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Gene × dietary pattern interactions in obesity: analysis of up to 68 317 adults of European ancestry

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Abstract

Obesity is highly heritable. Genetic variants showing robust associations with obesity traits have been identified through genome-wide association studies. We investigated whether a composite score representing healthy diet modifies associations of these variants with obesity traits. Totally, 32 body mass index (BMI)- and 14 waist–hip ratio (WHR)-associated single nucleotide polymorphisms were genotyped, and genetic risk scores (GRS) were calculated in 18 cohorts of European ancestry (n = 68 317). Diet score was calculated based on self-reported intakes of whole grains, fish, fruits, vegetables, nuts/seeds (favorable) and red/processed meats, sweets, sugar-sweetened beverages and fried potatoes (unfavorable). Multivariable adjusted, linear regression within each cohort followed by inverse variance-weighted, fixed-effects meta-analysis was used to characterize: (a) associations of each GRS with BMI and BMI-adjusted WHR and (b) diet score modification of genetic associations with BMI and BMI-adjusted WHR. Nominally significant interactions ($P = 0.006$–0.04) were observed between the diet score and WHR-GRS (but not BMI-GRS), two WHR loci (*GRB14* rs10195252; *LYPLAL1* rs4846567) and two BMI loci (*LRRN6C* rs10968576; *MTIF3* rs4771122), for the respective BMI-adjusted WHR or BMI outcomes. Although the magnitudes of these select interactions were small, our data indicated that associations between genetic predisposition and obesity traits were stronger with a healthier diet. Our findings generate interesting hypotheses; however, experimental and functional studies are needed to determine their clinical relevance.

Introduction

The recent obesity epidemic is widely believed to be driven by typical Westernized lifestyles, consisting of diets low in nutrient quality and high in calories, along with physical activity levels insufficient to offset high-caloric consumption. Despite these general relationships, people living within the same obesogenic environment display substantial between-person variability in body weight. Responses to overfeeding or underfeeding have been shown to depend, at least in part, on genetic background (1–3), suggesting that genetic susceptibility to weight change interacts with a person's environment.

Driven by large-scale meta-analyses of genome-wide association study (GWAS) data, the past decade has witnessed rapid progress in the discovery of genetic variants associated with obesity-related traits (4,5). Although these associations are robust across unique samples, there is little empirical evidence that lifestyle factors modify the effects associated with these variants. Several observational studies show that physical activity may attenuate the genetic predisposition to obesity (6–11). However, it is not known whether it is physical activity alone or other lifestyle factors that correlate with physical activity, such as diet, that underlie these interactions (12–14). Characterizing how diet influences the associations of genetic variants with obesity-related traits in observational studies may help determine the extent that dietary interventions can offset a person's genetic susceptibility to obesity, and further, may inform the design of clinical trials that are specifically designed to test gene–diet interactions (e.g. genotype-based recall studies). Many published observational studies and clinical trials reporting gene–lifestyle interaction were not designed to test such interactions, and, thus, are underpowered (15).

We previously created a composite diet score, ranking individuals on their intakes of various foods to characterize a generally healthy dietary pattern (16). This approach, compared with one focused on single foods or nutrients, captures the highly complex nature of diet and translates more intuitively to public health. Using this score, we sought to determine whether the associations of established body mass index (BMI)- and waist–hip ratio (WHR)-associated variants, individually or combined, are modified by a composite diet score in adults of European ancestry.

Results

CHARGE diet score

A higher, compared with lower, diet score (reflecting a healthier diet) was associated with lower BMI and BMI-adjusted WHR in models adjusted for potentially confounding physical characteristics and lifestyle factors (Table 1; Supplementary Material, Figs S1 and S2).

Associations of BMI-GRS and WHR-GRS with BMI and WHR

The BMI-GRS and WHR-GRS were positively associated with BMI and BMI-adjusted WHR, respectively (Table 2). Each additional risk allele in the BMI-GRS was associated with an average of 0.116 kg/m^2 [standard error (SE): 0.005] higher BMI ($P = 1.97 \times 10^{-124}$;

Table 1. Associations of diet score with BMI and WHR in all participants and by sex

