rs1121980 and rs9939609 *FTO* polymorphisms), anthropometric, dietary, PA, and other control variables were available. The protocol was approved by the IRB at the Universities of Alabama, Minnesota, Utah, and Tufts.

The BPRHS. The study comprised approximately 1200 Puerto Rican (Hispanics of Caribbean origin) participants aged 45–75 y in the greater Boston area (29) and was derived from the NIH-funded Centers on Population Health and Health Disparities. We included 1094 (315 men and 779 women) participants for whom genetic (rs1121980), anthropometric, dietary, PA, and other control variables were available. The protocol was approved by the IRB at Tufts University.

Anthropometric and PA determinations. Anthropometric variables, including height, weight, and waist circumference, were measured (28,29). BMI was calculated as weight (kg)/height (m^2). Participants with a BMI $\geq$30 kg/m^2 were considered obese. PA in the GOLDN was assessed by an interviewer-administered questionnaire containing questions on the number of h/d dedicated to different levels (heavy, slight, and sedentary) of activity as well as the average number of h/d without activity (29,30). Afterwards, a single score representing PA was calculated (25,29). In the BPRHS, a PA single score based on the Paffenbarger questionnaire of the Harvard Alumni Activity Survey (31) was also estimated. In both populations, a higher score indicates a greater amount of PA.

Dietary intake and other lifestyle variables. Diet was measured by validated questionnaires in each specific population (32–34). In the GOLDN, we estimated dietary intake with the Diet History Questionnaire (32,33). In the BPRHS, a specifically validated questionnaire for this population was used (34). All the included participants had valid dietary intake data from the FFQ (total daily energy within the range of 800–5500 kcal in men or 600–4500 kcal in women). The percentage of individuals outside the inclusion range was very low (3.6% in GOLDN and 4.2% in BPRHS).

Data on smoking and drinking were obtained as previously described (29,30).

Genetic analyses. DNA was isolated from blood (Qiagen). We performed *FTO* genotyping (rs1121980 and rs9939609 in the GOLDN and rs1121980 in the BPRHS) using Taqman assays with allele-specific probes on the ABIPrism 7900HT Sequence Detection System (Applied Biosystems). All genetic analyses were undertaken in the same laboratory. Quality control measures were applied. Genotype frequencies were consistent with Hardy-Weinberg equilibrium in both populations.

Statistical analyses. Chi square tests were used to test differences in percentages. Normality of continuous variables was examined. Intakes of total fat (g/d), carbohydrates (g/d), protein (g/d), and fatty acids (g/d) and PA scores were log-transformed for statistical testing. Spearman correlation coefficients (r_s) between nutrient intakes were estimated. We first analyzed the association between the *FTO* and anthropometric variables (weight, BMI, and waist circumference) by ANOVA, including a test for linear trend. Sample size calculations were carried out assuming an allele frequency for the minor *FTO* allele of 0.44 and the parameters from the meta-analysis by Frayling et al. (1), in which each additional copy of the *FTO* risk-allele was associated with a BMI increase of a mean of ~0.4 kg/m^2 (range, 0.3–0.5 kg/m^2). Enrollment of ~1040 participants would be necessary for our association study to have 80% power (α-level = 0.05) in each population. Therefore, post hoc power calculations showed that our study incorporating 1069 participants in the GOLDN and 1069 participants in the BPRHS had 81% power in each population to show significant associations with BMI at α = 0.05.

We also tested the statistical homogeneity by gender by checking the significance of the interaction term between the *FTO* SNP and gender, and men and women were analyzed together. Control for potential confounders was carried out by general mixed regression models. Models were adjusted for gender, age, tobacco smoking, and alcohol consumption. In the GOLDN, because this is a population in which some participants are related, additional adjustments for family relationships were undertaken as previously described (28). In the BPRHS, further adjustment for admixture using the first component variable derived from the analysis of 100 ancestry informative markers was undertaken (35).

