

Genes Versus Environment

The relationship between dietary fat and total and central abdominal fat

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OBJECTIVE — The influence of diet on body fat has not been quantified independently of genetic influences, although both are held to contribute to regulation of body fat stores. This study examined 1) the relationship between recent diet and total body and central abdominal fat in middle-aged female twins independent of genetic and important environmental factors and 2) evidence of interaction between diet and genetic predisposition.

RESEARCH DESIGN AND METHODS — Measurements in 436 healthy female twins (aged 58 ± 10 years) included dietary intake by food frequency questionnaire (validated against a 7-day food diary, $n = 162$), BMI, total body and central abdominal fat by dual-energy X-ray absorptiometry, and environmental covariates (smoking habit, hormone replacement, and physical activity) by standardized questionnaire. Dietary energy underreporters were excluded.

RESULTS — Intake of dietary fat (total and subtype) and carbohydrates was not related to BMI or to total or central fat, confirmed in quintile analysis. With genetic and environmental factors controlled in 90 monozygotic pairs, differences in the intake of energy, fat, or protein were not related to intrapair differences in total and central body fat. However, a minor inverse relationship between carbohydrate intake and total adiposity was confirmed ($r = -0.25$, $P = 0.02$). In paired analyses, the twin with the higher intake of total sugars had significantly lower total body and central abdominal adiposity. There was no evidence of a gene-environment interaction between intake of fat or carbohydrates contributing to greater body fat mass in subjects genetically predisposed to obesity.

CONCLUSIONS — Using validated dietary measures and direct measures of body fat and excluding underreporters, no relationship between dietary fat and body fat was found in middle-aged women, particularly after controlling for genetic and some environmental factors. The role of dietary factors in determining total body and central abdominal fat appears to have been overestimated in past cross-sectional studies.

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Obesity underlies a substantial proportion of cases of type 2 diabetes and is increasing in prevalence. In particular, central adiposity predicts the development of type 2 diabetes (1,2), independent of generalized adiposity. The contribution of diet to the long-term regulation of body fat stores is

not definitely established, nor is its role in the pathogenesis of obesity, despite current recommendations for reducing dietary fat as a strategy for attaining and maintaining healthy weight. The increasing prevalence of obesity despite evidence of declining dietary fat intake (3) and the failure of dietary fat

reduction in producing long-term weight reduction (4) challenge the primacy of dietary fat over other nutrients and/or environmental factors in causing obesity.

Several observational studies suggest a causative link, using varied measures of dietary intake and often indirect estimates of body fatness (5–11), but are not supported by other large (12) or prospective (6) studies or recent critical opinion and review (4,13,14). Prospective studies report an association between dietary fat intake and subsequent weight gain (15–17), but a more minor one than is usually appreciated. For example, in the hierarchy of environmental influences, dietary factors were subordinate to physical activity in predicting weight gain when this important covariate was assessed (16). The primacy of dietary fat over other macronutrients in predicting weight gain is also unsubstantiated, and sex differences are apparent: in overweight women, the relative risk of gaining 5 kg over 5 years was greatest in those with the highest intake of not only fat, but also carbohydrate and energy (15); in men, dietary intake did not predict weight gain (15).

Methodological deficiencies limiting the conclusions from the major reports include limited dietary assessments (5,10,11,17) (often with little data validation), indirect estimates of adiposity, such as BMI (5–8) (derived from self-reported weight and height in one study [6]), small sample size (9), selection bias from studying predominantly overweight or obese individuals (15,18), and, universally, no detection and exclusion of underreporters. Underreporting subjects more frequently report lower energy intake (19) or have a history of dieting (whether normal weight or overweight) (20) and may bias relationships observed, particularly if underreporting is selective for fat, as has been suggested (21,22).

Studies of dietary intake and central adiposity are limited by the adequacy of dietary assessment and the indirect estimation of central adiposity (9,11). When central fat was measured directly, no specific influence of dietary fat (by 3-day record) was found (23).

Genetic factors are strong determinants of total body and central abdominal fat, explaining ~50 and ~60%, respectively, of

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Abbreviations: BEE, basal energy expenditure; DEXA, dual-energy X-ray absorptiometry; DZ, dizygotic; %EI, percentage of energy intake; FFQ, food frequency questionnaire; GEE, generalized estimating equation; HRT, hormone replacement therapy; MZ, monozygotic.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

the population variance in middle-aged females (24). The direct influence of diet on total and central abdominal fat has not previously been examined independent of genetic factors, but it can be quantitated using the discordant monozygotic (MZ) twin model (co-twin case control study design). This experimental model allows for the estimation of the effect of one discordant environmental factor in twin pairs who are genetically identical and concordant for other environmental factors. Using this model, the influence of dietary factors can be estimated, isolated from the additional covariates that usually confound other studies.

