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Cross-sectional Associations between Healthy Eating Index and Sex Steroid Hormones in Men – National Health and Nutrition Examination Survey 1999–2002

Zhehui Chen[1], **Giulia Pestoni**[1], **Katherine A McGlynn**[2], **Elizabeth A Platz**[3,4,5], **Sabine Rohrmann**[1]

[1]Division of Chronic Disease Epidemiology; Epidemiology, Biostatistics and Prevention Institute; University of Zurich; Zurich, Switzerland

[2]Division of Cancer Epidemiology & Genetics, National Cancer Institute, NIH, DHHS, Bethesda, MD, USA

[3]Department of Epidemiology, Johns Hopkins Bloomberg School of Public Health, Baltimore, MD, USA

[4]Sidney Kimmel Comprehensive Cancer Center at Johns Hopkins, Baltimore, MD, USA

[5]Department of Urology and the James Buchanan Brady Urological Institute, Johns Hopkins University School of Medicine, Baltimore, MD.

Abstract

Background: Diet plays an important role in health and is a modifiable risk factor for chronic diseases. In men, sex steroid hormones influence, and are influenced, by a number of health states. Specific dietary patterns have been found to alter sex steroid hormone levels in observational and intervention studies. Thus, we hypothesized that dietary patterns captured the by the Healthy Eating Index (HEI) are associated with serum concentrations of sex steroid hormones and sex hormone-binding globulin (SHBG).

Objectives: The objective is investigating the association between HEI and sex steroid hormones and SHBG in a general US population of men.

Methods: We used data on serum sex steroid hormones and SHBG levels, HEI, and other variables collected in the National Health and Nutrition Examination Survey (NHANES), 1999–2002. A total of 550 men greater than 20 years old were included in the analysis. The cross-sectional associations between HEI (from 0 to 100 points, higher score equates to a healthier diet) with natural logarithm transformed concentrations of total and free testosterone, total and free estradiol, and SHBG were evaluated with multivariable linear regression models and adjusted for potential confounders. We also stratified by the body mass index (BMI) and race/ethnicity, and tested for interactions.

Corresponding author: Sabine Rohrmann, Division of Chronic Disease Epidemiology, Epidemiology, Biostatistics and Prevention Institute, University of Zurich, Zurich, Switzerland, sabine.rohrmann@uzh.ch, phone: +41 44 634 5256.

Results: HEI showed a significant inverse association with free estradiol (p=0.03), but was not associated with total or free testosterone, total estradiol or SHBG concentrations. Neither BMI nor race/ethnicity statistically significantly modified the association between HEI and sex steroid hormone levels.

Conclusion: The present cross-sectional analysis in a representative sample of US men showed no consistent association between eating habits, sex steroid hormones and SHBG. Longitudinal studies are needed to further investigate potential associations.

Keywords

androgens; hormones; diet; Healthy Eating Index

Introduction

Diet acts in diverse ways that affect nutrition status, body composition, life expectancy, and the development of diseases. Analyses of diet and health have mainly addressed individual nutrients to understand disease etiology; however, it also may be helpful to examine overall diet patterns and quality (1). One method to evaluate dietary patterns is to use diet-quality scores based on recommended diets or dietary guidelines. The Healthy Eating Index (HEI) measures the concordance of dietary patterns with *Dietary Guidelines for Americans* and the food guide pyramid (2), and has been used to predict chronic disease risks. In the Health Professionals Follow-up Study (HPFS), men with the highest HEI scores, which equate to a healthier diet, had a lower risk of cardiovascular diseases (CVD) than did those with the lowest scores (3). The same inverse association between HEI and CVD risks was observed among women in the Nurses' Health Study (NHS) (4).

Circulating concentrations of sex steroid hormones have been previously associated with health conditions in men, including type-2 diabetes (T2D) (5), chronic kidney disease (6) and CVD (7,8). In previous studies using data from the National Health and Nutrition Examination Survey (NHANES), men with low concentrations of free testosterone, bioavailable testosterone, and estradiol had a higher risk of premature death, whereas men with low sex hormone-binding globulin (SHBG) levels had a decreased risk (9), which was consistent with some, but not all previous studies (10).