To study gene-PA and gene-diet (macronutrient intake) interactions in determining BMI, we used multivariate linear regression models, including main effects and interaction terms. We included the same variables for each population. PA was considered both as categorical and continuous (log of the PA score). Dietary variables were also considered as categorical and as continuous. We adjusted analyses for gender, age, tobacco smoking, alcohol consumption, PA, and total energy intake. Considering the different options and controversies to categorize the variables of diet and the diverse ways to express nutrient intake (in g/d or percent of energy), we used two different approaches for the analyses. We used a model that uses dietary variables expressed in g/d and includes the adjustment for total energy intake (25). This model is known as the standard multivariate energy-adjusted model (36). In our analyses, macronutrient intakes were analyzed as categorical (based on the population means) as well as continuous. However, given that the previous study by Sonestedt et al. (20) found a significant interaction between the *FTO* SNP and total fat and carbohydrate intakes in determining BMI and obesity risk using dietary variables expressed in energy percentages (nutrient density model), we

also fitted additional models expressing total fat and carbohydrate intakes as percent of energy and adjusting for energy intake (multivariate nutrient density) to determine if the results of Sonestedt et al. (20) could be replicated. For this second approach, we categorized macronutrient intakes in gender-specific tertiles to closely reproduce the same statistical model of the previous work (20).

When the *FTO*-diet interaction was analyzed with dietary variables in continuous form, it was depicted by computing the predicted values for each individual from the adjusted regression model and plotting these values against fat intake by the *FTO* genotype. Stratified analyses by fat intake levels were also carried out. Logistic regression models, including main effects and interaction terms, were fitted to test the *FTO* associations and the gene-PA or gene-diet interactions for determining the OR of obesity. Multivariate adjustments were done as indicated.

Statistical analyses were conducted with SAS software (v.9.1; SAS Institute) and SPSS software (v.17.0). Standard regression diagnostic procedures were used to ensure the appropriateness of the fitted models. All reported probability tests were 2-sided. Differences were considered significant at $P < 0.05$. Because our study was conducted in two independent populations to discard random associations by chance, we did not proceed to adjust for multiple comparisons.

Results

Association of FTO variants with obesity measures and food intake. The two populations studied differed in demographic and lifestyle variables (**Table 1**). In the GOLDN study, we did not observe significant associations between the *FTO* polymorphisms (rs1121980) (Table 1) or rs9939609 (**Supplemental Table 1**) and anthropometric (weight, BMI, and waist circumference) or dietary variables (energy and macronutrient intakes). Both *FTO* polymorphisms were in high linkage disequilibrium (0.997; $P < 0.001$) and we obtained the same associations for both. After additional adjustment for gender, age, tobacco smoking, alcohol drinking, and family relationships, we did not find differences in the significance of the results. Like the GOLDN study, the minor allele at the rs1121980 SNP in the BPRHS (Table 1) was not associated with greater BMI or greater energy or fat intake in the codominant model. After adjustment for gender, age, tobacco smoking, alcohol drinking, and population admixture, the results did not change in significance. When the recessive model was tested, we did not find significant associations. The lack of associations with BMI in these populations was not due to a lack of statistical power but to the small magnitude of the effects associated with the risk-allele carriers (T allele for rs1121980 and the A allele for rs9939609).

Interactions between FTO *variants and PA.* In the GOLDN, we found a significant interaction between the rs1121980 and PA when three categories (based on the GOLDN population tertiles) were considered (**Supplemental Fig. 1***A*) or when PA was analyzed as a continuous variable (**Supplemental Fig. 2***A*). We also observed significant interaction (*P*-interaction = 0.002) between the rs1121980 and PA tertiles on obesity (results not shown). In contrast, in the BPRHS, there were no significant interactions between PA and the rs1121980 SNP on BMI when three categories of PA (based on the BPRHS population tertiles) were considered (Supplemental Fig. 1*B*) ($P = 0.10$) or when PA was analyzed as a continuous variable (Supplemental Fig. 2*B*) ($P = 0.07$). Likewise, there were no significant interactions between PA and the *FTO* genotype on obesity (not shown).