Finally, Astrup et al. (25) have postulated that a high-fat diet will produce obesity only in genetically predisposed subjects, but not in "normal" subjects, suggesting that a 1.6% difference in fat intake cross-sectionally could be associated with a 10-kg greater fat mass in such people. This hypothesis, that only genetically predisposed individuals are susceptible to obesity in the presence of a high-fat diet, can be examined in the twin model by testing for such a gene-environment interaction as previously described (26–28).

We therefore studied a cohort of female twins to 1) determine the cross-sectional relationships between nutrients and body composition, including total body and central abdominal fat; 2) quantify the influence of dietary intake independent of genetic and other environmental factors; and 3) examine whether a higher intake of dietary fat is associated with greater body fat in those genetically predisposed to adiposity (i.e., a gene-environment interaction).

RESEARCH DESIGN AND

METHODS — Volunteers were a twin subset of PHENOBASE, a database of intermediate phenotype and genetic information on unselected twins and affected sibships, recruited from a national media campaign to participate in a twin study of osteoporosis at St. Thomas' Hospital, London (29). Subjects gave informed consent, and the study was approved by the local research and ethics committee. Volunteers were unaware of any hypotheses regarding body fat or nutrition.

Twin pairs were assessed on the same day. Zygosity was determined by standardized questionnaire (30) and confirmed with multiplex DNA fingerprinting using the AmpFL STR Profiler polymerase chain reaction amplification kit (PE Applied Biosystems, Foster City, CA).

Weight and height were measured with the subject barefoot in light street clothes: weight to the nearest 0.1 kg and height to the nearest centimeter. Body composition was measured by dual-energy X-ray absorptiometry (DEXA) (Hologic QDR, Waltham, MA) (24). Central abdominal fat was determined by scan reanalysis by a single blinded investigator (K.S.) and is described elsewhere (24,31). The region was defined by cursor manipulation as that extending from the upper border of the second lumbar vertebra to the lower border of the fourth and laterally to the inner border of the ribcage. Central abdominal fat (mass and percentage) were calculated using the standard software calculation. This region contains maximal intra-abdominal fat in postmenopausal women (32), despite some subcutaneous fat inclusion, and has been reported by our group to be strongly related to insulin resistance (by hyperinsulinemic-euglycemic clamp) (33). Total and central abdominal fat measurements are expressed in kilograms and as a percentage of body mass (percent central abdominal fat is the percentage of fat in the total tissue mass in the regional window).

Energy intake and dietary composition were assessed in all subjects using the Oxford-type food frequency questionnaire (FFQ) (34) and a 7-day food diary (35) in a randomly selected subset of 162 subjects. Both dietary assessments were self-administered after instructions from trained nurses. The FFQ recorded the pattern of dietary intake for the last year and was based on the instrument used in the Nurses' Health Study (36). Lists of foods given in the questionnaire were modified by changing American food names to their British equivalent and by using the National Food Survey to identify additional foods that are important sources of nutrients in the average British diets (37). The questionnaire specified standard serving sizes and frequency of consumption, from which intake of each food was calculated. Serving sizes in the questionnaire were specified (slices, teaspoons, pints) or defined as portions that were based on published values (38). Missing answers in FFQ questionnaires were left missing in FFQ analyses. These were very infrequent, and <1% for total sugars. In FFQs where subjects left >10 answers missing, these subjects were excluded from any further analyses.

The 7-day food diary used estimates of portion size (based on published values [38]), demonstrated in numerous sets of

photographs included in each diary (34).

Both dietary assessments were analyzed using the European Prospective Investigation in Cancer and Nutrition (EPIC) Group nutrient database (Institute of Public Health, University of Cambridge, Cambridge, U.K.), which is based (with some modifications) on the Royal Society of Chemistry supplements of McCance and Widdowson's *The Composition of Foods* (39). The FFQ were analyzed using the Compositional Analyses for Food Frequency Estimates (CAFE) program (version 17-1). The coded responses of the 7-day food records were entered using the Data In, Diet Out program (40). Both dietary methodologies have been validated against weighed 16-day records (34) and 24-h urinary nitrogen and other biological markers (35). Dietary composition is given in both grams per day and as a percentage of energy intake (%EI).

Dietary assessment data were further validated in three ways: first, by confirming the necessary relationships between energy and weight, then energy and diet composition (41); second, by cross-checking FFQ data against that from food diaries (validity was accepted if correlation coefficients were >0.4, as suggested by Willett [42]); and third, by identifying and excluding underreporters. Basal energy expenditure (BEE) was estimated by use of a mathematical model (the Garby formula using measures of body composition) (22). Underreporters were identified as those whose BEE exceeded reported energy intake.