Testosterone and estradiol are the major sex steroids in the human body. The bioavailable fraction of steroid hormones can travel across cellular membranes and bind to the nuclear steroid receptors; they circulate either free or bound to albumin (11). The calculation of free testosterone and free estradiol using mass action models based on concentrations of total hormone levels and their affinity constants for albumin and SHBG has been shown to be valid (12,13).

Diet influences hormone production, metabolism, excretion and availability (14,15) and, in turn, may affect the incidence of hormone-related diseases. The association between diet and sex steroid hormones in men has been investigated in both observational studies and intervention trials. Regarding the choice of protein sources, omnivorous men have been reported to have significantly lower SHBG levels compared to vegetarians, and free

testosterone levels have been reported to be higher in omnivorous compared to vegetarian men (16). Soy product intake was significantly inversely correlated associated with serum estradiol concentrations among Japanese men (17). Moreover, high-fat, low-fiber diets have been reported to resulted in a marginally significant increase in total testosterone in healthy men (18). Increased SHBG levels and lowered testosterone/estradiol ratio have been observed when meat was replaced by isoenergetic soybean in a diet intervention among healthy men (19).

While some evidence supports that intakes of specific nutrients and diet patterns influence sex steroid hormone levels, it is relevant to consider the influence of overall dietary quality, since the health outcomes of a diet may be due to potential synergistic effects of nutrients within the diet as a whole. Fung et al. (20) observed that among postmenopausal women in the NHS, higher AHEI (Alternative Healthy Eating Index, a variant of HEI) was associated with decreased plasma estradiol and increased plasma SHBG concentrations. Similar results were found in NHS II, where AHEI was inversely associated with premenopausal estrogen concentrations (21).

Given the prior findings, we hypothesized that general eating patterns and overall diet quality may influence chronic disease risks via changes in sex steroid hormone among men. To date, we are not aware of any studies that have investigated the association between HEI and concentrations of sex steroid hormones and SHBG among men. The aim of our study was therefore to investigate whether general eating patterns, measured by the HEI, were associated with sex steroid hormone or SHBG concentrations in a nationally representative study of U.S. men.

Methods

The NHANES is a series of cross-sectional surveys conducted by the Centers for Disease Control and Prevention to assess the health and nutrition status of the U.S. population. For the current study, we used data collected in the 1999/2000 and 2001/2002 cycles. A total of 21,004 individuals were interviewed at home, of which 19,759 also completed an examination at the Mobile Examination Centers.

The HEI, used to express diet quality, was calculated for 17,102 individuals from dietary information (24-hours dietary recall). The HEI incorporates 10 components of five food groups (grains, vegetables, fruits, milk, meat), four nutrients (total fat, saturated fat, cholesterol, sodium), and a measure of variety in food intake. The total HEI score can range from 0 to 100 points, with 0 indicating poor adherence and 100 indicating maximal adherence to the *Dietary Guidelines for Americans*.(2)

We also used data on sex steroid hormones and SHBG concentrations measured in surplus serum for 1,007 males. Full hormone information was available for 989 of the men. In the current study, we excluded men younger than 20 years old (n = 347), invalid estradiol level (n = 51), extreme sex steroid hormone levels (negative value below the detection limit, n= 56), missing information on HEI (n = 27), missing data on body mass index (BMI) or waist circumference (n = 29), energy intake (n = 23), cigarette smoking (n = 1), alcohol intake (n =

31), physical activity (n = 29), and men previously diagnosed with prostate cancer (n = 13). After the exclusions, 550 men remained for this analysis.