TABLE 1 Anthropometric, dietary, and lifestyle characteristics depending on the *FTO* polymorphism (rs1121980) in the GOLDN and BPRHS participants[1]

	GOLDN participants (rs1121980)					BPRHS participants (rs1121980)				
	CC	CT	TT	P^2	P-trend[3]	CC	CT	TT	P^2	P-trend[3]
n	291	541	236			394	523	177		
Age, *y*	48.4 ± 15.9	49.2 ± 16.1	48.6 ± 16.6	0.76	0.90	57.6 ± 7.7	57.8 ± 7.5	56.3 ± 7.4	0.06	0.06
Weight, *kg*	83.1 ± 18.1	82.9 ± 18.3	82.5 ± 18.7	0.92	0.70	80.2 ± 17.4	79.5 ± 17.1	82.9 ± 18.1	0.07	0.08
BMI, kg/m^2	28.5 ± 5.6	28.3 ± 5.8	28.0 ± 5.4	0.64	0.34	32.2 ± 6.9	31.5 ± 6.5	32.6 ± 6.9	0.11	0.49
Waist, *cm*	96.1 ± 15.4	96.2 ± 17.4	96.3 ± 16.6	0.99	0.92	101.5 ± 14.9	101.1 ± 15.1	103.7 ± 15.6	0.14	0.11
Daily intakes										
Energy, *MJ*	8.67 ± 3.49	8.71 ± 3.72	8.16 ± 3.27	0.13	0.10	8.77 ± 3.52	8.68 ± 3.62	8.58 ± 3.64	0.83	0.56
Total fat, *g*	82.8 ± 38.8	83.2 ± 41.7	79.1 ± 37.2	0.41	0.29	73.2 ± 34.6	72.5 ± 34.1	72.3 ± 35.4	0.94	0.77
SFA, *g*	27.5 ± 13.2	28.2 ± 15.5	26.4 ± 13.2	0.28	0.42	22.8 ± 11.8	22.6 ± 11.6	22.8 ± 12.3	0.95	0.98
MUFA, *g*	31.2 ± 15.2	31.2 ± 15.9	29.9 ± 14.5	0.56	0.35	26.5 ± 12.9	26.2 ± 12.6	26.0 ± 13.2	0.88	0.65
PUFA, *g*	17.9 ± 8.9	16.5 ± 8.7	16.8 ± 8.4	0.29	0.12	17.6 ± 8.4	17.6 ± 8.8	17.3 ± 8.8	0.87	0.62
Proteins, *g*	80.6 ± 35.5	81.7 ± 37.2	78.5 ± 34.4	0.52	0.51	89.6 ± 40.4	89.7 ± 40.1	90.4 ± 42.3	0.98	0.84
Carbohydrates, *g*	249 ± 100	253 ± 111	235 ± 96	0.07	0.13	271 ± 105	267 ± 115	260 ± 108	0.54	0.27
Total fat, *% energy*	35.4 ± 7.0	35.4 ± 6.7	36.0 ± 6.2	0.52	0.33	30.9 ± 5.2	31.1 ± 5.4	31.3 ± 4.5	0.59	0.36
SFA, *% energy*	11.7 ± 2.6	11.9 ± 2.8	11.9 ± 2.6	0.55	0.29	9.5 ± 2.2	9.6 ± 2.3	9.8 ± 2.1	0.49	0.24
Proteins, *% energy*	15.6 ± 2.9	15.8 ± 2.8	16.1 ± 2.6	0.15	0.06	17.0 ± 3.2	17.5 ± 3.7	17.6 ± 3.2	0.11	0.08
Carbohydrates, *% energy*	48.7 ± 8.8	49.2 ± 8.5	48.6 ± 7.7	0.56	0.90	52.4 ± 7.8	51.7 ± 7.7	51.2 ± 6.6	0.16	0.08
PA score[4]	33.9 ± 5.9	34.3 ± 6.1	34.4 ± 6.9	0.52	0.32	31.8 ± 4.8	31.3 ± 4.5	31.8 ± 4.7	0.18	0.92
Current smokers, *%*	7.2	7.6	7.7	0.98	0.85	26.2	24.6	22.7	0.42	0.41
Current drinkers, *%*	52.6	49.6	49.6	0.69	0.47	58.4	55.5	53.7	0.63	0.35
Diabetes, *%*	9.3	7.9	6.1	0.37	0.16	38.4	40.4	38.3	0.80	0.88
Obesity, *%*	33.3	32.8	35.6	0.75	0.61	56.1	55.7	61.1	0.36	0.36

[1] Values are means ± SD or proportions. BPRHS, Boston Puerto Rican Health Study; GOLDN, Genetics of Lipid Lowering Drugs and Diet Network; PA, physical activity.
[2] *P* in the ANOVA test for continuous variables or chi square test for categorical variables.
[3] The polynomial contrast and chi square test were used to determine *P*-linear trend for continuous and categorical variables, respectively.
[4] PA score was estimated as described in "Methods."