This study further minimized bias by studying a healthy group unaware of any hypothesis regarding nutrition or fat. Finally, macronutrients are also expressed as a ratio of energy intake, a measure less sensitive to nutrient-specific underreporting than intake in grams (43).

Menopausal status, smoking habits, use of hormone replacement therapy (HRT), and physical activity were ascertained by standardized questionnaires. Menopause was defined as amenorrhea exceeding 12 months. Only subjects with known smoking and history of HRT were included. Physical activity was scored using a standardized questionnaire assessing activity at home, time spent in sporting activities per week, and weekly physical activity of an intensity to produce sweating (44). No subject had a history of diabetes or was receiving antidiabetic medication.

The relationship between dietary intake by FFQ and total and central fat

were examined in the cohort as a whole, using simple correlations, and in multiple regression to test for independence from other lifestyle factors, including smoking, HRT, and physical activity. As a further attempt to detect any relationships between diet and body fat, tertile and quintile analyses were also performed. Because both of these were negative, only quintile analyses are reported. Characteristics in twin pairs are not independently determined (because of common genetic and environmental influences). To correct for this possible bias, the generalized estimating equation (GEE) modeling approach was used (45), which allows for the correlation within twin pairs and yields consistent estimates of linear association terms and their corresponding standard errors. If this is not used, standard errors may be underestimated, and consequently, *P* values may be overstated.

MZ twin pairs provide a powerful tool for investigating the influence of environmental factors, since intrapair differences in body composition in genetically identical individuals must be due to differences in environment. The influence of diet was estimated using MZ pairs (using the co-twin case control study design) in two ways. First, the absolute intrapair differences in dietary composition were regressed against the intrapair variance in body composition through the origin (46). Second, the data were examined to evaluate the effect of increasing differences in dietary intake on body fat in genetically identical individuals and, conversely, in MZ pairs with increasing differences in body fat, to determine whether this may be related to differences in dietary intake. Thus, 1) MZ twin pairs concordant or discordant for dietary components were compared for intrapair differences in body fat; 2) MZ pairs concordant or discordant for body fat were compared for intrapair differences in dietary intake using analysis of variance (47); and 3) within each MZ pair, each twin was assigned to a higher or lower intake for each of dietary fat, carbohydrate, and total sugars (both grams per day and as %EI). Total body and central abdominal fat (kilograms and percent) were compared within each twin pair using paired *t* tests. To eliminate other known environmental influences, such as smoking status and estrogen replacement therapy (47), only MZ pairs concordant for these environmental factors were included in the MZ analyses.

The possibility that dietary fat only increases body fat in a genetically predis-

Table 1—Body composition and dietary intake (by FFQ and 7-day food records) in British middle-aged female twins

	Cohort characteristics	FFQ	7-day food diary	Correlations*
<i>n</i>	436	436	162	162
Age	57.7 ± 6.5	—	—	—
Weight	63.3 ± 9.9	—	—	—
BMI	24.3 ± 3.6	—	—	—
Total body fat				
kg	24.86 ± 8.07	—	—	—
%	38.2 ± 7.2	—	—	—
Central fat				
kg	1.56 ± 0.67	—	—	—
%	36.8 ± 10.0	—	—	—
Fat free mass (kg)	37.0 ± 4.5	—	—	—
Energy (kcal)	—	2,017 ± 651	1,853 ± 359	0.42†
Diet composition				
Total dietary fat	—	34.0 ± 20.5	34.3 ± 5.3	0.46†
Saturated	—	13.2 ± 8.8	—	—
Polyunsaturated	—	6.0 ± 3.0	—	—
Monounsaturated	—	11.9 ± 8.9	—	—
Carbohydrate	—	49.6 ± 17.0	48.2 ± 6.1	0.53†
Total sugars	—	25.6 ± 10.3	23.7 ± 6.2	0.56†
Protein	—	17.8 ± 8.9	16.4 ± 2.8	0.50†
Alcohol	—	2.0 ± 3.6	2.4 ± 3.8	0.92†

Data are means ± SD. Diet composition is expressed as a %EI. *Simple correlation coefficients: FFQ versus 7-day food records. †*P* < 0.0001.