Serum was separated from subsample blood obtained during the NHANES examination and stored at −80°C. Concentrations of total testosterone, total estradiol, and SHBG were measured at Children's Hospital Boston MA by electrochemiluminescence immunoassays on a 2010 Elecsys autoanalyzer (Roche Diagnostics, Indianapolis, IN). Laboratory methods and quality control statistics were reported previously (18). Using the mass action equation model (13), free testosterone was calculated from testosterone, albumin, and SHBG values; free estradiol was calculated from estradiol, albumin and SHBG concentrations (12).

All statistical analyses were performed with R (Version 3.4.1) taking NHANES sampling weights into account. HEI was categorized into quintiles, with 110 persons in each quintile. Hormones and SHBG levels were transformed using the natural logarithm. Geometric means and their 95% confidence intervals (CIs) were calculated from multivariable linear regression models. Model 1 was adjusted for age (continuous), race/ethnicity (non-Hispanic white, non-Hispanic black, Mexican-American, other). Model 2was further adjusted for BMI (continuous), waist circumference (continuous), energy intake (continuous), smoking status (never, former, current), alcohol consumption (non-drinker, > 0 to <1 drinks/week, ≥ 1 drinks/week to <1 drinks/day, ≥ 1 drinks/day), physical activity (neither moderate nor vigorous activity, either moderate or vigorous activity, both moderate and vigorous activity), and mutually adjusted for other hormones (total testosterone, total estradiol, and SHBG adjusted for each other; free testosterone adjusted for total estradiol; free estradiol adjusted for total testosterone). Since positive correlations were observed between total testosterone and total estradiol, total testosterone and SHBG and total estradiol and SHBG, we mutually adjusted for other hormones in the analyses to be able to identify independent associations. We additionally examined associations stratified by BMI (non-overweight vs. overweight defined as BMI ≥ 25 kg/m^2) and race/ethnicity (non-Hispanic white, non-Hispanic black, and Mexican-American; other race/ethnicity [not reported because of small sample size]) and tested for interactions for overweight and race/ethnicity by including a cross-product term in a separate model, which was evaluated using the Wald test.

Results

Men in this analysis were 20+ years old years old. The race/ethnicity distribution of the participants was 50.1% non-Hispanic white, 18.9% non-Hispanic black, 23.1% Mexican American, and 7.8% other race/ethnicity. The mean HEI score was 63.8 points. Men in higher quintiles of HEI were less likely to be non-Hispanic black, were better educated, had higher household income, were less likely to currently smoke or less likely to be exposed to passive smoke, and were more likely to engage in both moderate and vigorous physical activity (Table 1). BMI and waist circumference were associated with HEI quintiles in a U-shaped manner.

Table 2 presents adjusted geometric mean sex steroid hormone levels. In an age and race/ethnicity-adjusted model (model 1), HEI was inversely associated with both total (p-trend = 0.02) and free (p-trend = 0.01) estradiol levels, and possibly positively associated

with SHBG (p-trend = 0.07). No significant trends were observed between HEI and concentrations of total testosterone (p-trend = 0.71) or free testosterone (p-trend = 0.43).

In the multivariable model with mutual adjustments for hormones (model 2), the inverse association between HEI and free estradiol levels was attenuated but remained significant (p-trend = 0.03). In contrast, the association with total estradiol was no longer significant (p-trend = 0.13). No significant trends were observed between HEI and concentrations of total testosterone (p-trend = 0.92) or free testosterone (p-trend = 0.97).

We conducted further analyses stratified by BMI and race/ethnicity, as these two variables might be potential effect modifiers. No statistically significant association was observed within BMI groups, and no interaction was observed between BMI and HEI with sex steroid hormone or SHBG levels (Supplementary Table 1). Also, race/ethnicity did not modify the association between HEI and sex steroid hormone levels (Supplementary Table 2). However, a significant positive association was observed for non-Hispanic black men between free testosterone levels and HEI (p-trend = 0.03), and an inverse association was seen between SHBG concentrations and HEI (p-trend = 0.04); these associations were not observed among non-Hispanic white or Mexican-American men. Free estradiol concentrations were inversely associated with HEI (p-trend = 0.02) in non-Hispanic white men, but not in non-Hispanic black or in Mexican-American men. Table 3 shows a summary of adjusted associations of HEI and sex steroid hormones or SHBG by different regression models.

