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Dietary Intakes of Zinc and Heme Iron from Red Meat, but Not from Other Sources, Are Associated with Greater Risk of Metabolic Syndrome and Cardiovascular Disease[1–3]

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Abstract

Metabolic syndrome (MetS), Type 2 diabetes (T2D), and cardiovascular disease (CVD) share an inflammatory etiology and are known to be influenced by diet. We investigated associations of hypothesized prooxidative (Fe) and antioxidative (Zn, Mg, β-carotene, vitamin C, vitamin E) micronutrients with incident MetS, T2D, and CVD in the Multi-Ethnic Study of Atherosclerosis. Participants, 45–84 y at baseline (2000–2002), were followed through 2010. Diet was assessed by FFQ. After adjusting for demographics and behavioral confounders, including BMI, dietary vitamin E intake was inversely associated with incident MetS and CVD [HR for extreme quintiles: MetS = 0.78 (95% CI = 0.62, 0.97), *P*-trend = 0.01; CVD: HR = 0.69 (95% CI = 0.46, 1.03), *P*-trend = 0.04]. Intakes of heme iron and Zn from red meat, but not from other sources, were positively associated with risk of MetS [heme iron from red meat: HR = 1.25 (95% CI = 0.99,1.56), *P*-trend = 0.03; Zn from red meat: HR = 1.29 (95% CI = 1.03,1.61), *P*-trend = 0.04] and CVD [heme iron from red meat: HR = 1.65 (95% CI = 1.10,2.47), *P*-trend = 0.01; Zn from red meat: HR = 1.51 (95% CI = 1.02, 2.24), *P*-trend = 0.01]. Dietary intakes of nonheme iron, Mg, vitamin C, and β-carotene were not associated with risk of MetS, T2D, or CVD. Data provided little support for the associations between specific micronutrients and MetS, T2D, or CVD. However, nutrients consumed in red meat, or red meat as a whole, may increase risk of MetS and CVD. J. Nutr. 142: 526–533, 2012.

Introduction

Frequent intake of healthy foods may reduce the risk of MetS[10], T2D, and CVD (1–3). Intakes of fruits, vegetables, whole grain, and nuts have been associated with lower concentrations of markers of chronic inflammation and endothelial activation and with quantitative traits related to glucose metabolism (4–7). Micronutrients present in those foods, such as Zn and Mg, are essential components of several enzymes involved in inflammatory and metabolic pathways and may contribute to the reported associations between specific foods and disease (8–12). In addition, previous studies suggest that higher intakes of Zn, Mg, and other well-known antioxidants, such as β-carotene, vitamin C, and vitamin E, may attenuate oxidative stress (11,13–17), an etiology common among MetS, T2D, and CVD. In contrast to the hypothesized favorable effects of those nutrients, studies suggest greater heme iron intake and serum ferritin concentrations (reflective of body Fe stores and influenced by dietary Fe) are associated with the development of MetS, T2D, and CVD (8,18–20,20–22).

Despite these plausibly biological mechanisms, single nutrients have been inconsistently associated with disease outcomes. Some of this inconsistency may be due to differences in food sources of these nutrients across unique populations, particularly when predominant food sources of a single nutrient show opposite associations with disease. It is possible that some constituents of the food matrix interact with single nutrients,

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[3] Supplemental Tables 1–3 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.

[10] Abreviations used: BP, blood pressure; CVD, cardiovascular disease; MESA, Multi-Ethnic Study of Atherosclerosis; MetS, metabolic syndrome; T2D, type II diabetes.

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altering their bioavailability and influencing mechanisms that underlie disease development (23). Moreover, the assignment of disease associations to nutrients alone may be misleading, because intakes of single nutrients may serve as a marker of the entire matrix of constituents comprised by a given food.

The aims of this study were to characterize the associations of heme and nonheme iron, Zn, Mg, β-carotene, vitamin C, and vitamin E with incidence of MetS, T2D, and CVD in a multiethnic, population-based cohort. We hypothesized that intake of heme iron would be positively associated with MetS, T2D, and CVD incidence, whereas intakes of Zn, Mg, and vitamin antioxidants would be inversely associated with the same outcomes. Furthermore, because food source might influence the nature of nutrient associations, particularly for those nutrients found in both red meat [hypothesized to be unfavorably related to these outcomes (24,25)] and other sources, we also evaluated associations of heme iron and Zn from red meat and other sources.

Methods

Study population

The MESA is a prospective study initiated in July 2000 to investigate risk factors associated with subclinical CVD in a population-based sample (26). A total of 6814 adults, aged 45–84 y and free of clinical CVD, were recruited from six U.S. communities: Baltimore City and County, MD; Chicago, IL; Forsyth County, NC; New York, NY; Los Angeles County, CA; and St Paul, MN. The first examination was conducted between 2000 and 2002 (baseline), followed by examinations in 2002–2003, 2004–2005, and 2005–2007. Protocols were approved by local institutional review boards; all participants provided written informed consent.

We excluded participants whose dietary data did not meet quality control checks ($<$600 or $\geq$6000 kcal/d or unusual answering patterns, n = 801) and those with prevalent T2D (n = 859) or MetS at baseline (n = 2248) (depending on the primary outcome of interest). We also excluded participants who did not return for an exam after baseline (n = 303 and 219 for T2D and MetS analyses, respectively). For the CVD analysis, participants with prevalent T2D at baseline were excluded due to the potential for recent dietary change to confound the associations of interest.

Dietary assessment

A modified-block (27), 120-item FFQ was used at baseline to assess usual food intake over the previous year (28), with modifications to include Chinese food/beverage items (28,29). Mean correlation coefficients between log-transformed nutrients estimated from the original FFQ compared to mean across eight 24-h recalls were 0.40 for energy-adjusted vitamin C, 0.20 for energy-adjusted vitamin E, and 0.36 for total energy intake (28). Criterion validity of the modified MESA-specific FFQ was evaluated by comparison of macronutrient intake with plasma lipid concentrations within the MESA cohort (29).

To estimate daily nutrient intakes from diet, frequency and serving size for each food consumed were multiplied by the nutrient content of that food (Nutrition Data Systems for Research, University of Minnesota; Minneapolis). The content of heme iron was calculated as 40% of the total Fe intake from beef, poultry, and fish (30). Nonheme Fe was estimated as the difference between the intakes of total Fe and heme iron. Intakes of Zn from red meat and from all other sources were estimated separately by multiplying total Zn content in food items containing red meat by the daily food intake frequency and the age- and portion size-specific gram weights. Zn from all other sources was estimated by subtracting Zn from red meat only from the total Zn intake. Similarly, intakes of heme iron from red meat and other sources (poultry and fish) were estimated.

Participants were also asked about dietary supplement use, allowing quantification of intakes of nutrients from foods and supplements. However, in the present study, there were no substantial differences in results using total nutrient intake (from foods and supplements) compared to nutrients from food sources only. Therefore, only data from food-derived nutrients are presented.