posed subgroup has been raised (25,48). Thus, we investigated whether such a gene-environment interaction between genetic predisposition and dietary intake existed in this population. Using twin data, genetic susceptibility can be defined by the phenotype of a twin's co-twin (27,28) (in the current absence of validated susceptibility loci or genes). Thus, if a twin's co-twin is obese, it can be assumed that the twin also has a high genetic risk for being overweight. Data were analyzed as follows: degree of genetic risk for obesity was defined by the co-twin's ranking in tertiles of body fat for the whole cohort: highest risk 30.0–65.0 kg and lowest risk 8.6–20.6 kg body fat, with mean ± SD corresponding BMI values of 27.3 ± 2.9 and 21.2 ± 1.65 kg/m², respectively. That is, twin 1 had a high genetic risk for obesity if her co-twin's body fat was in the highest tertile and a low risk if the co-twin was in the lowest tertile. Only twin 1 (randomly chosen) was included in subsequent analyses. Only MZ and dizygotic (DZ) pairs concordant for smoking and HRT were included in these analyses. Dietary fat intake (in both grams per day and percentage of daily energy intake) was categorized into high and low intake based on tertiles (high

intake: 85–264 g/day, 35–55%; low intake: 19–61 g/day, 15–31%). After categorization for genetic risk and nutrient intake, there were 28 subjects in the low genetic risk tertile and 30 subjects in the high-risk tertile. The interaction between genetic risk category and dietary fat intake was examined using a two-factor analysis of variance, assessing for multiplicative effects of genetic risk category and dietary intake. That is, a gene-environment interaction was present not only if a higher intake of dietary fat was related to a greater body fat mass in the genetically predisposed, but if this increment in fat mass was significantly greater than that found in the low risk group. This method was repeated for central obesity and dietary fat intake. Again, genetic risk for central adiposity for twin 1 was defined by the central fat rank of her co-twin using tertile measures for central fat mass: highest risk 1.78–4.12 kg, lowest risk 0.31–1.22 kg. Data are presented with standard errors (± SEM).

RESULTS — The mean age (± SD) of the 436 twin women was 57.7 ± 6.4 years; 90% were postmenopausal, with a menopause duration of 10.4 ± 6.7 years

Table 2—Relationships between intrapair differences in dietary intake and intrapair variance in total body and central abdominal fat in 90 MZ middle-aged female twin pairs

Difference in dietary factor	Intrapair variance in total fat mass	Intrapair variance in central fat mass
Energy intake (kcal)	0.17	0.11
Dietary fat (%)	0.10	0.10
Saturated fat (%)	0.02	0.001
Polyunsaturated fat (%)	0.13	0.13
Monounsaturated fat (%)	0.15	0.15
Carbohydrate (%)	-0.25*	0.19
Total sugars (%)	0.12	0.08
Protein (%)	0.12	0.02

Energy and macronutrient intake were measured by FFQ. * $P = 0.02$.

(Table 1). HRT was used by 21% of subjects, and 15% smoked. There were 332 MZ and 104 DZ subjects. Dietary data are presented in Table 1.

Data validation

Simple correlations were used to assess dietary data validity. Energy intake by FFQ was related to weight ($r = 0.11$, $P = 0.03$), dietary fat (grams: $r = 0.89$, $P = 0.0001$; percent: $r = 0.26$, $P = 0.001$), and carbohydrate (grams: $r = 0.91$, $P = 0.0001$). In subjects with both FFQ and 7-day records ($n = 162$), energy and macronutrient intakes were similar and significantly related (Table 1). Underreporters were excluded ($n = 27$); they had a higher BMI (26.0 ± 0.8 vs. 24.2 ± 0.2 kg/m², $P = 0.01$) and fat mass (28.4 ± 2.0 vs. 24.6 ± 0.4 kg, $P = 0.02$).

Cross-sectional analysis

Across the cohort as a whole, there was no relationship found between dietary fat intake (%EI) and weight ($r = 0.13$, $P = 0.16$), total body fat ($r = 0.07$, $P = 0.18$), central abdominal fat ($r = 0.06$, $P = 0.22$), or fat-free mass ($r = 0.10$, $P = 0.08$), using GEE modeling. While a weak relationship was detected between BMI and dietary fat intake (%EI) ($r = 0.11$), it was not significant after GEE analysis ($P = 0.28$). No association was found between type of dietary fat and total body or central abdominal fat (data not shown). Similarly, no relationship was found between carbohydrate intake (%EI) and body fatness ($r = -0.11$, $P = 0.67$). Results did not differ if macronutrient intake was expressed as grams per day (data not shown).

Alcohol intake (in grams per day) was associated with a lower total body fat mass ($r = -0.15$, $P = 0.01$), but not with central fat mass ($r = -0.09$, $P = 0.10$). No relationship was found between fat-to-carbo-

hydrate ratio and total body or abdominal central fat (data not shown).

In multiple regression models, including smoking, HRT use, and physical activity, dietary fat intake still did not relate to total body or central abdominal fat (data not shown). In this model, there was a nonsignificant trend for an inverse relationship between sugar intake (%EI) and total body adiposity ($\beta = -0.18$, $P = 0.07$). Of all variables entered in the model, physical activity was the strongest predictor of total body fat ($\beta = -0.60$, $P = 0.01$).

Quintile analysis

Quintile groups (based on percent body fat) did not differ in intake of carbohydrate or fat (total or subtype, grams per day, or %EI) (data not shown).