Discussion

Sex steroid hormones and their binding protein SHBG play essential roles in men's health. Epidemiological studies have reported that sex steroid hormones are associated with age, race, BMI, alcohol consumption, smoking behaviors, and physical activity (22–24). In this study, we used multivariable models adjusted for these confounding factors to examine whether sex steroid hormone concentrations were associated with general eating patterns measured by the HEI.

To our knowledge, this is the first study to examine associations between HEI and sex steroid hormone levels in a sample of men representative of the general US population. Limited previous dietary intervention trials suggested possible associations between dietary components and the levels of total testosterone, free testosterone and SHBG (16,17,19,25–27). In this study, free estradiol was inversely associated with HEI, and the association remained statistically significant after adjustment for confounding variables.

The main characteristics of the study sample by HEI quintiles showed U-shaped associations for body measurements such as BMI and waist circumference. In fact, men in the highest HEI quintile had higher mean BMI and waist circumference than men in the third or fourth quintile. HEI as a matrix derived from 24-hour dietary recalls is dependent on the self-report of participants; therefore, it is possible that participants with high BMI or already in a status of obesity tend to report healthier eating patterns either because they do indeed have a healthier diet to lose weight or, more likely, because they misreport their food intake.

Another explanation might be that men in the fifth quintile of HEI have maintained their muscle mass (i.e., are more robust) and thus have higher BMI.

In the analyses, we adjusted for age, race/ethnicity, BMI, waist circumference, energy intake, tobacco smoking, alcohol consumption and physical activity, and also mutually adjusted for hormones and SHBG levels. Age, race, BMI, and waist circumference are widely accepted factors to be associated with sex steroid hormone levels, and thus to be adjusted as potential confounders (23,24,28–30). We adjusted for both BMI and waist circumference since they reflect overall and central obesity (28). Lifestyle factors such as tobacco smoking, alcohol consumption, and physical activity have also been shown to affect sex steroid hormone concentrations to some extent (22,24,29,30). In the design of original HEI (2), the recommended number of servings per day varies depending on computed energy requirements. However, it does not take into account total energy intake. Because total energy intake is related to HEI, we took this into account as an adjustment variable.

Both total and free estradiol concentrations were inversely associated with HEI when the model was adjusted only for age and race/ethnicity in both analyses by HEI quintiles. A similar pattern was observed by Fung et al. (20) in women. The fully adjusted model attenuated the significant association between free estradiol and HEI, which suggests that the association could partly be explained by other variables. So far, only studies in women observed inverse associations between HEI and circulating estradiol concentrations (20)(21). A lower estradiol concentration in women among those with high HEI might be attributable to higher intake of dietary fibers, vegetable fats, and omega-3 fatty acids, which were inversely associated with estradiol concentrations in previous studies (20)(21).

Neither BMI nor race/ethnicity modified the associations between HEI and hormone concentrations. Although we observed some statistically significant associations in ethnic/racial groups (positive association between HEI and free testosterone concentrations in non-Hispanic black men; SHBG levels were significantly lower in non-Hispanic black men with higher HEI; an inverse association between HEI and free estradiol was observed only in non-Hispanic white men), these might be due to chance. To our knowledge, no other studies investigated the association between HEI and sex hormone levels by racial and ethnic groups.

The population used in this study was from a large, nationally representative sample of the non-institutionalized U.S. population. The composition and size of the study population made it possible to stratify our analyses by BMI and race/ethnicity. Also, sex steroid hormone concentrations were measured under rigorous evaluation of assay performance and quality control.