TABLE 2 BMI and obesity risk according to *FTO* genotypes and the level of total fat and carbohydrate intake in the GOLDN and BPRHS participants[1]

	BMI								Obesity risk					
	n	*FTO*[2] 11+12, Mean ± SEM	*n*	*FTO* 22, Mean ± SEM	*P*-interaction[3,4]	*P*[4,5]	*P*-interaction[3,6]	*P*[4,6]	*FTO*[2] 11+12, OR	*FTO* 22, OR (95% CI)	*P*- interaction[3,4]	*P*[4,7]	*P*-interaction[3,6]	*P*[6,7]
GOLDN study														
Total fat intake,[8] *g/d*					0.027		0.023				0.09		0.07	
Low: <82.0	520	28.2 ± 0.4	115	27.2 ± 0.6		0.09		0.08	1 ref	0.91 (0.58, 1.41)		0.66		0.65
High: ≥82.0	361	28.3 ± 0.6	73	29.3 ± 0.7		0.17		0.17	1 ref	1.68 (0.98, 2.86)		0.06		0.06
Carbohydrate intake,[8] *g/d*					0.52		0.49				0.65		0.94	
Low: <247	485	28.8 ± 0.4	114	27.5 ± 0.7		0.43		0.36	1 ref	1.16 (0.74, 1.80)		0.52		0.59
High: ≥247	398	28.4 ± 0.6	72	27.6 ± 0.8		0.86		0.87	1 ref	1.15 (0.67, 1.98)		0.61		0.61
BPRHS study														
Total fat intake,[8] *g/d*					0.021		0.023				0.10		0.12	
Low: <72.6	517	31.1 ± 0.4	104	30.7 ± 0.7		0.69		0.63	1 ref	1.08 (0.69, 1.71)		0.71		0.88
High: ≥72.6	400	30.8 ± 0.5	73	33.2 ± 0.8		0.009		0.018	1 ref	1.93 (1.09, 3.34)		0.025		0.026
Carbohydrate intake,[8] *g/d*					0.26		0.24				0.23		0.26	
Low: <266	510	31.6 ± 0.4	102	31.8 ± 0.7		0.67		0.64	1 ref	1.17 (0.73, 1.85)		0.51		0.60
High: ≥266	407	30.2 ± 0.4	75	31.7 ± 0.7		0.06		0.12	1 ref	1.74 (0.99, 3.13)		0.05		0.06

[1] Values are adjusted means ± SEM or OR and 95% CI. BPRHS, Boston Puerto Rican Health Study; GOLDN, Genetics of Lipid Lowering Drugs and Diet Network; PA, physical activity.
[2] The *FTO* genotypes were rs9939609 (11+12: TT+TA; 22: AA) in the GOLDN study and rs1121980 (11+12: CC+CT; 22: TT) in the BPRHS study.
[3] *P*-interaction between the corresponding *FTO* genotype and total fat intake or carbohydrate intake in the corresponding multivariate model (lineal or logistic adjusted for the corresponding covariables in determining BMI or obesity risk.
[4] Models adjusted for gender, age, smoking, drinking, energy intake, and PA tertiles.
[5] *P* for mean comparison between the *FTO* genotypes in the corresponding strata.
[6] Models adjusted for gender, age, smoking, drinking, energy intake, PA tertiles, and total fat/carbohydrates.
[7] *P* for the OR corresponding to the homozygotes for minor allele in comparison with the other genotypes in each strata.
[8] Levels of intake were defined according to the corresponding population mean.

