ORIGINAL RESEARCH

Higher Dietary Acid Load Is Associated With an Increased Risk of Calcium Oxalate Kidney Stones

Fahimeh Haghighatdoost, PhD, ⋆ *Reyhaneh Sadeghian, BSc,* † *Cain C. T. Clark, PhD,* ‡ *and Behnood Abbasi, PhD* †

Objectives: Diet-dependent net acid load may influence the risk of kidney stone formation by affecting calcium and citrate excretion. However, to date, little research has investigated the relationship between dietary acid load and kidney stones. Therefore, this study sought to assess whether a diet high in potential acid load was related to the risk of calcium oxalate stone formation.

Methods: This case-control study was conducted on 430 participants (including 215 newly diagnosed patients with calcium oxalate stones and 215 controls matched for sex and age). Dietary intake was assessed using a validated food frequency questionnaire over the preceding year. Dietary acid load was estimated based on the potential renal acid load (PRAL) and net endogenous acid production (NEAP). The association between dietary acid load indices and kidney stone was examined using multivariable logistic regression.

Results: Mean PRAL (standard error) was significantly lower in cases versus controls (−5.3 ± 1.3 vs. −1.7 ± 1.3, $P = .048$). Corresponding values for NEAP were 39.4 ± 0.8 and 41.8 ± 0.8, respectively ($P = .032$). After adjustment for potential confounders, the odds ratios (95% confidence intervals) of calcium oxalate stones in the top tertile of PRAL and NEAP were 1.45 (0.89-2.38, $P = .136$) and 1.88 (1.14-3.09, $P = .013$), respectively. Adjustment for potassium and protein in 2 separate models did not substantially change the results.

Conclusions: A diet high in potential acid load, measured by NEAP, was associated with higher risk of calcium oxalate stone formation, independent of potassium and protein intake. Prospective longitudinal studies are warranted to confirm the veracity of our results.

Introduction

NEPHROLITHIASIS, A CONDITION associated with the presence and formation of kidney stones, is highly prevalent, with a growing trend worldwide,[1] and has been shown to double the risk of chronic kidney disease or end-stage renal disease.[2] Calcium stones, particularly calcium oxalate stones, are the most frequent type of kidney stones, and are found in up to 80% of cases.[3,4] Although 46% of stone events in women and 57% in men are attributable to hereditary, non-modifiable, factors,[5] approximately half of all stone events are caused by modifiable risk factors, and therefore can be prevented through altering environmental and lifestyle factors.

There is strong evidence highlighting the role of dietary intake in kidney stone formation.[6–9] Indeed, the available literature suggests that a healthy dietary pattern, such as the dietary approach to stop hypertension diet, rich in whole grains, fruit, vegetables, calcium, and unsaturated fatty acids, but low in salt, saturated fatty acids, sugar-sweetened beverages, and animal proteins, is associated with lower risk of kidney stones.[6–9] A mechanism by which dietary intake may influence the risk of kidney stone formation is altering dietary acid load. Although the components of a healthy diet including calcium, fruit, and vegetables (which are rich in potassium and magnesium) may increase urinary pH, the components of an unhealthy dietary pattern such as animal proteins and SSB may decrease urinary pH, and consequently favor calcium oxalate stone formation.[10–12] Indeed, in response to metabolic acidosis, the kidneys attempt to restore acid-base balance, resulting in an increased calcium and oxalate excretion and decreased excretion of citrate, thereby facilitating the formation of calcium oxalate crystals.[13,14] In addition, urinary calcium and oxalate excretion may be increased by dietary calcium and animal proteins,[15] which can potentially affect dietary acid load.

In epidemiologic studies, dietary acid load has been calculated using 2 approaches: (1) the potential renal acid load (PRAL), which takes into account the intestinal absorption rates of protein, potassium, phosphorus, magnesium, and calcium[16,17] and (2) the net endogenous acid

⋆*Isfahan Cardiovascular Research Center, Cardiovascular Research Institute, Isfahan University of Medical Sciences, Isfahan, Iran.*

†*Department of Nutrition, Electronic Health and Statistics Surveillance Research Center, Science and Research Branch, Islamic Azad University, Tehran, Iran.*

‡*Centre for Intelligent Healthcare, Coventry University, Coventry, United Kingdom.*

Financial Disclosure: The authors declare that they have no relevant financial interests.