	Cohorts (*N*)	*N*	β	95% CI	P-value	I^2
Outcome: BMI						
All[a]	19	68 317	−0.034	(−0.0419, −0.0266)	<0.0001	83.9% (76.1%; 89.2%)
All[b]	19	66 493	−0.017	(−0.0247, −0.0088)	<0.0001	82.6% (73.9%; 88.4%)
Women[a]	17	46 916	−0.043	(−0.0540, −0.0316)	<0.0001	72.5% (55.4%; 83.1%)
Women[b]	17	45 796	−0.014	(−0.0257, −0.0025)	0.017	69.7% (50.3%; 81.6%)
Men[a]	18	29 992	−0.022	(−0.0320, −0.012)	<0.0001	79.0% (67.4%; 86.4%)
Men[b]	18	29 228	−0.014	(−0.0245, −0.0039)	0.007	77.8% (65.3%; 85.8%)
Outcome: BMI-adjusted WHR						
All[c]	17	58 393	−0.0010	(−0.0011, −0.0009)	<0.0001	21.7% (0%; 56.2%)
All[d]	17	57 666	−0.0007	(−0.0008, −0.0006)	<0.0001	0% (0%; 47.4%)
Women[c]	15	41 176	−0.0009	(−0.0010, −0.0007)	<0.0001	35.3% (0%; 65.1%)
Women[d]	15	40 116	−0.0006	(−0.0007, −0.0004)	<0.0001	29.6% (0%; 62.1%)
Men[c]	16	25 809	−0.0012	(−0.0013, −0.001)	<0.0001	0% (0%; 2.5%)
Men[d]	16	25 081	−0.0008	(−0.0010, −0.0007)	<0.0001	0% (0%; 0%)

[a]Associations adjusted for study center and/or family structure (as applicable), age, sex (where relevant) and kcal/day.
[b]Associations adjusted for study center and/or family structure (as applicable), age, sex (where relevant), kcal/day, education, physical activity, smoking and alcohol intake.
[c]Associations adjusted for study center and/or family structure (as applicable), age, sex (where relevant), kcal/day and BMI.
[d]Associations adjusted for study center and/or family structure (as applicable), age, sex (where relevant), kcal/day, BMI, education, physical activity, smoking and alcohol intake.

Table 2. Associations of BMI-GRS and WHR-GRS with BMI and WHR, respectively, in all participants and by sex

Group	Marker	Cohorts (N)	N	β	SE	P-value	Direction of association across cohorts
Outcome: BMI							
All[a]	BMI-GRS	18	57 075	0.116	0.005	1.97E−124	++++++++++++++++++
Women[a]	BMI-GRS	16	31 903	0.131	0.007	9.56E−72	++++++++++++++−+
Men[a]	BMI-GRS	17	25 172	0.102	0.007	1.36E−55	+++++++++++++++++
Outcome: BMI-adjusted WHR							
All[b]	WHR-GRS	17	54 294	0.0016	0.0001	2.15E−62	+++++++++++++++−+
Women[b]	WHR-GRS	15	30 196	0.0022	0.0001	1.14E−48	+++++++++++++++
Men[b]	WHR-GRS	16	24 098	0.0008	0.0001	1.55E−08	++−+−+++++++++−+

[a]Associations adjusted for study center and/or family structure (as applicable), age and sex (where relevant).
[b]Associations adjusted for study center and/or family structure (as applicable), age, sex (where relevant) and BMI.

Table 3. Interactions of diet score with BMI-GRS, WHR-GRS or select (<0.05)[a] individual SNPs for BMI or WHR (women and men combined)

	Nearest gene	Risk allele	$\beta_{\text{interaction}}$	SE	$P^{a}_{\text{interaction}}$	Direction of association across cohorts
Healthy diet score × BMI-GRS for BMI[b]	—	—	−0.0003	0.001	0.792	−−−+−−+−+−−++−−−−+
Healthy diet score × rs10968576 for BMI[b]	*LRRN6C*	G	0.0119	0.006	0.040	−++−−+−++−++++−+−+−
Healthy diet score × rs4771122 for BMI[b]	*MTIF3*	G	0.017	0.006	0.008	+−−?+++++−+−++++−−+
Healthy diet score × WHR-GRS for BMI-adjusted WHR[c]	—	—	4.77E−05	2.32E−05	0.040	+++++++−++++−−−−+
Healthy diet score × rs10195252 for BMI-adjusted WHR[c]	*GRB14*	T	1.74E−04	0.00008	0.028	++−+−+++++++++−−+
Healthy diet score × rs4846567 for BMI-adjusted WHR[c]	*LYPLAL1*	G	2.31E−04	0.00008	0.006	++−−−++++++++++++