MZ twin pair analyses (co-twin case-control analyses).

Intrapair analyses Among genetically identical twin pairs ($n = 90$), all concordant for smoking status and HRT use, and excluding underreporters, the mean intrapair difference in total body fat mass was 3.48 ± 3.1 kg and distributed normally (range 0.03–13.48 kg). The intrapair differences in central abdominal fat ranged from 0.003 to 1.20 kg (mean \pm SD 0.33 ± 0.28 kg) and were also normally distributed. Intrapair differences in energy intake, dietary fat (total or type, grams per day, or %EI), or protein did not explain the intrapair variance in total body or central abdominal fat (Table 2). Only a minor inverse relationship between intrapair differences in carbohydrate intake (%EI) and intrapair variance in total body fat (but not central abdominal fat) was found ($\beta = -0.011$, $r^2 = 0.06$, $P = 0.02$) (Table 2, Fig. 1).

Within each MZ twin pair, the twin with the higher dietary fat intake (grams

per day and %EI) had total body and central abdominal fat similar to the twin with the lower dietary fat intake (Table 3). Similar results were found with total carbohydrates (grams per day and %EI), but not with total sugars (grams per day), where the twin with the higher intake of total sugars had a lower total body (%) and central abdominal (%) fat (Table 3).

Concordant versus discordant pair comparisons. To search for any relationship with diet, MZ twin pairs were classified as concordant or discordant for total body and central abdominal fat based on several cut-off levels of difference within each pair. The intrapair differences in energy intake and diet composition were similar between pairs concordant or discordant for total body fat, despite testing across a wide range of discordance within identical twin pairs (data not shown). Only when examining large discordances in total body and central abdominal fat, differences in carbohydrate intake showed a negative relationship of borderline significance. When discordance for total body fat mass exceeded 7 kg within a pair ($n = 13$ pairs), the difference in carbohydrate intake in these pairs was greater than that in MZ pairs who were similar (concordant) in their total body fat ($n = 77$ pairs) (7.3 ± 1.8 vs. 4.5 ± 0.45 %EI, $P = 0.04$). Similarly, in MZ twin pairs who differed by >7 kg of total body fat (discordant), the difference in intake of total sugars within the pair was greater than that in concordant pairs (7.2 ± 1.3 vs. 4.7 ± 0.5 %EI, respectively), again of borderline significance ($P = 0.04$). Similar relationships were also found for pairs discordant for central abdominal fat only when the difference in central abdominal fat within pairs exceeded 500 g: the difference in intake was significantly greater in discordant MZ pairs ($n = 17$) compared with concordant pairs ($n = 73$) for both carbohydrate (7.3 ± 1.6 vs. 4.3 ± 0.4 %EI, respectively, $P = 0.01$) and total sugars (7.0 ± 1.1 vs. 4.6 ± 0.5 %EI, respectively, $P = 0.03$).

Conversely, MZ pairs were also classified as concordant or discordant for dietary intake based on several cut-off levels for differences within each pair for energy, total dietary fat, carbohydrate, and total sugars. Across a wide range of cut-off levels to define discordance in energy intake, dietary total fat, or carbohydrate (%EI), the differences in total body fat or central abdominal fat within pairs were similar in concordant and discordant pairs (Table 4). Similarly, the differences in body fat measures in pairs discordant for intake of fat subtypes did not