Associations between disease outcomes and food groups were estimated for the major food sources of the nutrients of interest (**Supplemental Table 1**).

Assessment of incident disease

T2D. Fasting serum glucose was quantified by rate reflectance spectrophotometry using thin film adaptation of the glucose oxidase method on the Vitros analyzer (Johnson & Johnson Clinical Diagnostics). The laboratory analytical CV was 1.1%. Incident T2D cases were identified at follow-up examinations by self-reported diagnosis, serum glucose $\geq$ 6.99 mmol/L, or new use of hypoglycemic medication.

MetS. MetS was defined according to the AHA/National Heart, Lung, and Blood Institute criteria (any three of the following: waist circumference $\geq$ 102 cm in men or $\geq$88 cm in women; TG $\geq$1.7 mmol/L or treatment for elevated TG; HDL-C $<$1.03 mmol/L in men or $<$1.29 mmol/L in women; BP $\geq$130 mm Hg systolic BP, $\geq$85 mm Hg diastolic BP or drug treatment for hypertension; and fasting glucose $\geq$5.6 mmol/L or treatment for elevated glucose). Participants who did not meet at least three of the MetS criteria at baseline but did meet at least three criteria at any of the follow-up examinations were identified as incident MetS cases.

CVD. Total CVD events comprise occurrence of any of the following: myocardial infarction, resuscitated cardiac arrest, definite and probable angina, stroke, stroke death, coronary heart disease death, other atherosclerotic death, or other CVD death. Information on cardiovascular events was obtained through cohort clinic visits, telephone calls to participants, medical record abstractions, or obituaries. Self-reported diagnoses, death certificates, autopsy reports, and medical records for hospitalizations as well as selected cardiovascular diagnoses and procedures were reviewed by a medical endpoints committee. Deaths for subjects with loss to follow-up were identified by contacting next-of-kin and medical records were obtained on 98% of reported hospitalized events and 95% of outpatient procedures (31). A standard protocol was used to classify events and assign incident dates based on the records available (1,32).

Other covariates

A combination of interviewer-administered and self-completed questionnaires was used at the baseline exam to gather information on demographics, education, medication use, and smoking (status and pack-years) along with alcoholic beverage consumption (via FFQ). The MESA Typical Week Physical Activity Survey (33) adapted from the Cross-Cultural Activity Participation Study (34) assessed time spent in, and frequency of, various physical activities during a typical week in the past month. During examinations, participants provided samples for measurement of fasting glucose and lipids concentrations and had anthropometric measurements taken. HDL cholesterol was measured in EDTA plasma using the cholesterol oxidase cholesterol method (Roche Diagnostics) after precipitation of non-HDL cholesterol with Mg/dextran. LDL cholesterol was calculated in plasma specimens having a TG value $<$4.5 mmol/L using the formula of Friedewald et al. (35); the laboratory analytical CV was 1.6%. TG concentrations were measured in EDTA plasma using Triglyceride GB reagent (Roche Diagnostics); the analytical CV was 4.0%. Resting seated BP was measured three times using a Dinamap model Pro 100 automated oscillometric sphygmomanometer (Critikon). The mean of the last two measurements was used in the analysis.

Statistical analysis

Age- and sex-adjusted Spearman correlation coefficients were used to evaluate correlations between energy-adjusted nutrients. Cox proportional hazard regression models estimated HR and 95% CI for T2D, MetS, and CVD across quintiles of nutrient or food intake, using the lowest quintile as the referent (SAS 9.2; SAS Institute). Tests for linear trend were conducted by using median values of energy-adjusted nutrient quintiles as continuous variables in the multivariable-adjusted models.

Model 1 adjusted for sex, age (years), race/ethnicity, energy intake (kcal/d), field center, education level (less than high school, high school, more than high school), active leisure (walking, sport, conditioning in metabolic min/wk), inactive leisure (television viewing, reading, light sitting activities in metabolic min/wk), alcohol intake (ethanol g/d), smoking (never, former, or current smoker and pack-years of cigarette smoking), BMI (kg/m^2), dietary fiber (g/d), and dietary supplement use (at least weekly, yes/no). Model 2 further adjusted for the polyunsaturated: saturated fat ratio and other nutrients of interest: Mg, Zn, heme iron, nonheme iron, β-carotene, vitamin E, and vitamin C (mutual adjustment, i.e., nutrient of interest as main predictor plus all other nutrients listed as covariates).

Because previous studies observed differences in the relationships between nutrient intake and health outcomes according to sex, ethnicity, and alcohol consumption (20,36–38), stratified analyses evaluated whether the associations of interest were modified by these three factors (alcohol intake categorized as 0–9.99, 10.00–29.99, and ≥30.00 ethanol g/d). All P values were two-sided, with $P < 0.05$ indicating significance.

Results

Among the 5285 participants in the incident CVD analytic sample, 42.9% were non-Hispanic whites, 24.4% were African Americans, 20.6 were Hispanics, and 12.1% were Chinese Americans (**Table 1**). No major differences in demographic characteristics were identified among analytic samples for each prospective analysis. Age- and sex-adjusted correlation among energy-adjusted nutrients ranged from −0.17 to 0.61 (**Table 2**). Coefficients were significant at the 0.05 level. The strongest correlations were observed between adjusted intakes of Zn and Mg (r = 0.61) and between nonheme iron and intakes of each Mg and Zn (r = 0.50). All adjusted nutrient-nutrient correlations were positive, except correlations with vitamin E. Coefficients were similar for all analytic samples (MetS, T2D, and CVD).

MetS. Between 2000 and 2007, 46.7 new cases of MetS per 1000 person-years were identified among 3828 participants without MetS at baseline (mean follow-up time: 4.8 y). Higher intakes of vitamin E were associated with lower risk of MetS (P-trend = 0.01), whereas higher intakes of Zn were associated with greater relative risk of MetS (P-trend = 0.03) (model 1, **Table 3**). After further adjustment for potential nutritional confounders (model 2), these associations were slightly attenuated and no longer significant (P-trend = 0.06 for vitamin E and Zn) (model 2, Table 3). There were no associations between incident MetS and intakes of total heme- and nonheme iron, Mg, vitamin C, or β-carotene (Table 3).