The italicized values represents the *P* value.
[a]A priori alpha for interactions: diet score × SNP interactions <0.0018 for outcome BMI, diet score × WHR-GRS for outcome WHR < 0.025 and diet score × SNP interactions for outcome WHR < 0.0016.
[b]Interaction β adjusted for study center and/or family structure (as applicable), age, sex and kcal/day; see Supplementary Material, Table S5 for interactions $P > 0.05$, which ranged from 0.056 to 0.99.
[c]Interaction β adjusted for study center and/or family structure (as applicable), age, sex, kcal/day and BMI; see Supplementary Material, Table S6 for interactions $P > 0.05$, which ranged from 0.16 to 0.98.

equivalent to 335 g for a person 1.7 m tall), and each additional risk allele in the WHR-GRS was associated with a 0.002 (SE: 0.0001) higher BMI-adjusted WHR ($P = 2.1 \times 10^{-62}$). Results were directionally consistent between sexes, although the association of the WHR-GRS with WHR was more evident in women than men (Table 2), as expected given that the majority of these loci were discovered in women (4). Individual single nucleotide polymorphism (SNP) associations with BMI and BMI-adjusted WHR are reported in the total sample of males and females in Supplementary Material, Tables S3 and S4, respectively.

Gene × diet score interactions

Diet score did not modify the association of the BMI-GRS with BMI ($P_{\text{interaction}} = 0.79$; Table 3, Supplementary Material, Fig. S3A), whereas there was nominal evidence that a higher diet score (representing a healthier diet) strengthened the association of WHR-GRS with BMI-adjusted WHR ($\beta_{\text{interaction}}$ ($SE_{\text{interaction}}$) = 4.77E−5 (2.32E−5); $P_{\text{interaction}} = 0.04$; Table 3, Supplementary Material, Fig. S3B). In analyses modeling the interactions of each individual SNP and diet score on BMI and BMI-adjusted WHR, two tests of interaction for BMI and two for WHR were nominally statistically significant (Table 3, Supplementary Material, Tables S5 and S6). All four of these interaction effect estimates (βs) were also positive, again, indicating a stronger association between genotype and the respective outcome with higher diet score: *LRRN6C* rs10968576 ($P_{\text{interaction}} = 0.040$) and *MTIF3* rs4771122 ($P_{\text{interaction}} = 0.008$) for BMI, and *GRB14* rs10195252 ($P_{\text{interaction}} =$ 0.028) and *LYPLAL11* rs4771122 ($P_{\text{interaction}} = 0.006$) for BMI-adjusted WHR. However, these diet score × SNP interactions were not statistically significant after correction for multiple testing ($P < 0.0011$ based on Bonferroni correction for 46 tests).

Discussion

We conducted a broad assessment of the role of a multifactorial diet score on the genetic susceptibility to obesity by examining 32 common variants that have been reliably associated with BMI (5) and an additional 14 common variants that have been associated with BMI-adjusted WHR (4) in populations of European ancestry. Our study is the largest of its kind to date, utilizing a centrally designed and harmonized analysis plan and including cohorts with relatively diverse dietary habits and prevalence of obesity.

Overall, we observed nominal evidence of interaction between the WHR-GRS and the diet score, such that the GRS effect was stronger in those with higher versus lower diet scores. Similarly, we observed suggestive evidence that healthy diet augments the associations of variants in or near four loci with BMI (*LRRN6C* and *MTIF3*) and BMI-adjusted WHR (*GRB14* and *LYPLAL1*). While these observations counter the general hypothesis that healthy behaviors can offset risk, it is important to note that although genetic susceptibility was slightly more pronounced in those with healthier diets, at any one level of genetic susceptibility, the BMI or BMI-adjusted WHR was lower in those with healthier versus less-healthy diets (higher versus lower diet scores). Nevertheless, the nature of these interactions differs from that observed in studies on the modification of genetic effects by other lifestyle factors, such as those reporting an attenuating influence of physical activity on genetic predisposition to obesity-related traits (6–12,14). Proxy measures of both diet and physical activity contain an appreciable amount of random measurement error (17), requiring large sample sizes to achieve adequate statistical power. Most of the previous studies on physical activity were larger than the present analysis, and it is also possible that the true sizes of the interactions differ, with larger modifying effects of physical activity than of diet. Sources of systematic error (bias) also exist and are specifically relevant to studies of obesity; in such studies, bias can occur, for example, by over- or underreporting of dietary intake (or physical activity) in people who are over- or underweight, in part, because participants may be well aware of the links between lifestyle and body corpulence, and this awareness may impact their response to lifestyle-related questions. While the valid assessment of lifestyle is difficult in large cohorts, so too is differentiating the influence of the observed lifestyle factors and their unmeasured correlates on genetic susceptibility. Thus, further investigation is necessary to elucidate these dynamics, both in terms of study design and physiology, perhaps using more precise tools to assess diet or in more powerful studies of different design (e.g. intervention studies adequately powered to test gene–treatment interactions).