Address correspondence to Behnood Abbasi, PhD, Department of Nutrition, School of Medical Sciences and Technology, Science and Research Branch, Islamic Azad University, Shohada Hesarak Blvd, Daneshgah Square, Sattari Expressway, Tehran 1477893855, Iran. E-mail: abbasi.b@srbiau.ac.ir

1051-2276/$36.00

https://doi.org/10.1053/j.jrn.2020.08.012

production (NEAP), which is based on the total amount of ingested protein and potassium.[18] Moreover, the validity of these approaches has been approved in several studies.[16-18]

To our knowledge, only a few studies have investigated the association of dietary acid load with calcium oxalate stones to date.[12,19,20] Although these studies have consistently shown a higher risk for kidney stones in individuals with higher scores of dietary acid load,[12,19,20] due to the distinct paucity of data in this context, further studies in populations with different lifestyle and risk factors are warranted to better elucidate the association. Thus, the present study sought to evaluate whether an acidifying diet is associated with risk of calcium oxalate stones formation.

Materials and Methods

Study Population

This case-control study was carried out between September 2017 and June 2018. A convenience sample of 215 cases referring to Shahid Hasheminezhad hospital, Tehran, Iran, whose nephrolithiasis was pathologically confirmed by a nephrologist during the past 3 months was selected. Age- (5-year age group) and sex-matched controls (n = 215) were recruited from the general population through advertisements and word of mouth through people who participated in our study, or who opted to participate but were not eligible. Prior to data collection, ethical approval (ethical approval number: IR.IAU.SRB.REC.1398.146) was obtained from Iran national committee for ethics in biomedical research. The major inclusion criteria for both cases and controls were (1) aged ≥18 years and (2) a body mass index (BMI) <35 kg/m^2. We excluded subjects with BMI >35 kg/m^2, due to additional medications for metabolic disorders. Indeed, variations in the number and doses of these medicines, along with other medical interventions, which has known, direct effects on urine composition, in addition to unknown interactions, made it impossible to control or match the effect of these interventions. Subjects were excluded if they had one of the following criteria: (1) being on a specific diet; (2) having a history of inflammatory bowel disease, irritable bowel syndrome, ileostomy, celiac, hyperthyroidism, or hyperparathyroidism; and (3) being pregnant or post-menopausal. At the beginning of study, all participants provided a written informed consent.

Dietary Intake Assessment

Data regarding dietary intake over the preceding year were collected using a Persian validated, self-reported, 147-item food frequency questionnaire (FFQ).[21] Participants were instructed on how to complete the FFQ by a trained nutritionist, and asked to determine how often they had consumed each food item during the past year, according to the given portion sizes. All food items were converted to grams/day using household measures,[22] and daily energy and nutrients intake was estimated using Nutritionist IV software modified for Iranian foods (First Databank Division, The Hearts Corporation, San Bruno, CA). In the present study, subjects who left more than 20% of FFQ items unanswered or their estimated energy intake was <800 or >4200 kcal/d were excluded.

Dietary Acid Load Measurement

To estimate dietary acid load indices, we first adjusted the components of dietary acid load indices for energy using the residual method,[23] and subsequently used in the suggested formula for PRAL and NEAP. The PRAL score was calculated using the formula suggested by Remer and Manz, which takes into account the intestinal absorption of protein, phosphorus, potassium, calcium, and magnesium[24]:

$$\mathrm{PRAL(mEq/d)} = (\mathrm{protein(g/d)} \times 0.49) + (\mathrm{P(mg/d)} \times 0.037) - (\mathrm{K(mg/d)} \times 0.021) - (\mathrm{Ca(mg/d)} \times 0.013) - (\mathrm{Mg(mg/d)} \times 0.026)$$

The NEAP was also measured through the formula suggested by Frassetto et al.[18]:

$$\mathrm{NEAP} = [(54.5 * \mathrm{protein(g/d)}) / \mathrm{potassium\ (mEq/d)}] - 10.2.$$