To better understand the role of food source on the positive association observed between incident MetS and Zn intake, which was contrary to our a priori hypothesis, we estimated the associations of Zn intake from red meat separately from the associations of Zn intake from all other sources. Only Zn derived from red meat sources was associated with greater risk of incident MetS [HR for extreme quintiles of Zn from red meat sources = 1.29 (95% CI = 1.03, 1.61), P-trend = 0.04; HR for Zn from all other sources = 1.13 (95% CI = 0.90, 1.42), P-trend = 0.22)] (model 1, **Table 4**). The association remained consistent after further adjustment for the ratio of polyunsaturated fat: saturated fat, Mg, and total heme iron intake. In a similar analysis, we observed a positive association between heme iron from red meat, but not for heme iron from poultry and fish, and incident MetS [HR for extreme quintiles of heme iron from red meat sources = 1.25 (95% CI = 0.99, 1.56), P-trend = 0.03; HR for extreme quintiles of heme iron from poultry and fish = 1.03 (95% CI = 0.81, 1.31), P-trend = 0.97] (model 1, Table 4). HR for extreme quintiles were similar after further adjustment for intakes of total Zn, Mg, nonheme iron, and antioxidants; however, the association was slightly attenuated and no longer

TABLE 1 Baseline characteristics of MESA participants, sample included in follow-up of incident CVD (n = 5285)[1]

Demographic and lifestyle characteristics	
Sex, %	
Female	52.7
Male	47.3
Age, *y*	61.8 ± 10.3
Race/ethnicity, %	
White	42.9
African American	24.4
Hispanic	20.6
Asian	12.1
Education, %	
<High school	16.1
High school	17.6
>High school	66.3
Smoking status, %	
Never	50.6
Former	36.8
Current	12.6
Physical activity, *MET-min/wk*	
Inactive leisure	1690 ± 1130
Active leisure	2500 ± 3050
BMI, *kg/m^2*	27.9 ± 5.3
Alcohol intake, *g/d*	5.5 ± 12.3
Dietary intake	
Total energy,[2] *kcal/d*	1680 (794)
Fiber, *g/d*	16.8 (8.4)
Heme iron, *mg/d*	0.78 (0.63)
Nonheme iron, *mg/d*	11.2 (5.4)
Zn, *mg/d*	8.27 (4.38)
Mg, *mg/d*	254 (118)
Vitamin C, *mg/d*	100 (60.1)
Vitamin E, *mg/d*	10.4 (6.7)
β-Carotene, *mg/d*	3.26 ± 2.40

[1] Values are percent, mean ± SE, or median (SE). CVD, cardiovascular disease; MESA, Multi-Ethnic Study of Atherosclerosis; MET, metabolic equivalent.
[2] 1 kcal/d = 4.18 kJ/d.

TABLE 2 Spearman correlation coefficients for energy-adjusted nutrients in 5285 MESA participants[1,2]

	Heme iron	Nonheme iron	Mg	Zn	Vitamin C	Vitamin E	β-Carotene
Heme iron	1	0.09	0.02	0.40	0.07	−0.09	0.25
Nonheme Fe		1	0.50	0.50	0.35	−0.08	0.31
Mg			1	0.61	0.44	−0.15	0.39
Zn				1	0.20	−0.17	0.25
Vitamin C					1	0.11	0.55
Vitamin E						1	0.08

[1] Correlations adjusted for age, sex, and race/ethnicity. MESA, Multi-Ethnic Study of Atherosclerosis.
[2] All coefficients are significant at the 0.05 level.

TABLE 3 HR and 95% CI for MetS by increasing quintiles of nutrient intake estimates in 3828 MESA participants[1,2]

	Quintiles of energy-adjusted nutrient intakes					
	Q1	Q2	Q3	Q4	Q5	*P*-trend
n	765	766	765	766	766	
Heme iron, mg/d	≤0.45	0.46–0.63	0.64–0.80	0.81–1.08	≥1.09	
Model 1	REF	1.08 (0.86, 1.35)	1.00 (0.80, 1.26)	1.27 (1.02, 1.59)	1.14 (0.90, 1.43)	0.17
Model 2	REF	1.07 (0.85, 1.36)	1.02 (0.80, 1.30)	1.28 (1.01, 1.63)	1.06 (0.81, 1.40)	0.55
Nonheme iron, mg/d	≤8.5	8.6–9.9	10.0–11.3	11.4–13.4	≥13.5	
Model 1	1.00	0.99 (0.79, 1.23)	1.04 (0.82, 1.31)	1.21 (0.96, 1.53)	1.01 (0.78, 1.31)	0.67
Model 2	1.00	1.02 (0.81, 1.28)	1.06 (0.83, 1.36)	1.22 (0.94, 1.57)	0.95 (0.70, 1.29)	0.95
Zn, mg/d	≤6.7	6.8–7.6	7.7–8.4	8.5–9.7	≥9.8	
Model 1	REF	0.99 (0.79, 1.25)	1.20 (0.96, 1.50)	1.10 (0.88, 1.38)	1.26 (1.01, 1.57)	0.03
Model 2	REF	1.00 (0.78, 1.28)	1.20 (0.93, 1.55)	1.13 (0.85, 1.49)	1.33 (0.97, 1.82)	0.06
Mg, mg/d	≤206	207–237	238–263	264–299	≥300	
Model 1	REF	1.02 (0.81, 1.29)	1.20 (0.94, 1.52)	1.23 (0.95, 1.59)	1.09 (0.82, 1.43)	0.39
Model 2[5]	REF	1.00 (0.79, 1.27)	1.16 (0.90, 1.49)	1.18 (0.90, 1.55)	1.01 (0.73, 1.39)	0.78
Vitamin C, mg/d	≤57	58–80	81–103	104–135	≥136	
Model 1	REF	1.06 (0.85, 1.32)	1.03 (0.82, 1.29)	1.02 (0.81, 1.29)	1.15 (0.89, 1.49)	0.36
Model 2	REF	1.07 (0.85, 1.34)	1.04 (0.82, 1.32)	1.01 (0.79, 1.29)	1.18 (0.90, 1.54)	0.32
Vitamin E, mg/d	≤7.3	7.4–8.7	8.8–10.1	10.2–12.4	≥12.5	
Model 1	REF	0.97 (0.78, 1.20)	1.01 (0.81, 1.26)	0.85 (0.68, 1.06)	0.78 (0.62, 0.97)	0.01
Model 2	REF	1.00 (0.79, 1.26)	1.03 (0.81, 1.31)	0.84 (0.65, 1.09)	0.76 (0.56, 1.03)	0.06
β-Carotene, mg/d	≤1.61	1.62–2.34	2.35–3.29	3.30–4.74	≥4.75	
Model 1	REF	0.96 (0.78, 1.19)	0.93 (0.74, 1.17)	1.00 (0.79, 1.25)	0.96 (0.73, 1.26)	0.91
Model 2	REF	0.94 (0.75, 1.17)	0.91 (0.71, 1.16)	0.98 (0.77, 1.26)	0.94 (0.70, 1.27)	0.92

[1] Values are HR (95% CI) for risk of MetS according to quintiles of micronutrient intakes. MESA, Multi-Ethnic Study of Atherosclerosis; MetS, metabolic syndrome.
[2] Statistical model 1 was adjusted for energy intake (kcal/d), age (y), sex, race-ethnicity (non-Hispanic whites, African Americans, Hispanics, and Chinese Americans), education (<high school, high school, >high school), study center, alcohol intake (g/d), physical activity (active and inactive leisure in metabolic equivalents per min/wk), BMI (kg/m^2), fiber intake (g/d), cigarette smoking (never, current, or former smoker), and dietary supplement use (>1/wk, yes or no). Statistical model 2 was adjusted for all variables in model 1 and the ratio of polyunsaturated fat intake:saturated fat intake and mutual adjustment for Mg, Zn, heme iron, nonheme iron, and antioxidant intake.

significant after further adjustment for trans-fat intake [HR for extreme quintiles of heme iron from red meat sources = 1.21 (95% CI = 0.99, 1.61), *P*-trend = 0.09; data not tabulated].