However, there are a few limitations in this study. The measurements of hormones were conducted with electrochemiluminescence immunoassays rather than HPLC tandem mass spectrophotometry. Regarding the bioavailable hormone concentrations, we calculated free testosterone and free estradiol levels by using mass action equations (12,13). However, the calculation method for free estradiol was only calibrated and compared with direct measurements of radioimmunoassay in postmenopausal women, which might

not be appropriate to directly apply in men. In addition, we relied on a single time point measurement of sex steroid hormones, which may not represent the men's usual concentrations. As for the dietary patterns, the data used for HEI calculation were collected from a 24-hours dietary recall, and does not necessarily reflect an individual's habitual diet. A longitudinal cohort study is needed to investigate the association between chronic eating patterns and the long-term effect on sex steroid hormones.

Conclusion

In conclusion, our analyses did not show consistent associations between healthy eating patterns and circulating levels of sex steroid hormones or SHBG. A significantly inverse association between HEI and free estradiol concentrations, indicating a healthier eating pattern might lower the concentrations of free estradiol in adult men, was mainly seen for non-Hispanic white men, but not for other racial/ethnic groups.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

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Table 1

Weighted Characteristics by Quintiles of HEI for Adult Men, NHANES 19999–2002

	HEI quintile				
	Q1	**Q2**	**Q3**	**Q4**	**Q5**
N (unweighted)	110	110	110	110	110
Age (years), mean (se)	42.99 (1.42)	40.08 (1.44)	42.70 (1.33)	43.63(1.36)	46.52 (1.50)
Race/ethnicity					
Non-Hispanic white	75.3%	70.5%	66.4%	71.7%	77.0%
Non-Hispanic black	12.2%	11.8%	12.0%	6.6%	6.4%
Mexican-American	7.5%	6.3%	8.1%	8.1%	7.5%
Others	5.1%	11.4%	13.4%	13.5%	9.1%
BMI (kg/m^2), mean (se)	28.69 (0.49)	27.59 (0.47)	27.25 (0.68)	27.04 (0.46)	28.61 (0.50)
Waist circumference (cm), mean (se)	101.05 (1.36)	97.17 (1.33)	96.73 (1.58)	97.23 (1.23)	101.74 (1.37)
Education					
Less than High School	26.1%	21.6%	18.5%	18.4%	13.7%
High School Diploma	27.6%	32.4%	20.3%	16.7%	17.3%
More than High School	44.5%	46.0%	61.2%	64.8%	69.0%
Unknown	1.7%	0.0%	0.0%	0.0%	0.0%
Household income					
Low	33.69%	30.06%	27.89%	20.34%	23.38%
Middle	32.15%	26.26%	29.54%	22.36%	26.73%
High	34.15%	43.68%	42.57%	57.29%	49.89%
Cigarette smoking					
Never	44.7%	37.7%	33.4%	56.6%	49.5%
Former	21.9%	29.5%	36.4%	24.0%	32.1%
Current	33.33%	32.8%	30.2%	19.4%	18.4%
Cigarette smoke exposure					
Unexposed	40.8%	41.7%	50.7%	63.9%	69.6%
Passively exposed	13.5%	22.7%	11.4%	8.1%	6.8%
Actively exposed	45.7%	35.6%	37.9%	28.0%	23.6%
Alcohol consumption					
Non-drinker	31.6%	18.4%	14.8%	19.9%	21.1%
>0 to <1 /week	19.5%	22.1%	30.4%	24.9%	29.0%
≥1/week to <1/day	25.5%	34.4%	40.0%	28.2%	26.3%
≥1/day	23.3%	25.0%	14.8%	27.0%	23.6%
Physical activity					
No moderate or vigorous	34.6%	30.7%	38.0%	24.1%	29.9%
Moderate or vigorous	42.6%	38.5%	39.3%	45.6%	37.5%
Moderate and vigorous	22.9%	30.8%	22.7%	30.3%	32.6%

Abbreviations: HEI, Healthy Eating Index; Q1~Q5, HEI quintile 1~5; N, number of participants in each group; BMI, body mass index

Table 2

Geometric Mean (95% CI) of Serum Sex Steroid Hormone Concentrations by Quintiles of HEI for Adult Men, NHANES 1999–2002