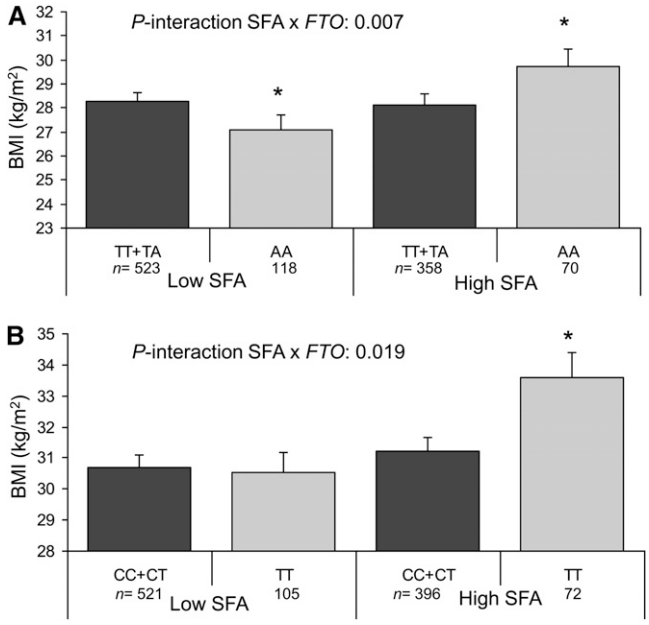

FIGURE 1 BMI in participants in the GOLDN study (*A*) and the BPRHS (*B*) depending on the SFA intake (2 levels, based on the population mean: 27.6 g/d in GOLDN and 22.7 g/d in BPRHS) and the *FTO* polymorphisms (recessive model; rs9939609 in GOLDN and rs1121980 in BPRHS). Values are adjusted means ± SEM. Models were adjusted for gender, age, tobacco smoking, alcohol drinking, PA, and total energy intake. *P* values for mean comparison in each saturated fat strata were also adjusted for covariates. *Different from CC+TT, $P < 0.05$. BPRHS, Boston Puerto Rican Health Study; GOLDN, Genetics of Lipid Lowering Drugs and Diet Network; PA, physical activity.

Interactions between FTO variants and dietary intake. A higher total fat intake was associated with higher BMI in homozygous participants for the minor allele of the *FTO* in both populations; these interactions reached a higher significance level in the GOLDN participants than in the BPRHS participants. Thus, in the GOLDN (**Table 2; Supplemental Table 2**), we found significant interactions between the *FTO* polymorphism and total fat intake on BMI whether total fat was expressed either as gender-specific tertiles of percent of energy ($P = 0.017$) or as two categories based on the mean intake in g/d ($P = 0.027$). In BPRHS participants (Table 2; Supplemental Table 2), we only found a significant interaction when total fat intake was expressed in g/d ($P = 0.021$). We did not observe a significant interaction with carbohydrate intake (Table 2; **Supplemental Table 3**).

Further, we investigated the specific effects of SFA intake on this interaction. In both the GOLDN and BPRHS, we obtained significant interactions both as categorical (**Fig. 1**) and continuous (**Fig. 2**) variables. In both populations, an SFA intake higher than the mean was associated with a higher mean of BMI in homozygous participants for the minor allele compared with the other genotypes (29.7 ± 0.7 vs. 28.1 ± 0.5 kg/m^2; $P = 0.037$ in GOLDN and 33.6. ± 0.8 vs. 31.2 ± 0.4 kg/m^2; $P = 0.006$ in PBRHS). Accordingly, in the GOLDN, homozygous individuals for the minor allele did not have a higher risk of obesity than other genotypes when the SFA intake was low [below the population mean; OR = 0.79 (95% CI = 0.51–1.25)] ($P = 0.32$), whereas in the high-SFA intake stratum, the risk was higher and significant [OR = 2.10 (95% CI = 1.22–3.62)] ($P = 0.008$). In the BPRHS, these estimates were OR = 1.16 (95% CI = 0.74–1.82) ($P = 0.51$) for the low and OR = 1.75 (95% CI = 0.99–3.06) ($P = 0.05$) for the high-SFA intake stratum.