T2D. Among the 4982 MESA participants, 16.7 new T2D cases/1000 person-years were identified during the follow-up time (mean follow up time = 4.8 y). We found no significant associations

TABLE 4 HR and 95% CI for MetS by increasing quintiles of heme iron and Zn intake estimates in 3828 MESA participants, stratified by food source[1,2]

	Quintiles of energy-adjusted nutrient intakes					
	Q1	Q2	Q3	Q4	Q5	*P*-trend
n	765	766	765	766	766	
Heme iron from red meat, mg/d	≤0.18	0.19–0.30	0.31–0.41	0.42–0.58	≥0.59	
Model 1	1.00	1.06 (0.85, 1.34)	1.25 (0.99, 1.57)	1.33 (1.06, 1.66)	1.25 (0.99, 1.56)	0.03
Model 2	1.00	1.07 (0.84, 1.36)	1.19 (0.93, 1.51)	1.29 (1.02, 1.63)	1.25 (0.98, 1.58)	0.03
Heme iron from poultry and fish, mg/d	≤0.16	0.17–0.26	0.27–0.37	0.38–0.54	≥0.55	
Model 1	1.00	1.17 (0.94, 1.47)	1.04 (0.82, 1.31)	1.16 (0.93, 1.46)	1.03 (0.81, 1.31)	0.97
Model 2	1.00	1.19 (0.94, 1.50)	1.08 (0.85, 1.38)	1.16 (0.91, 1.47)	1.00 (0.77, 1.30)	0.71
Zinc from red meat, mg/d	≤0.52	0.53–0.87	0.88–1.17	1.18–1.65	≥1.66	
Model 1	1.00	1.13 (0.90, 1.42)	1.41 (1.12, 1.78)	1.25 (0.99, 1.58)	1.29 (1.03, 1.61)	0.04
Model 2	1.00	1.19 (0.93, 1.53)	1.46 (1.12, 1.89)	1.25 (0.95, 1.64)	1.42 (1.07, 1.89)	0.04
Zinc from other sources,mg/d	≤5.55	5.56–6.41	6.42–7.19	7.20–8.41	≥8.42	
Model 1	1.00	1.04 (0.83, 1.30)	1.09 (0.87, 1.37)	1.21 (0.96, 1.51)	1.13 (0.90, 1.42)	0.22
Model 2	1.00	1.00 (0.79, 1.27)	1.06 (0.82, 1.37)	1.16 (0.89, 1.51)	1.13 (0.83, 1.54)	0.33

[1] Values are HR and 95% CI for risk of MetS according to quintiles of micronutrient intakes. MESA, Multi-Ethnic Study of Atherosclerosis; MetS, metabolic syndrome.
[2] Statistical model 1 was adjusted for energy intake (kcal/d), age (y), sex, race-ethnicity (non-Hispanic whites, African Americans, Hispanics, and Chinese Americans), education (<high school, high school, >high school), study center, alcohol intake (g/d), physical activity (active and inactive leisure in metabolic equivalents per min/wk), BMI (kg/m^2), fiber intake (g/d), cigarette smoking (never, current, or former smoker), and dietary supplement use (>1/wk, yes or no). Statistical model 2 was adjusted for all variables in model 1 and the ratio of polyunsaturated fat intake:saturated fat intake, Mg, nonheme iron, antioxidant intake, and mutual adjustment for total Zn and total heme iron intake.

between intakes of the studied micronutrients and risk of incident T2D (**Supplemental Tables 2** and **3**).

CVD. Among the 5285 participants included in this analysis, the incidence rate of CVD events between 2000 and 2007 was 8.5 cases/1000 person-years (mean follow-up time = 6.2 y). There was a significant positive trend across quintiles of dietary intakes of heme iron and risk of incident CVD (*P*-trend = 0.02) (model 1, **Table 5**), whereas the trend was inverse across quintiles of dietary vitamin E intakes and risk of CVD (*P*-trend = 0.04) (model 1, Table 5). Results were similar after further adjustment for nutritional covariates (model 2, Table 5).

To further investigate the positive association with heme iron, associations were evaluated for different strata of heme iron intake. Heme iron from red meat, but not heme iron from poultry and fish, was associated with incident CVD [HR for extreme quintiles of heme iron from red meat sources = 1.65 (95% CI = 1.10, 2.47), *P*-trend = 0.001; HR for extreme quintiles of heme iron from all other sources = 1.03 (95% CI = 0.67, 1.57), *P*-trend = 0.34] (model 1, **Table 6**). The association remained significant after further adjustment for other nutrients (model 2). In a similar analysis, Zn from red meat, but not Zn from other sources, was associated with risk of CVD [HR for extreme quintiles of Zn from red meat sources = 1.51 (95% CI = 1.02–2.24), *P*-trend = 0.01; HR for extreme quintiles of Zn from all other sources = 0.81 (95% CI = 0.53, 1.23), *P*-trend = 0.29] (model 2, Table 6). However, the association with Zn from red meat was attenuated after adjustment for total heme iron intake [HR for extreme quintiles = 1.36 (95% CI = (0.85, 2.17), *P*-trend = 0.10] (data not tabulated).

No associations with risk of CVD were observed for dietary intakes of nonheme iron, Mg, Zn, vitamin C, and β-carotene.

Food groups. We evaluated associations between the three outcomes of interest and the major food sources of the studied nutrients in the MESA cohort. Red meat intake was positively associated with MetS [HR for extreme quintiles = 1.29 (95% CI = 1.01, 1.64), *P*-trend = 0.04)] and with CVD [HR for extreme quintiles = 1.59 (95% CI = 1.03, 2.47), *P*-trend = 0.03]. On the other hand, we observed inverse associations between intakes of other vegetables and risk of MetS [HR for extreme quintiles = 0.77 (95% CI = 0.60, 0.99), *P*-trend = 0.01], and between intakes of whole grain and risk of CVD [HR for extreme quintiles = 0.57 (95% CI = 0.37, 0.87), *P*-trend = 0.02]. Fish, poultry, white bread, salty snacks, fats and oils, and legumes were not associated with risk of MetS, T2D, and CVD (data not tabulated).

Interactions. The relationships between micronutrient intake and incident MetS, T2D, and CVD were not modified by sex, race-ethnicity, or alcohol intake (*P*-interaction $>$ 0.05 for all tests) (data not shown).