Assessment of Covariates

Data regarding demographic variables, including age, sex, education, alcohol drinking, smoking, and past medical history, were collected using a self-reported questionnaire. Physical activity was assessed using the International Physical Activity Questionnaire,[25] where its validity and reliability have been approved.[26] Body weight was measured, while participants were minimally clothed and unshod, using SECA scales, and recorded to the nearest 100 g. Height was measured using a fixed-wall tape, with participants unshod. BMI was calculated by dividing body weight (kg) by height (m^2).

Statistical Analysis

Participants were divided into 3 groups according to the tertiles of dietary acid load indices (PRAL and NEAP). Differences in general characteristics of participants, between cases and controls, were assessed using an independent sample *t*-test for continuous variables and a chi-squared test for categorical variables. Data were reported as means ± standard error (SE). Age-, sex-, and energy-adjusted dietary intakes across the tertiles of PRAL and NEAP were compared using analysis of covariance. Multiple logistic regression was performed to estimate odds ratios (OR) and 95% confidence intervals (CIs) for having kidney stones in the crude and adjusted models. In model 1, we

Table 1. Comparison of General Characteristics Between Cases and Controls*

Characteristic	Cases (n = 215)	Controls (n = 215)	*P* value†
Age (y)	39.2 ± 0.8	39.6 ± 0.8	.78
BMI (kg/m^2)	27.1 ± 0.3	26.9 ± 0.3	.56
Physical activity (MET.min)/wk	2,270.9 ± 188.7	2,708.5 ± 186.9	.10
Hypertension or diabetes (%)	22.8	14.9	.036
Alcohol drinking (%)	18.1	12.1	.080
Smoker (%)	26.5	11.2	<.0001
Calcium supplements (%)	5.1	10.2	.046
Multivitamin supplements (%)	5.1	9.3	.093
Education (y)			<.0001
<12	77.2	54.0	
12-16	14.9	25.6	
16	7.9	20.5	
Dietary intakes			
Total energy (kcal/d)	2,394.9 ± 98.7	2,448.3 ± 58.2	.642
Carbohydrate (g/d)	393.9 ± 2.9	390.6 ± 2.9	.405
Protein (g/d)	93.4 ± 1.0	98.5 ± 1.0	<.0001
Fat (g/d)	88.8 ± 1.2	88.2 ± 1.2	.736
Cholesterol (mg/d)	335.1 ± 13.6	313.2 ± 13.6	.257
Calcium (mg/d)	1,117.2 ± 21.0	1,198.3 ± 21.0	.007
Magnesium (mg/s)	473.7 ± 5.7	496.9 ± 5.7	.004
Potassium (mg/d)	4,132.1 ± 54.6	4,155.8 ± 54.6	.759
Phosphorus (mg/d)	1,690.8 ± 19.9	1,778.2 ± 19.9	.0002
Sodium (mg/d)	4,592.3 ± 58.9	4,335.6 ± 58.9	.002
Dietary fiber (g/d)	52.8 ± 1.3	50.5 ± 1.3	.217
Vitamin C (mg/d)	172.1 ± 4.7	162.4 ± 4.9	.167
Fruits (g/d)	376.3 ± 13.7	317.9 ± 13.7	.003
Vegetables (g/d)	297.4 ± 12.8	324.1 ± 12.8	.142
Meat (g/d)	105.0 ± 3.4	106.2 ± 3.4	.792
Whole grains (g/d)	138.8 ± 6.0	144.9 ± 6.0	.473
Refined grains (g/d)	361.9 ± 8.9	351.2 ± 8.9	.397
Dairy (g/d)	350.1 ± 15.8	397.6 ± 15.8	.034
Nuts (g/d)	13.0 ± 1.1	13.6 ± 1.1	.687
Legumes (g/d)	65.0 ± 3.1	67.1 ± 3.1	.624
Soft drink (cc/d)	58.0 ± 5.6	40.5 ± 5.6	.027
Sugar sweetened beverages (cc/d)	23.9 ± 2.8	14.6 ± 2.8	.018
Tea (cc/d)	883.5 ± 56.8	757.6 ± 56.8	.118
Coffee (cc/d)	16.9 ± 3.6	22.8 ± 3.6	.252
PRAL	−5.3 ± 1.3	−1.7 ± 1.3	.048
NEAP	39.4 ± 0.8	41.8 ± 0.8	.032

ANCOVA, analysis of covariance; BMI, body mass index; NEAP, net endogenous acid production; PRAL, potential renal acid load; SE, standard error.