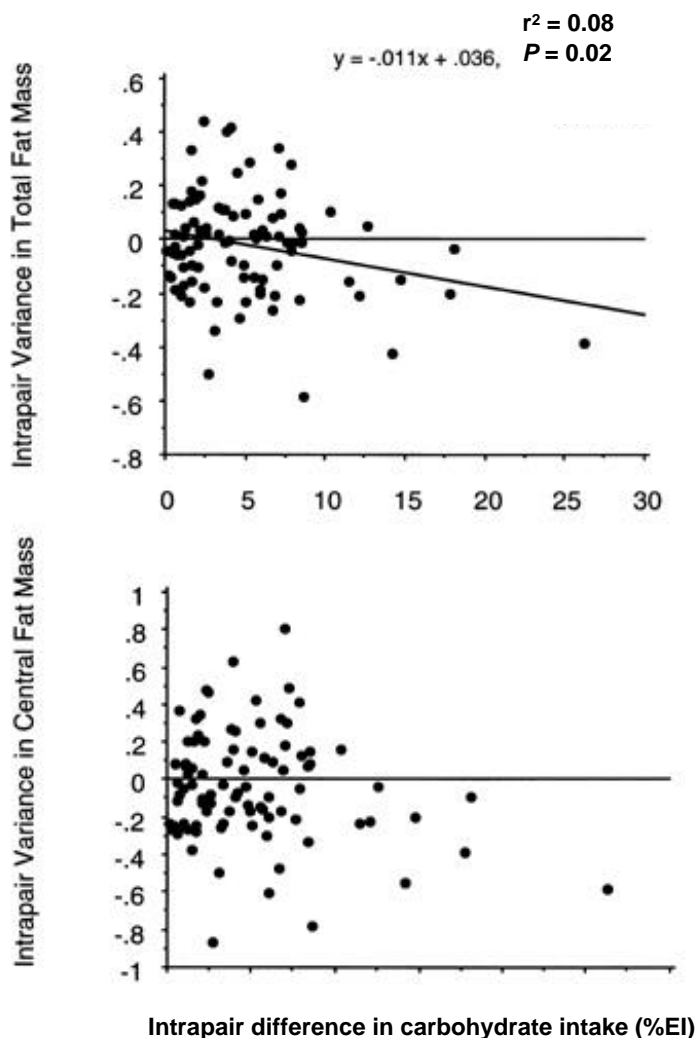


Figure 1—Intrapair differences in carbohydrate intake (%EI) predict intrapair variance in total fat mass but not in central abdominal fat mass in MZ middle-aged female twin pairs.

differ from that in concordant pairs (data not shown).

Discordance in intake of total sugars (exceeding 5% of daily energy intake) was associated with significantly greater differences in total body and central abdominal fat, compared with those differences in body fat in concordant pairs (i.e., with a similar intake of total sugars) (Table 4). This degree of difference in intake of total sugars accounted for a mean difference in total body and central abdominal fat mass of 1.5 kg and 110 g, respectively. MZ analyses were also performed controlling for physical activity level. Results were unchanged, although twin pair numbers in analyses were reduced (data not shown).

Gene-environment interaction analysis

In examining for a contributory gene-environment interaction in those predisposed to obesity (i.e., a greater dietary fat intake producing greater body fat mass in genetically predisposed subjects), we found that a higher intake of dietary fat was not associated with greater total body or central abdominal fat in subjects with a genetic predisposition to generalized obesity (Fig. 2). Similarly, higher intakes of carbohydrate (total and as simple sugars) in the genetically predisposed were not associated with significantly different total body and central abdominal fat mass. Results were similar whether dietary intakes were expressed as percent or grams per day (not shown). Similar for central abdominal fat, a greater intake of dietary fat or carbohydrate was not associated with any signifi-

Table 3—MZ twin analysis (co-twin case control study): higher intakes of dietary fat, carbohydrate, and sugars and total body and central abdominal fat in 90 middle-aged female twin pairs

Macronutrient	Within-pair comparison										
	Nutrient intake		Total body fat (kg)		Total body fat (%)		Central fat (kg)		Central fat (%)		
	Higher intake	Lower intake	Higher intake	Lower intake	Higher intake	Lower intake	Higher intake	Lower intake	Higher intake	Lower intake	
Fat											
Grams/day	88.2 ± 3.6	62.5 ± 2.3*	23.0 ± 0.7	23.5 ± 0.7	36.7 ± 0.7	37.0 ± 0.7	1.45 ± 0.06	1.47 ± 0.06	35.6 ± 1.0	35.8 ± 1.0	
%	34.4 ± 0.6	29.8 ± 0.6*	23.3 ± 0.7	23.3 ± 0.7	36.9 ± 0.7	36.9 ± 0.7	1.45 ± 0.06	1.47 ± 0.06	36.0 ± 1.0	35.4 ± 1.0	
Carbohydrate											
Grams/day	291.3 ± 9.7	217.9 ± 6.3*	23.3 ± 0.7	23.3 ± 0.7	36.9 ± 0.7	36.9 ± 0.7	1.47 ± 0.06	1.45 ± 0.06	35.6 ± 1.0	35.8 ± 1.0	
%	52.0 ± 0.6	47.1 ± 0.6*	23.8 ± 0.7	23.4 ± 0.7	36.7 ± 0.7	37.0 ± 0.7	1.46 ± 0.07	1.46 ± 0.06	35.1 ± 1.0	36.3 ± 0.9	
Total sugars											
Grams/day	159.4 ± 6.5	115.7 ± 3.9*	23.0 ± 0.7	23.6 ± 0.7	36.4 ± 0.8	37.3 ± 0.7†	1.45 ± 0.07	1.47 ± 0.06	35.0 ± 1.0	36.5 ± 0.09†	
%	29.1 ± 0.6	24.1 ± 0.5*	23.4 ± 0.7	23.2 ± 0.7	36.9 ± 0.7	36.8 ± 0.7	1.48 ± 0.07	1.44 ± 0.06	35.8 ± 1.0	35.7 ± 1.0	

Data are means ± SEM. Energy and macronutrient intake were measured by FFQ. Macronutrient intake is expressed as %EI. * $P < 0.0001$, † $P < 0.05$ for paired analysis (within twin pairs) of higher-intake versus lower-intake twin.