Discussion

Over 6 y of follow-up in this multiethnic cohort, dietary vitamin E was inversely associated with risk of MetS and CVD but not with T2D. Intake of heme iron and Zn, and more specifically, heme iron and Zn from red meat but not from other sources, was

TABLE 5 HR and 95% CI for total CVD by increasing quintiles of nutrient intake estimates in 5285 MESA participants[1,2]

	Quintiles of energy-adjusted nutrient intakes					
	Q1	Q2	Q3	Q4	Q5	*P*-trend
n	1057	1057	1057	1058	1056	
Heme iron, mg/d	≤0.44	0.45–0.63	0.64–0.80	0.81–1.06	≥1.07	
Model 1	1.00	0.89 (0.60, 1.30)	1.04 (0.71, 1.53)	1.26 (0.86, 1.86)	1.45 (0.96, 2.18)	0.02
Model 2	1.00	0.87 (0.58, 1.31)	1.13 (0.75, 1.69)	1.31 (0.86, 1.98)	1.57 (0.99, 2.49)	0.02
Nonheme iron, mg/d	≤8.5	8. 6–9.9	10.0–11.3	11.4–13.3	≥13.4	
Model 1	1.00	1.24 (0.82, 1.87)	1.52 (1.01, 2.27)	1.18 (0.77, 1.83)	0.88 (0.55, 1.42)	0.26
Model 2	1.00	1.28 (0.84, 1.96)	1.59 (1.04, 2.42)	1.13 (0.71, 1.79)	0.89 (0.52, 1.52)	0.31
Zinc, mg/d	≤6.7	6.8–7.6	7.7–8.4	8.5–9.7	≥9.8	
Model 1	1.00	1.15 (0.77, 1.70)	1.17 (0.79, 1.75)	1.45 (0.98, 2.14)	1.12 (0.74, 1.69)	0.46
Model 2	1.00	1.15 (0.76, 1.76)	1.22 (0.79, 1.90)	1.43 (0.90, 2.27)	1.12 (0.66, 1.90)	0.66
Mg,mg/d	≤206	207–237	238–263	264–299	≥300	
Model 1	1.00	1.34 (0.87, 2.06)	1.68 (1.09, 2.60)	1.78 (1.13, 2.79)	1.15 (0.68, 1.96)	0.83
Model 2[5]	1.00	1.35 (0.87, 2.09)	1.73 (1.10, 2.72)	1.84 (1.14, 2.99)	1.27 (0.70, 2.28)	0.66
Vitamin C, mg/d	≤57	58–80	81–103	104–135	≥136	
Model 1	1.00	1.19 (0.80, 1.77)	1.29 (0.86, 1.94)	1.42 (0.94, 2.17)	1.04 (0.65, 1.67)	0.92
Model 2	1.00	1.17 (0.78, 1.77)	1.27 (0.83, 1.94)	1.34 (0.86, 2.07)	1.01 (0.62, 1.65)	0.94
Vitamin E, mg/d	≤7.3	7.4–8.7	8.8–10.0	10.1–12.3	≥12.4	
Model 1	1.00	0.82 (0.55, 1.21)	1.06 (0.73, 1.55)	0.70 (0.47, 1.06)	0.69 (0.46, 1.03)	0.04
Model 2	1.00	0.75 (0.50, 1.13)	0.93 (0.62, 1.39)	0.56 (0.35, 0.89)	0.50 (0.29, 0.86)	0.01
β-Carotene, mg/d	≤1.61	1.62–2.34	2.35–3.29	3.30–4.74	≥4.75	
Model 1	1.00	1.20 (0.82, 1.75)	0.99 (0.66, 1.47)	0.92 (0.60, 1.41)	1.06 (0.66, 1.70)	0.83
Model 2	1.00	1.23 (0.83, 1.83)	1.06 (0.70, 1.60)	0.80 (0.50, 1.26)	1.08 (0.66, 1.77)	0.70

[1] Values are HR and 95% CI for risk of CVD according to quintiles of micronutrient intakes. CVD, cardiovascular disease; MESA, Multi-Ethnic Study of Atherosclerosis.

[2] Statistical model 1 was adjusted for energy intake (kcal/d), age (y), sex, race-ethnicity (non-Hispanic whites, African Americans, Hispanics, and Chinese Americans), education (<high school, high school, >high school), study center, alcohol intake (g/d), physical activity (active and inactive leisure in metabolic equivalents per min/wk), BMI (kg/m^2), fiber intake (g/d), cigarette smoking (never, current, or former smoker), and dietary supplement use (>1/wk, yes or no). Statistical model 2 was adjusted for all variables in model 1 and the ratio of polyunsaturated fat intake:saturated fat intake, Mg, nonheme iron, antioxidant intake, and mutual adjustment for total Zn and total heme iron intake.

TABLE 6 HR and 95% CI for total CVD by increasing quintiles of heme iron and Zn intake estimates in 5285 MESA participants, stratified by food source[1,2]

	Quintiles of energy-adjusted nutrient intakes					
	Q1	Q2	Q3	Q4	Q5	*P*-trend
n	1057	1057	1057	1058	1056	
Heme iron from red meat, mg/d	≤0.18	0.19–0.30	0.31–0.41	0.42–0.58	≥0.59	
Model 1	1.00	1.06 (0.71, 1.59)	1.25 (0.83, 1.87)	1.22 (0.81, 1.84)	1.65 (1.10, 2.47)	0.01
Model 2	1.00	1.14 (0.75, 1.75)	1.38 (0.90, 2.13)	1.28 (0.82, 2.01)	1.85 (1.18, 2.88)	0.01
Heme iron from poultry and fish, mg/d	≤0.15	0.16–0.26	0.27–0.36	0.37–0.52	≥0.53	
Model 1	1.00	0.77 (0.52, 1.14)	0.84 (0.57, 1.24)	1.22 (0.83, 1.78)	1.03 (0.67, 1.57)	0.34
Model 2	1.00	0.98 (0.69, 1.38)	1.10 (0.78, 1.55)	1.02 (0.72, 1.45)	1.25 (0.88, 1.77)	0.17
Zinc from red meat, mg/d	≤0.54	0.55–0.89	0.90–1.19	1.20–1.68	≥1.69	
Model 1	1.00	1.01 (0.67, 1.53)	1.10 (0.72, 1.68)	1.37 (0.91, 2.06)	1.51 (1.02, 2.24)	0.01
Model 2	1.00	1.04 (0.68, 1.61)	1.17 (0.76, 1.81)	1.40 (0.91, 2.15)	1.66 (1.10, 2.49)	<0.01
Zinc from other sources,mg/d	≤5.53	5.54–6.41	6.42–7.19	7.20–8.41	≥8.42	
Model 1	1.00	1.02 (0.70, 1.49)	0.91 (0.61, 1.35)	0.99 (0.67, 1.47)	0.81 (0.53, 1.23)	0.29
Model 2	1.00	0.98 (0.65, 1.46)	0.88 (0.56, 1.38)	0.89 (0.54, 1.44)	0.68 (0.35, 1.30)	0.24

[1] Values are HR and 95% CI for risk of CVD according to quintiles of micronutrient intakes. CVD, cardiovascular disease; MESA, Multi-Ethnic Study of Atherosclerosis.