Previous studies involving the genetic regions highlighted in our analyses [*LYPLAL1* (18,19), *MTIF3* (20,21), *GRB14* (22,23) and *LRRN6C* (24)] delineate their roles in physiology (see also Supplementary Material, Table S7), but few studies have investigated how diet might interact with these loci to influence body composition (25–28). While one longitudinal observation study reported no interactions between various lifestyle factors and *LYPLAL1* variation (27), the Diabetes Prevention Program (DPP), an intensive lifestyle intervention study, did observe evidence of interaction on weight change at this locus (25). Specifically, of the 12 loci examined in the DPP study, *LYPLAL1* (rs2605100, $r^2 = 0.48$ with rs4846567) was one of three loci for which the test of interaction was statistically significant: the G (versus A) allele conveyed greater short-term weight loss following lifestyle intervention versus control intervention (~0.34 kg per G allele from baseline to 6 months) (25). The Look AHEAD Study (26) examined relationships between 12 obesity-associated gene variants, including *MTIF3* (rs7988412, $r^2 = 0.68$ with rs4771122), and caloric intake and eating patterns at baseline. The authors found no association between the variant and baseline caloric intake ($P = 0.99$) or the number of eating occasions ($P = 0.62$). However, in a joint analysis from the DPP and Look AHEAD trials, all loci studied in the present report were examined for interaction with intensive lifestyle modification in relation to weight loss (up to 4 years post-randomizations) (the DPP and Look AHEAD Study groups, personal communication, P.W. Franks). Of the loci studied, the one with the strongest evidence for gene–lifestyle interaction on weight loss was the *MTIF3* rs1885988 variant ($r^2 = 0.72$ with the rs4771122 variant studied here). There are no other reports in the published literature on gene–diet interactions for obesity at the *LYPLAL1* or *MTIF3* loci to our knowledge.

Like most clinically prescribed weight-conscious diets, both the Look AHEAD and DPP lifestyle interventions were designed around general principles of healthy eating, each focusing on calorie and fat goals to guide healthy food selections. In a similar sense, our diet score broadly captures several dietary characteristics; therefore, neither the clinical trials data nor those from our analyses allow us to speculate on the effect modifying roles of individual dietary components. Hence, it is possible that a reductive approach (one focused on individual foods or nutrients) might identify interactions of different magnitudes and directions that could be masked by combining these into a summary score, such as we have done. However, studying each component of the score separately would require many more hypothesis tests, which we concluded that our study is not powered to accommodate. Further, studies that characterize diet more broadly (i.e. as multiple-component dietary patterns) are more easily applied to public health. Similarly, while the GRS allow assessment of overall genetic susceptibility, studying the role of individual variants within the GRS may provide insight into the biology that potentially underlies any observed interactions.

Taking variants that were top ranked in marginal effects GWAS meta-analyses and testing these for interactions with environmental exposures, as we did here, is a pragmatic data reduction strategy; this is so because those variants (or the loci that they tag) are, with high probability, likely to reside on causal pathways for the traits of interest. Although this does not necessarily mean that those variants should interact with environmental exposures, many argue that it is a hypothesis worth testing. In all likelihood, many other variants, which would not be picked up by marginal effects tests, but which modulate the effects of environmental exposures, exist (29).

The present report, alongside others, points to *MTIF3* as a region that may interact with dietary factors to influence aspects of adiposity. The remaining results suggest that diet, as represented by our composite score, does not appreciably modify the effects of several loci, singly or collectively, on BMI and BMI-adjusted WHR. This area of research would benefit from future studies that utilize more detailed and precise information on dietary intake, alternative study designs (such as interventions) and other genetic regions that do not reach genome-wide statistical significance in main effects GWAS.