Finally, we examined the interaction effect of MUFA and PUFA intakes with the *FTO* polymorphism on BMI (**Supplemental Table 4**) and found consistent results in the GOLDN and BPRHS. PUFA intake did not interact with the *FTO* polymorphisms in determining BMI (*P*-interaction = 0.18 in GOLDN and *P*-interaction = 0.53 in BPRHS). However, we obtained significant interactions with MUFA in both the GOLDN and BPRHS participants (*P*-interaction = 0.012 and *P*-interaction = 0.021, respectively, for the categorical variables based on the population means). These results, mimicking that observed for

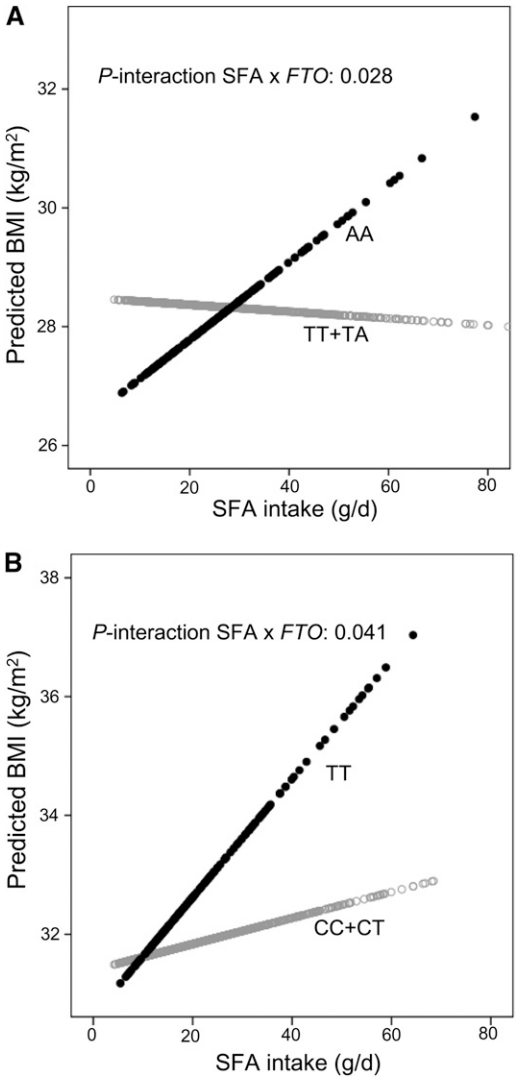

FIGURE 2 Predicted values of BMI by the *FTO* polymorphisms (recessive model) in the GOLDN study (*A*) and the BPRHS (*B*) plotted against the SFA intake ($n = 881$ TT+TA and $n = 188$ AA in GOLDN and $n = 917$ CC+CT and $n = 177$ TT in BPRHS). Predicted values were calculated from the regression models containing the SFA intake (as continuous), the *FTO* polymorphism, their interaction term, and the potential confounders (gender, age, smoking, drinking, PA, and total energy intake). The *P* value for the interaction term between SFA intake and the corresponding *FTO* polymorphism (rs9939609 in GOLDN and rs1121980 in BPRHS participants) was obtained in the hierarchical multivariate adjusted interaction model in which SFA intake was logarithmically transformed. BPRHS, Boston Puerto Rican Health Study; GOLDN, Genetics of Lipid Lowering Drugs and Diet Network; PA, physical activity.

SFA intake, may reflect the high correlation between the MUFA and SFA intake ($r_s = 0.94$; $P < 0.001$ for GOLDN and $r_s = 0.95$; $P < 0.001$ for BPRHS) in these populations.

Discussion

We found that carriers of the minor allele (obesity risk allele) of the *FTO* gene did not present a greater BMI than noncarriers, either in the GOLDN study or in the BPRHS when the population was analyzed as a whole, despite the fact that many other studies have described significant associations (1–8). It has been reported that the effects of *FTO* variation on BMI diminish as the age of participants increases (7,37). Because we were studying two middle-aged populations, the *FTO* effects on BMI may have been of a lesser magnitude than those on children or adolescents. Age could also have another influence insofar as we did not find any association between *FTO* gene variation and dietary intake, because the most significant results have been found in children (26,27,38) and recent literature does not provide a strong support for the effect in adults (38–40).