[2] Statistical model 1 was adjusted for energy intake (kcal/d), age (y), sex, race-ethnicity (non-Hispanic whites, African Americans, Hispanics, and Chinese Americans), education (<high school, high school, >high school), study center, alcohol intake (g/d), physical activity (active and inactive leisure in metabolic equivalents per min/wk), BMI (kg/m^2), fiber intake (g/d), cigarette smoking (never, current, or former smoker), and dietary supplement use (>1/wk, yes or no). Statistical model 2 was adjusted for all variables in model 1 and the ratio of polyunsaturated fat intake:saturated fat intake, Mg, nonheme iron, antioxidant intake, and mutual adjustment for total Zn and total heme iron intake.

associated with greater risk of CVD and MetS. Dietary intakes of nonheme iron, Mg, vitamin C, and β-carotene were not associated with risk of MetS, T2D, or CVD.

The inverse association between dietary intake of vitamin E and incident MetS and CVD is in agreement with our a priori hypothesis. These associations are also consistent with a cross-sectional analysis of data from the Third NHANES that reported significantly lower blood concentrations of vitamin E in participants with MetS compared to participants without MetS (39). However, unlike our study, the NHANES III study also reported cross-sectional associations between MetS prevalence and serum concentrations of vitamin C and β-carotene. The difference in characterization of these exposures (intake vs. serum levels) and study designs (cross-sectional vs. prospective) may partly explain the discrepancy between our findings. Of the few other prospective studies evaluating the relationship between diet and risk of MetS, most have focused on foods and not nutrients (40,41). Higher intakes of vegetables, the main source of vitamin E in the MESA population, were associated with a 30% decrease in MetS prevalence among women living in Tehran (41). Higher intakes of vegetables and nuts, two major sources of vitamin E in the MESA population, were also associated with a lower risk of isquemic stroke (RR and 95% CI for 1 serving/d increment in green leafy vegetables intake: RR = 0.79 [95% CI = 0.62–0.99]) (42) and myocardial infaction (RR for >5 times/wk vs. <1 time/wk: RR = 0.49) (43). On the other hand, most studies investigating associations between dietary vitamin E and CVD have reported weak inverse relationships, not always reaching significance (44). A pooled analysis including results from nine cohort studies reported an inverse association of borderline significance with coronary heart disease risk [RR and 95% CI extreme quintiles: RR = 0.84 (95% CI = 0.71, 1.00), *P*-trend = 0.17] (45), which is consistent with our results. It is important to note, however, that high-dose vitamin E supplementation has been associated with a small increase in all-cause mortality in clinical trials (46).

In MESA, red meat was the main source of heme iron and Zn, accounting for 47 and 14% of those intakes, respectively. The positive association observed between heme iron from red meat sources and CVD corroborate previous results from prospective studies evaluating the relationships of CVD with heme iron (21,47,48) and red meat (24,49–51). Red meat was also positively associated with MetS and CVD in our analysis. Similarly, data showed that Zn from red meat, but not from other sources, was associated with risk of CVD and MetS. This association is supported by findings from the Supplementation en Vitamines et Mineraux Antioxydants Study, showing a positive association between serum Zn concentration and MetS (52). However, because intakes of heme iron and Zn from non-red meat sources had no effect on the outcomes of interest, our findings suggest that the associations observed might reflect other constituents of red meat (53) or that such components might interact with heme iron and Zn, altering their bioavailability and producing the observed associations with incident disease. Also, because the correlation between heme iron from red meat and Zn from red meat is 0.90, it is possible that the positive association with Zn might reflect heme iron intake and its biological activity. Other factors associated with a meat-based dietary pattern might also drive these associations, as has been previously suggested by Supplementation en Vitamines et Mineraux Antioxydants Study investigators (52). We did observe attenuation on the measures of association after further adjustment for trans-fat intake; however, the interpretation of this observation is not clear given that trans-fat intake is difficult to estimate due to variation across brands and food preparations that are not well captured by FFQ. The absence of association with nutrients from other sources cannot be attributed to the lack of statistical power, because intakes of Zn and heme iron from these sources were either similar or higher than those from red meat alone. Altogether, observations from our study suggest that other elements present in red meat may influence risk of CVD and MetS.

The majority of tested relationships between the selected nutrients and the outcomes of interest were not significant, which confirms reports from other investigations, especially experimental trials. A meta-analysis including 15 clinical trials showed that intake of β-carotene (ranging from 15 to 50 mg/d) had no beneficial effects on CVD outcomes in various groups of men and women (54). A recent review on the role of minerals and antioxidants (Mg, vitamin C, vitamin E, and β-carotene) in T2D prevention concluded that the current scientific evidence does not support the beneficial role of individual nutrients for prevention (55). Results from prospective studies have been inconclusive. The Rotterdam Study showed no association between dietary intakes of vitamin C and myocardial infarction in healthy participants living in The Netherlands [RR and 95% CI for extreme tertiles = 1.05 (0.65–1.67)]; however, they reported lower risk of myocardial infarction among participants with a higher intake of β-carotene [RR and 95% CI for extremes tertiles = 0.55 (0.34, 0.83), *P*-trend = 0.01]. Results from the Iowa's Women Health Study showed no association between intakes of Zn, heme and nonheme iron and cardiovascular mortality in those consuming <10 g/d of alcohol (20), which was also true for over 90% of women in our cohort (data not shown). On the other hand, the same study showed significant associations between intakes of heme and nonheme iron and incident T2D (19), which contrasts with our observations. Similarly, our results oppose those of a meta-analysis including seven cohort studies that reported an overall 15% reduction in T2D risk associated with a 100-mg increase in daily Mg intake (95% CI = 0.79, 0.92) (56). Taken together, these observations do not support the individual role of nutrients on the studied outcomes but suggest that higher intakes of food sources containing nutrients such as vitamin E could be beneficial to individuals at risk for CVD and MetS. However, we cannot discard the potential for type II error due to random measurement error associated with the dietary assessment method.

These apparent inconsistencies underscore the challenges of the single nutrient approach to nutritional epidemiology. The study of nutritional factors in relation to chronic diseases is complex. First, nutrients are not consumed singly but as part of a food matrix that interacts with its components (23). Second, high correlations among nutrients make it difficult to disentangle associations attributed to a single nutrient from associations attributable to that nutrient's food source(s) or companion nutrients. Finally, in most cases, the magnitude of associations between dietary factors and chronic diseases are moderate, and the presence of measurement error may attenuate effect measures and reduce the statistical power to detect such associations (57,58). Although intakes of individual nutrients showed only weak or no associations with outcomes, other work in this cohort does suggest that more holistic expressions of dietary intake, namely dietary patterns, are associated with incident T2D and CVD (1,2).

Our study has important strengths. The comparison of associations with heme iron and Zn from red meat compared to other sources provided insights into the role these and other components of red meat may play in cardiometabolic diseases. The ethnic diversity of MESA allows for the study of a greater range of dietary behaviors and susceptibility to the health outcomes than other cohorts of more ethnically homogeneous composition. The prospective design allowed us to estimate incidence of disease with less concern for reverse causality between diet and outcomes. Finally the evaluation of nutrient intake from different food sources provided new insights into the relationship between nutrients and disease. Our study also has limitations, including potential measurement error in exposure assessment. Although likely random with respect to outcome, this could have attenuated measures of association. Moreover, residual confounding is possible due to covariate measurement error, imperfect proxies for underlying constructs, or yet-to-be identified, unmeasured confounding factors.