Materials and Methods

This project was coordinated by the Nutrition Working Group of the Cohorts for Heart and Aging Research in Genomic Epidemiology (CHARGE) consortium (30). Each of the 18 contributing cohorts executed analyses locally according to a uniform analysis plan and shared summary statistics with a central data hub for meta-analyses. One of these cohorts, DIetary, Life style and Genetic determinants of Obesity and Metabolic syndrome (DILGOM), provided two independent samples (metabochip and GWAS samples) that were analyzed separately. The 18 cohorts, providing up to 68 317 adults, are described in Table 4. Written informed consent and institutional review board approvals were obtained locally by each participating study. All studies were conducted in accordance with the Helsinki Declaration of 1975 as revised in 1983.

Anthropometry

BMI was calculated as weight in kilograms (kg) divided by height in meters squared (m^2). In all except two cohorts, body weight and height were measured by clinical staff at the examination sites; in the Nurses Health Study (NHS) and the Health Professionals Follow-Up Study (HPFS), height and body weight were self-reported by questionnaire (correlation of self-reported with directly measured values: $r = 0.97$) (31). Waist and hip circumferences, used to calculate WHR, were directly measured in 15 cohorts, self-reported in two cohorts (NHS and HPFS), and unavailable in two other cohorts (GLACIER and Health ABC).

Dietary intake and the diet score

Self-reported dietary intake was assessed by food frequency questionnaire (13 cohorts), by a combination of food frequency questionnaire and diet records (one cohort), or by diet records (four cohorts) (Supplementary Material, Table S2). The methods

Table 4. Clinical characteristic of the study participants by cohort

Cohort	Abbreviation	Country	Exam year[b]	N[c]	BMI (kg/m^2)		WHR (cm/cm)		Age (years)		Sex	Smoking	Energy intake (kcal/day)		Diet Score[c,d]		
					Mean	SD	Mean	SD	Mean	SD	% Women	% Current	Mean	SD	Range	Mean	Median
Atherosclerosis Risk in Communities Study	ARIC	USA	1987–1989	8586	26.7	4.6	0.92	0.08	54.2	5.7	53.7	24.4	1642	604	1–27	13.7	13
Cardiovascular Health Study	CHS	USA	1989–1990	2761	26.0	4.3	0.91	0.09	72.3	5.4	62.1	11.5	2016	648	1–27	13.7	14
DIetary, Life style, and Genetic determinants of Obesity and Metabolic Syndrome	DILGOM (metabochip sample)	Finland	2007	3467	26.6	4.5	0.91	0.09	52.4	13.5	52.0	22.4	2313	783	3–27	13.8	14
DIetary, Life Style, and Genetic determinants of Obesity and Metabolic Syndrome	DILGOM (GWAS sample)	Finland	as above	604	26.9	4.7	0.91	0.09	51.5	13.4	55.2	19.2	2533	912	2–25	13.7	14
Family Heart Study	Family HS	USA	1992	3185	27.4	5.3	0.91	0.09	51.4	13.6	53.6	14.7	1749	615	1–26	13.2	13
Framingham Offspring Study and Framingham Third Generation Study	Framingham	USA	1991–1995	5827	26.7	5.0	0.89	0.09	46.1	11.5	54.6	17.2	1982	662	0–26	13.7	14
Gene–Lifestyle interactions and Complex Traits Involved in Elevated Disease Risk Study	GLACIER	Sweden	1985–2007	5277	25.7	4.0	NA	NA	49.2	8.6	61.4	22.3	1762	605	0–24[d]	11.6	12
Health 2000	Health 2000	Finland	2000–2001	1935	27.3	4.5	0.92	0.08	50.5	10.9	51.4	28.8	2245	783	2–27	13.7	14
Health, Aging and Body Composition	Health ABC	USA	1997–1998	1266	26.2	4.0	NA	NA	74. 8	2.8	50.6	6.3	1807	599	3–27	15.7	16
Health Professionals Follow-Up Study	HPFS	USA	1986	896	25.0	2.7	0.94	0.05	55.0	8.3	0	10.6	2041	604	2–26	13.6	13
Helsinki Birth Cohort Study	HBCS	Finland	2001–2004	1584	27.4	4.4	0.92	0.09	61.5	2.9	58.4	24.4	2238	821	2–26	13.3	13
Invecchiare in Chianti	InCHIANTI	Italy	1997	991	27.1	4.1	0. 90	0.08	67.0	15.4	56.2	19.0	2036	595	2–20[d]	10.7	11
Malmö Diet and Cancer Study	Malmö	Sweden	1991–1996	20319[e]	25.4	3.8	0.85	0.09	58.5	7.6	59.4	27.6	2342	581	1–26	13.7	14
Multi-Ethnic Study of Atherosclerosis	MESA	USA	2000–2002	2146	27.5	4.9	0.92	0.08	62.6	10.3	52.4	11.0	1699	718	1–27	13.6	14
Nurses Health Study	NHS	USA	1986	1187	24.8	4.5	0.77	0.07	54.0	6.7	100	11.6	1787	510	1–25	13.5	13
Rotterdam Study	Rotterdam	The Netherlands	1990–1993	3932	26.2	3.6	0.90	0.09	67.1	7.6	58.3	23.4	1985	509	1–21[d]	10.1	10
The Hellenic Study of Interactions between SNPs and Eating in Atherosclerosis Susceptibility	THISEAS	Greece	2006–2010	543	28.2	4.6	0.92	0.10	55.9	13.6	48.5	33.6	1778	1023	0–26	11.9	12
Uppsala Longitudinal Study of Adult Men	ULSAM	Sweden	1991–1995	932	26.0	3.2	0. 94	0.05	71.0	0.6	0	19.5	1774	449	3–24	13.5	13
Cardiovascular Risk in Young Finns Study	YFS	Finland	2007	1709	25.8	4.5	0.88	0.09	37.8	5.0	55.4	28.1	2386	769	2–26	13.6	14