Moreover, we analyzed gene-environment interactions replicating previous findings and obtaining new interesting results. We first analyzed the interaction with PA, because there are numerous previous studies that reported that a greater level of PA could reduce the effects of the *FTO* risk allele in determining greater BMI (16–20,38,41). We found a significant interaction between *FTO* gene variation and PA in determining BMI in the participants of the GOLDN study but not in participants of the BPRHS. Although the validity of the PA variable may be limited because it was self-reported in both populations, our results in the GOLDN study successfully replicated the earlier observation that a high PA level attenuates the effect of the *FTO* risk alleles on BMI. Several confounding factors related to the specific characteristics of the BPRHS participants (different ethnic background, greater mean age, higher prevalence of obesity, etc.) could contribute to the differences in results. However, other studies have also not been able to find any interaction between PA and the *FTO* polymorphisms in determining BMI in other populations (22–24).

In contrast, we found more consistent results when we analyzed gene–diet interactions. In agreement with Sonestedt et al. (20), who described an interaction of *FTO* rs9939609 with total fat and carbohydrate intake in determining BMI in Swedish men, we found some significant gene–diet interactions with dietary fat intake. Both in participants of the GOLDN study and in those of the BPRHS, the effects of the *FTO* risk allele increasing BMI increased with a high-fat diet. These effects were examined by using different approaches to express nutrient intake, because in gene–diet interaction studies there is a controversy over which is the best way to express nutrient contribution (whether in g/d or in percent of energy) as well as over the choice of the cutoff points to create categories of intake, given that the results may differ (36). Having analyzed the different statistical models, the interaction with total fat intake was more significant and consistent with the results reported by Sonestedt et al. (20) in the GOLDN population than in participants of the BPRHS. One reason for these results may be that the GOLDN population is closer to the Swedish population in European genetic ancestry and age and in the amount of total fat intake than the BPRHS participants.

In addition, it is possible that some types of fatty acids had a greater effect than others in this interaction and that they were consumed more in the GOLDN population than in the BPRHS. When we studied the effect of the different types of fatty acids (SFA, MUFA, and PUFA) in greater depth, we observed stronger and more significant results on considering SFA intake. Both in participants of the GOLDN study and in those of the BPRHS, *FTO* polymorphisms significantly interacted with SFA intake (both as categorical and as continuous variables) in determining BMI in each population. These results suggested that high-SFA intake instead of total fat intake may be more relevant in increasing the effects of the *FTO* risk allele on BMI. Considering that Sonestedt et al. (20) did not examine the effect of SFA, this is the first time to our knowledge that a significant interaction between SFA intake and the *FTO* polymorphisms in determining BMI has been reported. Moreover, taking into account that we found this interaction in two independent populations, our results have a high level of both internal and external consistency. We obtained similar results for the interaction terms with MUFA intake due to the strong correlation that exists between MUFA and SFA consumption in North American populations. Additional studies in Mediterranean populations, where there are important differences in the sources of MUFA (mainly olive oil) and SFA (meats, milk, etc.), are required to specifically test the separate role of SFA and MUFA intake on this interaction. Nevertheless, the biological mechanisms underlying this interaction in determining BMI remain unknown and require directed molecular research.

Strengths of the present study include the analysis of interactions in two well-characterized independent populations, the use of well-validated dietary questionnaires, and the use of consistent models for statistical analysis. The main limitation derives form the cross-sectional study design.

In conclusion, our study confirms previous findings that total fat intake interacts with the *FTO* gene variation in determining BMI. On studying that interaction in greater depth, we found that the effects of SFA intake on modulating the association between the *FTO* risk-allele and higher BMI seem stronger than that of total fat. We found this interaction in two American populations, obtaining a greater level of consistency than for the interaction with PA.

Acknowledgments

D.C., D.K.A., K.L.T., and J.M.O. designed research; D.K.A., K. L.T., E.K.K., M.T., L.D.P., C-Q.L., Y-C.L., D.W., P.N.H., and J.M.O. conducted research; D.C., D.K.A., K.L.T., and J.M.O. analyzed and interpreted data; D.C. and J.M.O. wrote the paper; and J.M.O. and D.C. had primary responsibility for final content. All authors read and approved the final manuscript.

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