This prospective analysis showed modest inverse associations of dietary vitamin E with incident MetS and CVD and positive associations of dietary Zn and heme iron, only from red meat sources, with risk of MetS and CVD. These observations do not provide support for the association of other studied, food-supplied nutrients with MetS, CVD, or, particularly, T2D. However, nutrients consumed in red meat, or consumption of red meat itself, may be associated with greater risk of MetS and CVD.

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Literature Cited

1. Nettleton JA, Polak JF, Tracy R, Burke GL, Jacobs DR Jr. Dietary patterns and incident cardiovascular disease in the Multi-Ethnic Study of Atherosclerosis (MESA). Am J Clin Nutr. 2009;90:647–54.
2. Nettleton JA, Steffen LM, Ni H, Liu K, Jacobs DR Jr. Dietary patterns and risk of incident type 2 diabetes in the Multi-Ethnic Study of Atherosclerosis (MESA). Diabetes Care. 2008;31:1777–82.
3. Kastorini CM, Milionis HJ, Esposito K, Giugliano D, Goudevenos JA, Panagiotakos DB. The effect of Mediterranean diet on metabolic syndrome and its components: a meta-analysis of 50 studies and 534,906 individuals. J Am Coll Cardiol. 2011;57:1299–313.
4. Jiang R, Jacobs DR Jr, Mayer-Davis E, Szklo M, Herrington D, Jenny NS, Kronmal R, Barr RG. Nut and seed consumption and inflammatory markers in the multi-ethnic study of atherosclerosis. Am J Epidemiol. 2006;163:222–31.
5. Nettleton JA, Steffen LM, Mayer-Davis EJ, Jenny NS, Jiang R, Herrington DM, Jacobs DR Jr. Dietary patterns are associated with biochemical markers of inflammation and endothelial activation in the Multi-Ethnic Study of Atherosclerosis (MESA). Am J Clin Nutr. 2006;83:1369–79.
6. Basu A, Rhone M, Lyons TJ. Berries: emerging impact on cardiovascular health. Nutr Rev. 2010;68:168–77.
7. Sargeant LA, Khaw KT, Bingham S, Day NE, Luben RN, Oakes S, Welch A, Wareham NJ. Fruit and vegetable intake and population glycosylated haemoglobin levels: the EPIC-Norfolk study. Eur J Clin Nutr. 2001;55:342–8.
8. Ahluwalia N, Genoux A, Ferrieres J, Perret B, Carayol M, Drouet L, Ruidavets JB. Iron status is associated with carotid atherosclerotic plaques in middle-aged adults. J Nutr. 2010;140:812–6.
9. Barbagallo M, Dominguez LJ. Magnesium metabolism in type 2 diabetes mellitus, metabolic syndrome and insulin resistance. Arch Biochem Biophys. 2007;458:40–7.
10. Chambers EC, Heshka S, Gallagher D, Wang J, Pi-Sunyer FX, Pierson RN Jr. Serum magnesium and type-2 diabetes in African Americans and Hispanics: a New York cohort. J Am Coll Nutr. 2006;25:509–13.
11. Evans P, Halliwell B. Micronutrients: oxidant/antioxidant status. Br J Nutr. 2001;85 Suppl 2:S67.
12. De Oliveira Otto MC, Alonso A, Lee DH, Delclos GL, Jenny NS, Jiang R, Lima JA, Symanski E, Jacobs DR Jr, Nettleton JA. Dietary micronutrient intakes are associated with markers of inflammation, but not with markers of subclinical atherosclerosis. J Nutr. 2011; 141:1508–15.
13. Guerrero-Romero F, Rodriguez-Moran M. Hypomagnesemia, oxidative stress, inflammation, and metabolic syndrome. Diabetes Metab Res Rev. 2006;22:471–6.