HEI Quintile	Model 1*		Model 2*	
Total testosterone (ng/ml)	mean	95% CI	mean	95% CI
Q1	4.52	(4.06, 5.02)	4.70	(4.29, 5.15)
Q2	4.85	(4.36, 5.38)	4.78	(4.37, 5.23)
Q3	4.97	(4.46, 5.54)	4.84	(4.41, 5.30)
Q4	4.83	(4.34, 5.36)	4.75	(4.34, 5.19)
Q5	4.27	(3.84, 4.75)	4.43	(4.05, 4.84)
p trend	0.71		0.92	
Free testosterone (ng/ml)				
Q1	0.090	(0.082, 0.098)	0.090	(0.081, 0.100)
Q2	0.098	(0.090, 0.107)	0.096	(0.087, 0.106)
Q3	0.091	(0.083, 0.010)	0.091	(0.082, 0.101)
Q4	0.091	(0.083, 0.099)	0.091	(0.083, 0.101)
Q5	0.079	(0.072, 0.087)	0.081	(0.073, 0.090)
p trend	0.43		0.97	
Total estradiol (pg/ml)				
Q1	27.85	(24.43, 31.74)	28.07	(23.94, 32.90)
Q2	30.31	(26.62, 35.52)	29.95	(25.68, 34.93)
Q3	25.23	(22.07, 28.85)	25.18	(21.51, 29.48)
Q4	23.82	(20.88, 27.17)	23.70	(20.33, 27.64)
Q5	24.75	(21.64, 28.30)	25.23	(21.59, 29.48)
p trend	0.02		0.13	
Free estradiol (pg/ml)				
Q1	0.710	(0.621, 0.811)	0.716	(0.601, 0.852)
Q2	0.776	(0.679, 0.886)	0.771	(0.652, 0.914)
Q3	0.613	(0.535, 0.703)	0.610	(0.514, 0.724)
Q4	0.591	(0.517, 0.676)	0.589	(0.498, 0.697)
Q5	0.612	(0.534, 0.702)	0.617	(0.520, 0.732)
p trend	0.008		0.03	
SHBG (nmol/L)				
Q1	32.33	(28.73, 36.38)	33.75	(29.80, 38.22)
Q2	32.48	(28.89, 36.51)	32.55	(28.81, 36.76)
Q3	38.69	(34.28, 43.66)	37.60	(33.19, 42.60)
Q4	37.05	(32.90, 41.72)	35.99	(31.89, 40.62)
Q5	35.86	(31.78, 40.47)	37.20	(32.90, 42.06)
p trend	0.07		0.08	

*Analyses were adjusted for age, race/ethnicity (Model 1), plus BMI, waist circumference, energy intake, tobacco smoking, alcohol consumption, physical activity, and mutually adjusted for other hormones (total testosterone, total estradiol, and SHBG adjusted for each other; free testosterone adjusted for total estradiol; free estradiol adjusted for total testosterone; Model 2)

Abbreviations: HEI, Healthy Eating Index; Q1~Q5, HEI quintile 1~5; CI, confidence interval; SHBG, sex hormone-binding globulin

Table 3

Summary of Adjusted Associations between HEI Quintile and Sex Steroid Hormone Levels in Adult Men, NHANES 1999–2002

		BMI		Race/ethnicity		
	Multivariate and other hormones adjusted	**Overweight and obesity**	**Non-overweight**	**Non-Hispanic White**	**Non-Hispanic black**	**Mexican American**
Total testosterone	-	-	-	-	↑	-
Free testosterone	-	-	-	-	↑*	-
Total estradiol	-	-	-	-	-	-
Free estradiol	↓*	↓	-	↓*	-	-
SHBG	↑	-	-	↑	↓*	-

↓* denotes a statistically significant inverse association. The arrows without an asterisk denote a possible association ($0.05 \leq P \leq 0.10$).

↑* denotes a statistically significant positive association.

Abbreviations: BMI, Body Mass Index; Q1~Q5, HEI quintile 1~5; CI, confidence interval; SHBG sex hormone-binding globulin