Cohort names (as listed also in Table 4): ARIC, Atherosclerosis Risk In Communities Study; CHS, Cardiovascular Health Study; DILGOM, DIetary, Life Style, and Genetic determinants of Obesity and Metabolic Syndrome; Family HS, Family Heart Study; Framingham, Framingham Offspring Study and Framingham Third Generation Study; GLACIER, Gene–Lifestyle interactions and Complex Traits Involved in Elevated Disease Risk Study; Health 2000 (no abbreviation used); Health ABC, Health, Aging and Body Composition; HPFS, Health Professionals Follow-Up Study; HBCS, Helsinki Birth Cohort Study; InCHIANTI, Invecchiare in Chianti; Malmö, Malmö Diet and Cancer Study; MESA, Multi-Ethnic Study of Atherosclerosis; NHS, Nurses Health Study; Rotterdam, Rotterdam Study; THISEAS, the Hellenic Study of Interactions between SNPs and Eating in Atherosclerosis Susceptibility; ULSAM, Uppsala Longitudinal Study of Adult Men; YFS, Cardiovascular Risk in Young Finns Study.

[a]More information on populations and study designs can be found online Supplementary Material, Table S1.

[b]Single examinations spanned several years (single time points); see Supplementary Material, Table S1 for more information.

[c]Largest contributing value; analysis-specific sample sizes presented in subsequent corresponding tables.

[c]Diet score = sum of quartile ranks of nine food groups (exceptions noted in Footnote e). Favorable: whole grains, fish, fruit, vegetables, nuts = 0–3 points per ascending quartile; Unfavorable: red or processed meats, desserts and sweets, sugar-sweetened beverages, fried potatoes = 0–3 points per descending quartile.

[d]Diet score in select cohorts is based on eight, instead of nine, food groups; no data collected on fried potatoes (InCHIANTI & Rotterdam) or nuts (GLACIER).

[e]Sample varies widely in SNP-based analyses; see other tables.

and rationale behind the construction of the CHARGE diet score and its criterion validity for predicting fasting glucose and insulin concentrations have been described in detail (16). Intakes of foods/beverages were modeled in servings per day for all cohorts except the sample from the Uppsala Longitudinal Study of Adult Men (ULSAM), where grams per day were used. Briefly, the score is based on the cohort-specific quartile ranks of nine food/beverage groups, where favorable food groups including fruits (not including juices), vegetables (not including white potatoes), whole grains, fish and nuts were assigned values of 0–3 according to ascending quartile ranks, and unfavorable food/beverage groups including red or processed meats, desserts and sweets, sugar-sweetened beverages and fried potatoes were assigned values of 0–3 according to descending quartile ranks. The resulting score is a continuous variable with a theoretical range of 0–27, where a higher diet score represents healthier food and beverage choices (Table 4).