14. Zago MP, Oteiza PI. The antioxidant properties of zinc: interactions with Fe and antioxidants. Free Radic Biol Med. 2001;31:266–74.
15. Montonen J, Knekt P, Jarvinen R, Reunanen A. Dietary antioxidant intake and risk of type 2 diabetes. Diabetes Care. 2004;27:362–6.
16. Asplund K. Antioxidant vitamins in the prevention of cardiovascular disease: a systematic review. J Intern Med. 2002;251:372–92.
17. Ellingsen I, Seljeflot I, Arnesen H, Tonstad S. Vitamin C consumption is associated with less progression in carotid intima media thickness in elderly men: a 3-year intervention study. Nutr Metab Cardiovasc Dis. 2009;19:8–14.
18. Jehn M, Clark JM, Guallar E. Serum ferritin and risk of the metabolic syndrome in U.S adults. Diabetes Care. 2004;27:2422–8.
19. Lee DH, Folsom AR, Jacobs DR Jr. Dietary Fe intake and type 2 diabetes incidence in postmenopausal women: The Iowa Women's Health Study. Diabetologia. 2004;47:185–94.
20. Lee DH, Folsom AR, Jacobs DR Jr. Iron, zinc, and alcohol consumption and mortality from cardiovascular diseases: The Iowa Women's Health Study. Am J Clin Nutr. 2005;81:787–91.
21. van der A DL, Peeters PH, Grobbee DE, Marx JJ, van der Schouw YT. Dietary haem Fe and coronary heart disease in women. Eur Heart J. 2005;26:257–62.
22. Jiang R, Ma J, Ascherio A, Stampfer MJ, Willett WC, Hu FB. Dietary Fe intake and blood donations in relation to risk of type 2 diabetes in men: a prospective cohort study. Am J Clin Nutr. 2004;79:70–5.
23. Jacobs DR Jr, Tapsell LC. Food, not nutrients, is the fundamental unit in nutrition. Nutr Rev. 2007;65:439–50.
24. Sinha R, Cross AJ, Graubard BI, Leitzmann MF, Schatzkin A. Meat intake and mortality: a prospective study of over half a million people. Arch Intern Med. 2009;169:562–71.
25. Aune D, Ursin G, Veierod MB. Meat consumption and the risk of type 2 diabetes: a systematic review and meta-analysis of cohort studies. Diabetologia. 2009;52:2277–87.
26. Bild DE, Bluemke DA, Burke GL, Detrano R, Diez Roux AV, Folsom AR, Greenland P, Jacob DR Jr, Kronmal R, Liu K, et al. Multi-ethnic study of atherosclerosis: objectives and design. Am J Epidemiol. 2002;156:871–81.
27. Block G, Hartman AM, Dresser CM, Carroll MD, Gannon J, Gardner L. A data-based approach to diet questionnaire design and testing. Am J Epidemiol. 1986;124:453–69.
28. Mayer-Davis EJ, Vitolins MZ, Carmichael SL, Hemphill S, Tsaroucha G, Rushing J, Levin S. Validity and reproducibility of a food frequency interview in a multi-cultural epidemiology study. Ann Epidemiol. 1999;9:314–24.
29. Nettleton JA, Rock CL, Wang Y, Jenny NS, Jacobs DR Jr. Associations between dietary macronutrient intake and plasma lipids demonstrate criterion performance of the Multi-Ethnic Study of Atherosclerosis (MESA) food frequency questionnaire. Br J Nutr. 2009;102:1220–7.
30. Monsen ER, Hallberg L, Layrisse M, Hegsted DM, Cook JD, Mertz W, Finch CA. Estimation of available dietary Fe. Am J Clin Nutr. 1978;31:134–41.
31. Folsom AR, Kronmal RA, Detrano RC, O'Leary DH, Bild DE, Bluemke DA, Budoff MJ, Liu K, Shea S, Szklo M, et al. Coronary artery calcification compared with carotid intima-media thickness in the prediction of cardiovascular disease incidence: the Multi-Ethnic Study of Atherosclerosis (MESA). Arch Intern Med. 2008;168:1333–9.
32. Detrano R, Guerci AD, Carr JJ, Bild DE, Burke G, Folsom AR, Liu K, Shea S, Szklo M, Bluemke DA, et al. Coronary calcium as a predictor of coronary events in four racial or ethnic groups. N Engl J Med. 2008;358:1336–45.
33. Bertoni AG, Whitt-Glover MC, Chung H, Le KY, Barr RG, Mahesh M, Jenny NS, Burke GL, Jacobs DR. The association between physical activity and subclinical atherosclerosis: the Multi-Ethnic Study of Atherosclerosis. Am J Epidemiol. 2009;169:444–54.
34. Ainsworth BE, Irwin ML, Addy CL, Whitt MC, Stolarczyk LM. Moderate physical activity patterns of minority women: the Cross-Cultural Activity Participation Study. J Womens Health Gend Based Med. 1999;8:805–13.
35. Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem. 1972;18:499–502.
36. Sullivan JL. Iron and the sex difference in heart disease risk. Lancet. 1981;1:1293–4.
37. Bhattacharya SK, Ahokas RA, Carbone LD, Newman KP, Gerling IC, Sun Y, Weber KT. Macro- and micronutrients in African-Americans with heart failure. Heart Fail Rev. 2006;11:45–55.
38. Lindquist CH, Gower BA, Goran MI. Role of dietary factors in ethnic differences in early risk of cardiovascular disease and type 2 diabetes. Am J Clin Nutr. 2000;71:725–32.
39. Ford ES, Mokdad AH, Giles WH, Brown DW. The metabolic syndrome and antioxidant concentrations: findings from the Third National Health and Nutrition Examination Survey. Diabetes. 2003; 52:2346–52.
40. Lutsey PL, Steffen LM, Stevens J. Dietary intake and the development of the metabolic syndrome: the Atherosclerosis Risk in Communities Study. Circulation. 2008;117:754–61.
41. Esmaillzadeh A, Kimiagar M, Mehrabi Y, Azadbakht L, Hu FB, Willett WC. Fruit and vegetable intakes, C-reactive protein, and the metabolic syndrome. Am J Clin Nutr. 2006;84:1489–97.
42. Joshipura KJ, Ascherio A, Manson JE, Stampfer MJ, Rimm EB, Speizer FE, Hennekens CH, Spiegelman D, Willett WC. Fruit and vegetable intake in relation to risk of ischemic stroke. JAMA. 1999; 282:1233–9.
43. Sabaté J. Nut consumption, vegetarian diets, ischemic heart disease risk, and all-cause mortality: evidence from epidemiologic studies. Am J Clin Nutr. 1999;70 Suppl 3:S500–3.
44. Willcox BJ, Curb JD, Rodriguez BL. Antioxidants in cardiovascular health and disease: key lessons from epidemiologic studies. Am J Cardiol. 2008;101:D75–86.
45. Knekt P, Ritz J, Pereira MA, O'Reilly EJ, Augustsson K, Fraser GE, Goldbourt U, Heitmann BL, Hallmans G, Liu S, et al. Antioxidant vitamins and coronary heart disease risk: a pooled analysis of 9 cohorts. Am J Clin Nutr. 2004;80:1508–20.
46. Miller ER III, Guallar E. Vitamin E supplementation: what's the harm in that? Clin Trials. 2009;6:47–9, discussion 50–1.
47. Klipstein-Grobusch K, Grobbee DE, den Breeijen JH, Boeing H, Hofman A, Witteman JC. Dietary Fe and risk of myocardial infarction in the Rotterdam Study. Am J Epidemiol. 1999;149:421–8.
48. Qi L, van Dam RM, Rexrode K, Hu FB. Heme Fe from diet as a risk factor for coronary heart disease in women with type 2 diabetes. Diabetes Care. 2007;30:101–6.
49. Hu FB, Rimm EB, Stampfer MJ, Ascherio A, Spiegelman D, Willett WC. Prospective study of major dietary patterns and risk of coronary heart disease in men. Am J Clin Nutr. 2000;72:912–21.
50. Snowdon DA, Phillips RL, Fraser GE. Meat consumption and fatal ischemic heart disease. Prev Med. 1984;13:490–500.
51. Bernstein AM, Sun Q, Hu FB, Stampfer JE, Willet M, Willet WC. Major dietary protein sources and risk of coronary heart disease in women. Circulation. 2010;122:876–83.
52. Czernichow S, Vergnaud AC, Galan P, Arnaud J, Favier A, Faure H, Huxley R, Hercberg S, Ahluwalia N. Effects of long-term antioxidant supplementation and association of serum antioxidant concentrations with risk of metabolic syndrome in adults. Am J Clin Nutr. 2009;90:329–35.
53. Willett WC, Buzzard MI. Foods and nutrients. In: Willet W, editor. Nutritional epidemiology. 2nd ed. New York: Oxford University Press; 1998. p. 18–32.
54. Vivekananthan DP, Penn MS, Sapp SK, Hsu A, Topol EJ. Use of antioxidant vitamins for the prevention of cardiovascular disease: meta-analysis of randomised trials. Lancet. 2003;361:2017–23.
55. Martini LA, Catania AS, Ferreira SR. Role of vitamins and minerals in prevention and management of type 2 diabetes mellitus. Nutr Rev. 2010;68:341–54.
56. Larsson SC, Wolk A. Magnesium intake and risk of type 2 diabetes: a meta-analysis. J Intern Med. 2007;262:208–14.
57. Kipnis V, Freedman LS. Impact of exposure measurement error in nutritional epidemiology. J Natl Cancer Inst. 2008;100:1658–9.
58. Kipnis V, Freedman LS, Brown CC, Hartman AM, Schatzkin A, Wacholder S. Effect of measurement error on energy-adjustment models in nutritional epidemiology. Am J Epidemiol. 1997;146:842–55.