Table 4—Increasing discordance for dietary intake: its influence on intrapair differences in total and central fat mass in MZ female twins

Discordance in dietary factor	Number of twin pairs		Intrapair differences			
			Total fat (kg)		Central fat (kg)	
			Concordant	Discordant	Concordant	Discordant
Energy intake (kcal)						
>96	12	78	2.21 ± 0.38	3.61 ± 0.36	0.30 ± 0.08	0.33 ± 0.03
>240	30	60	3.30 ± 0.61	3.49 ± 0.38	0.38 ± 0.06	0.29 ± 0.03
>480	52	38	3.20 ± 0.42	3.73 ± 0.50	0.34 ± 0.04	0.30 ± 0.04
Dietary fat (%)						
>3	44	46	4.15 ± 0.53	2.74 ± 0.35	0.36 ± 0.05	0.29 ± 0.03
>5	60	30	3.58 ± 0.43	3.11 ± 0.44	0.34 ± 0.04	0.29 ± 0.04
>10	80	10	3.34 ± 0.34	4.15 ± 0.96	0.32 ± 0.03	0.39 ± 0.07
Carbohydrate (%)						
>3	37	54	3.21 ± 0.48	3.58 ± 0.44	0.30 ± 0.04	0.34 ± 0.04
>5	54	36	3.42 ± 0.40	3.42 ± 0.50	0.30 ± 0.04	0.32 ± 0.05
Total sugars (%)						
>3	32	58	2.83 ± 0.45	3.75 ± 0.42	0.28 ± 0.03	0.35 ± 0.03
>5	53	37	2.84 ± 0.35	4.27 ± 0.58*	0.28 ± 0.03	0.39 ± 0.05*

Data are n or means ± SEM. Energy and macronutrient intake were measured by FFQ. Discordance for each dietary factor was defined by the degree of difference for that factor within each MZ twin pair. *P < 0.05, concordant vs. discordant.

cant difference in central abdominal fat in those subjects with a genetic predisposition to central obesity (Fig. 2). Thus, no evidence of higher dietary intake contributing to greater body fat mass in subjects with susceptibility to obesity was found using this twin model.

CONCLUSIONS — Reports of a relationship between dietary fat and body fat and obesity have profoundly influenced medical practice and community attitudes, yet the apparent reduction in fat intake in several Western countries over recent decades has been accompanied by increases in the prevalence of obesity (3). Past studies have been flawed by their cross-sectional nature, methodological inadequacies, such as surrogate estimates of adiposity (5,6,9, 12,17), small sample size (9), inadequacy of dietary measures (5,9,10,17), selection bias (15,16), and inclusion of underreporters. Recent critical reviews emphasize the paucity of conclusive evidence of a relationship between diet and the regulation of body fat stores (13, 14). This study overcomes the limitations of prior studies by using a direct measure of body fatness (DEXA) and well-validated dietary measures. Bias was minimized twofold: data bias by excluding underreporters and selection bias by studying a cohort with a wide distribution of weight whose members were unaware of any hypothesis regarding body fatness. It also used the unique and robust twin model to examine the nutrient/body fat hypothesis.