SNP selection, genotyping and genetic risk scores (BMI-GRS and WHR-GRS)

At each SNP locus, genotypes were coded as 0, 1 and 2 or imputed to indicate the number of risk alleles for the 32 and 14 variants that have been previously associated with BMI (5) and WHR (4), respectively (SNPs listed in Supplementary Material, Table S3). For each participant, a genetic risk score (GRS) was then calculated by summing up the number of risk alleles separately for the BMI and WHR SNPs. In cohorts where genotypes were directly assessed (i.e. not imputed), missing genotypes were estimated in participants with >70% genotype information available by using mean imputation, as described previously (32) (Supplementary Material, Table S1).

The BMI-GRS was not calculated in the sample from DILGOM that was genotyped using the Metabochip, owing to a high number of missing SNPs (with no suitable proxy). In three cohorts, the BMI-GRS was based on 31 SNPs [Malmö Diet and Cancer Study (MDC), the Hellenic Study of interactions between SNPs and Eating in Atherosclerosis Susceptibility (THISEAS) and Young Finns Study (YFS)], owing to the absence of one SNP. The WHR-GRS was calculated in all cohorts except those with no WHR data (GLACIER and Health ABC); in MDC, the WHR-GRS was based on 13 rather than 14 SNPs, owing to the absence of one SNP. The approximate mean (SD) across cohorts for the BMI-GRS and WHR-GRS were 28.2 (3.5) and 14.3 (2.4), respectively.

Statistical analysis

Statistical analyses were conducted within each study according to a uniform analysis plan and subsequently meta-analyzed (details below).

Associations of diet score with BMI and WHR

The associations between diet score and BMI and WHR were calculated using multivariable linear regression, with the diet score modeled as a continuous exposure, adjusting for age, sex (where relevant), energy intake (kcal/day) and study center and/or population substructure (as necessary); where WHR was the outcome of interest, BMI was included as an additional covariate (BMI-adjusted WHR). In a second model, associations were further adjusted for education, physical activity, smoking and alcohol intake. Sex-stratified analyses were also conducted using these models. Details concerning the methods used to assess and characterize lifestyle within cohorts are provided in Supplementary Material, Table S1.

Associations of GRS and individual loci with BMI and WHR

Associations of the individual BMI- and WHR-relevant SNPs and BMI- and WHR-GRSs with their respective outcomes were also calculated using multivariable linear regression, adjusting for age, sex and field center and/or population substructure; as with the individual SNP models, where WHR was the outcome of interest, BMI was also included among covariates (BMI-adjusted WHR). Sex-stratified analyses were also conducted for BMI and WHR-adjusted BMI traits, respectively.

Diet score interactions with GRS and individual loci

Interactions were assessed by including a product term (diet score × SNP or diet score × GRS) in the regression models, adjusting for age, sex, energy intake (kcal/day) and study center and/or population substructure (as needed); as above, where WHR was the outcome, models were additionally adjusted for BMI (BMI-adjusted WHR). To maximize sample size (and by proxy, statistical power) for interaction tests, sex-stratified analyses were not conducted.

Meta-analyses. Summary statistics from each cohort were combined using inverse variance-weighted, fixed-effects meta-analysis. Meta-analyses for the diet score associations with BMI or WHR were performed using the *rmeta* package (version 2.16) in R 2.13.1 (http://www.R-project.org/). Meta-analyses of the interactions and main effects of SNP and GRS tests were conducted using METAL (http://www.sph.umich.edu/csg/abecasis/Metal/index.html). Heterogeneity was assessed by the I^2 statistic (33). Meta-regression was used to explore sources of heterogeneity in the meta-analyses using the metafor package in *R*. Meta-regression included region (Europe versus USA) and sex ratio as cohort-specific covariates. The meta-regression did not indicate either region or sex ratio as sources of heterogeneity ($P > 0.48$).

Supplementary Material

Supplementary Material is available at *HMG* online.

Conflict of Interest statement: D.M. reports *ad hoc* honoraria or consulting from Nutrition Impact, Amarin, Astra Zeneca, Boston Heart Diagnostics and Life Sciences Research organization; and scientific advisory board, Unilever North America. All other authors declare no competing interests.

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DIetary, Life style, and Genetic determinants of Obesity and Metabolic syndrome (DILGOM)

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Gene–lifestyle interactions and complex traits involved in elevated disease risk (GLACIER)

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