*Values are mean ± SE unless indicated.

†*P*-values were calculated by independent *t*-test for continuous variables, and χ^2 for categorical variables and ANCOVA for dietary intakes (adjusted for age, sex, energy).

controlled for the confounding effect of education. In model 2, additional adjustment was made for calcium supplement consumption, smoking, hypertension, and diabetes mellitus. Due to independent role of protein and potassium in the development of kidney stone according to the literature,[12] model 3 and model 4 were further controlled for dietary potassium and protein intake, respectively. *P* for linear trends was determined by considering tertiles of PRAL and NEAP as linear continuous variables in the logistic regression model. All statistical analyses were performed using Statistical Package for Social Sciences (SPSS, Inc., Chicago, IL; version 20). Statistical significance was accepted at $P < .05$.

Results

The mean age of participants was 39.4 ± 11.9 y (77.2% male), and mean (SE) of PRAL and NEAP was significantly different between cases and controls (PRAL in cases: −5.3 ± 1.3 and in controls: −1.7 ± 1.3, $P = .048$; and NEAP in cases: 39.4 ± 0.8 and in controls: 41.8 ± 0.8, $P = .032$).

Table 1 compares the general characteristics as well as dietary intakes between cases and controls. Overall, compared with controls, cases were more likely to be hypertensive, diabetic, a smoker, consume alcoholic beverages, and less educated. In contrast, calcium supplement consumption was more prevalent among controls than

Table 2. Dietary Intakes of Participants Across the Tertiles of Dietary Acid Load Indices*