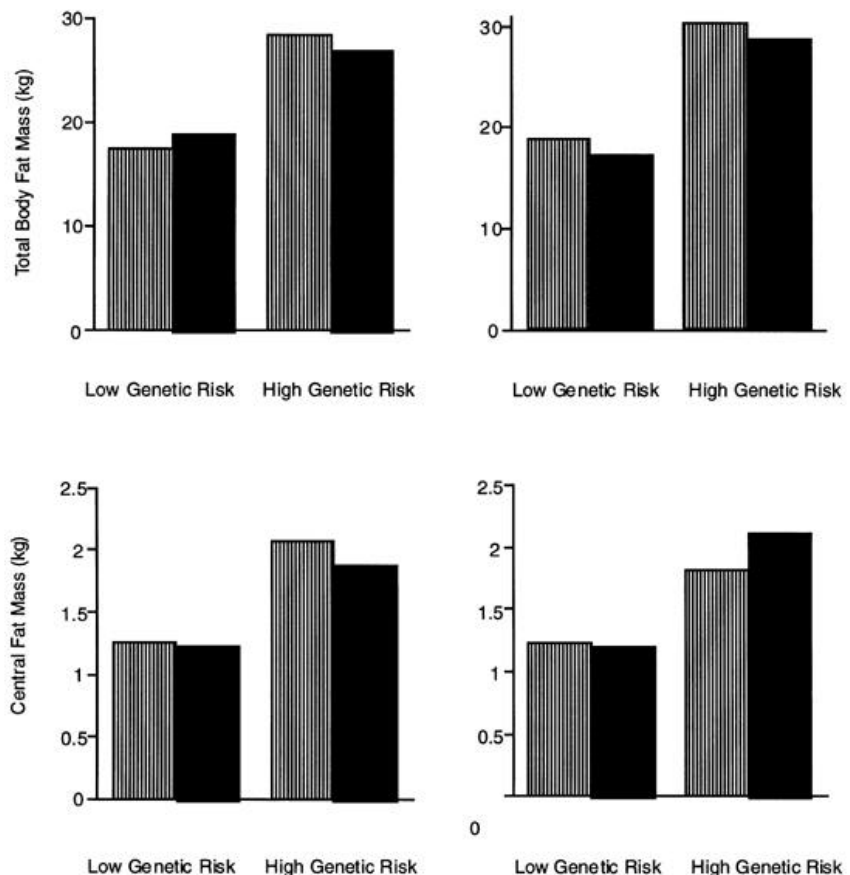


Figure 2—The influence of high (■) and low (▨) intake of dietary fat and carbohydrate (%EI) on total body and central abdominal fat mass in subjects with high and low genetic susceptibility to generalized and central abdominal obesity. High genetic susceptibility to generalized obesity: co-twin with total body fat mass 30.0–65 kg (mean ± SD BMI 27.3 ± 2.9 kg/m²). High genetic susceptibility to central abdominal obesity: co-twin with central abdominal fat mass 1.78–4.12 kg.

First, this study confirms some previous cross-sectional findings: that of a weak relationship between diet fat and BMI (non-significant after GEE modeling, however) and a small inverse relationship between alcohol and total adiposity. No relationship was found between recent amount or type of dietary fat intake and total body and central abdominal fat (both as grams per day and as %EI), confirming findings of some studies (12), particularly those using directly measured body fat (11,23). No relationship was found between body fatness and the fat-to-carbohydrate ratio, a ratio representing a high proportion of dietary fat-derived energy that has been implicated in obesity (3,18) but has not previously been examined against directly measured body fat in a population with a wide distribution of adiposity. In multivariate analysis including smoking, HRT, and physical activity, the lack of any relationship between dietary fat intake and body fatness remained.

The nonsignificant association between fat intake and weight and BMI deserves further discussion, since previous reports of significant relationships have perpetuated the paradigm that recent dietary fat intake is related to, and possibly responsible for, current body fat mass. In this study, dietary fat did not relate to body fat mass (measured accurately with DEXA), confirming the findings of other studies that measured body fat directly (11,23).

This study also dissects the complex relationships between genes, diet, and adiposity. In the co-twin case control (MZ) analyses, genetic factors and measured environmental factors were controlled for by studying genetically identical twins concordant for smoking and HRT but discordant for one of a variety of dietary factors. These analyses found that differences in total body and central abdominal fat mass within pairs were significantly greater only when the difference in daily intake of total sugars exceeded 5% of total energy intake, then accounting for 1.5 kg total body and 110 g central abdominal fat. No such effect was observed for any other dietary factor, and importantly, not for dietary fat. Similarly, when the dietary intake of MZ twin pairs discordant for total body and central abdominal fat were examined, only carbohydrate and total sugar intake (%EI) differed between pairs discordant and concordant for body fat measures. Cross-sectionally, this does not necessarily imply causation, since subjects with weight concerns may consciously reduce their sugar

intake. No relationship between total body and central abdominal fat and dietary fat (grams per day or %EI) was found using this powerful model.

A hypothesis exists that dietary fat only affects body fat in those predisposed to obesity (25). Based on previously described methods (26–28), we found no evidence in this cohort for such a gene-environment interaction between high dietary fat intake and higher contemporaneous total fat mass in the genetically predisposed. This may be due, in part, to the relatively low statistical power of the methodology used to detect gene-environment effects, so that diet-body fat gene-environment interactions may be detected in larger populations or in those with a specific phenotype.

This study has clarified some of the controversies existing in previous cross-sectional studies by using direct measures of body fat and validated dietary intake (measured with the most direct and accurate means applicable in large populations) and by excluding underreporters. Because the unique body composition of underreporters (21) may bias relationships between nutrients and body composition, their exclusion improves the accuracy of this study. Using the robust co-twin case control study design (MZ twin model), only differences in simple carbohydrate intake (%EI) predicted a very small proportion of the differences in body fatness. Overall, these cross-sectional results suggest that whatever the influence of dietary intake and composition on adiposity, it is small and relatively minor in comparison with the influence of genetic and other environmental factors. Due to the cross-sectional nature of the study, however, confounding may be present, and prospective studies in normal populations are necessary with similarly stringent validation of dietary data, assessment of body composition, and behavioral covariates.

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