	PRAL				NEAP			
Indicator	T1 (n = 143)	T2 (n = 144)	T3 (n = 143)	*P* value†	T1 (n = 143)	T2 (n = 143)	T3 (n = 144)	*P* value†
Cases (n)	80	72	63		87	65	63	
Total energy (kcal/d)	2,795.5 ± 58.6	2,527.8 ± 58.1	2,721.2 ± 59.1	.004	2,823.0 ± 58.2	2,499.6 ± 58.0	2,720.5 ± 58.6	<.001
Carbohydrate (g/d)	405.1 ± 3.4	392.4 ± 3.4	379.3 ± 3.5	<.0001	400.6 ± 3.5	396.0 ± 3.5	380.2 ± 3.5	<.0001
Protein (g/d)	89.5 ± 1.1	93.2 ± 1.1	105.2 ± 1.2	<.0001	88.1 ± 1.2	95.8 ± 1.2	103.9 ± 1.2	<.0001
Fat (g/d)	87.3 ± 1.5	89.5 ± 1.5	88.8 ± 1.5	.563	89.7 ± 1.5	86.9 ± 1.5	88.9 ± 1.5	.390
Cholesterol (mg/d)	286.7 ± 16.6	309.0 ± 16.6	376.8 ± 16.7	<.001	289.4 ± 16.8	316.7 ± 16.8	366.1 ± 16.8	.005
Calcium (mg/d)	1,157.6 ± 26.2	1,186.7 ± 26.1	1,128.7 ± 26.3	.301	1,162.0 ± 26.0	1,216.9 ± 26.0	1,094.8 ± 26.0	.005
Magnesium (mg/d)	506.3 ± 7.0	468.8 ± 7.0	480.9 ± 7.0	.001	495.1 ± 7.1	491.2 ± 7.1	469.6 ± 7.1	.026
Potassium (mg/d)	4,776.0 ± 54.9	4,027.7 ± 54.7	3,629.1 ± 55.1	<.0001	4,741.5 ± 54.7	4,132.8 ± 54.7	3,561.8 ± 54.6	<.0001
Phosphorus (mg/d)	1,666.3 ± 24.3	1,709.0 ± 24.2	1,828.4 ± 24.4	<.0001	1,676.6 ± 24.8	1,762.5 ± 24.8	1,764.3 ± 24.8	.018
Sodium (mg/d)	4,388.0 ± 73.6	4,427.7 ± 73.3	4,576.5 ± 74.0	.170	4,345.9 ± 73.5	4,435.0 ± 73.5	4,610.0 ± 73.5	.038
Dietary fiber (g/d)	52.1 ± 1.7	53.1 ± 1.6	49.8 ± 1.7	.363	49.6 ± 1.7	53.5 ± 1.7	51.9 ± 1.7	.260
Vitamin C (mg/d)	228.5 ± 5.6	154.7 ± 5.6	116.8 ± 5.6	<.0001	220.4 ± 5.8	162.1 ± 5.8	117.8 ± 5.8	<.0001
Fruits (g/d)	492.6 ± 14.2	339.7 ± 14.2	209.1 ± 14.3	<.0001	477.9 ± 14.6	353.4 ± 14.6	211.0 ± 14.6	<.0001
Vegetables (g/d)	389.2 ± 15.2	288.2 ± 15.1	255.1 ± 15.2	<.0001	382.7 ± 15.3	295.7 ± 15.3	254.3 ± 15.3	<.0001
Meat (g/d)	89.1 ± 3.9	97.2 ± 3.9	130.6 ± 3.9	<.0001	87.8 ± 3.9	98.9 ± 3.9	129.9 ± 3.9	<.0001
Whole grains (g/d)	122.0 ± 7.2	131.8 ± 7.2	171.8 ± 7.3	<.0001	126.3 ± 7.4	137.9 ± 7.4	161.3 ± 7.4	.003
Refined grains (g/d)	316.0 ± 10.8	364.2 ± 10.7	389.5 ± 10.8	<.0001	311.2 ± 10.7	363.0 ± 10.7	395.3 ± 10.7	<.0001
Dairy (g/d)	338.6 ± 15.7	374.3 ± 15.8	348.7 ± 15.8	.258	350.0 ± 15.7	389.1 ± 15.7	322.5 ± 15.7	.011
Nuts (g/d)	16.9 ± 1.4	11.9 ± 1.4	11.1 ± 1.4	<.0001	14.4 ± 1.4	13.8 ± 1.4	11.7 ± 1.4	.367
Legumes (g/d)	63.7 ± 3.8	64.2 ± 3.8	70.4 ± 3.8	.394	59.9 ± 3.8	71.1 ± 3.8	67.2 ± 3.8	.107
Soft drink (cc/d)	42.4 ± 6.9	53.5 ± 6.9	51.9 ± 7.0	.471	48.1 ± 6.9	40.4 ± 6.9	59.3 ± 6.9	.161
Sugar sweetened beverages (cc/d)	21.1 ± 3.5	19.4 ± 3.5	17.1 ± 3.5	.717	22.5 ± 3.5	19.6 ± 3.5	15.6 ± 3.5	.373
Tea (cc/d)	1,221.3 ± 65.9	760.5 ± 65.6	480.3 ± 66.2	<.0001	1,185.1 ± 66.5	801.9 ± 66.6	477.1 ± 66.5	<.0001
Coffee (cc/d)	25.0 ± 4.5	13.9 ± 4.5	20.8 ± 4.5	.215	24.2 ± 4.5	19.3 ± 4.5	16.2 ± 4.5	.456
PRAL	−23.0 ± 0.9	−3.3 ± 0.9	15.8 ± 0.9	<.0001	−22.3 ± 0.9	−3.2 ± 0.9	14.8 ± 0.9	<.0001
NEAP	30.0 ± 0.6	39.4 ± 0.6	52.4 ± 0.6	<.0001	29.7 ± 0.6	39.4 ± 0.6	52.6 ± 0.6	<.0001

NEAP, net endogenous acid production; PRAL, potential renal acid load; SE, standard error.

*Values are mean ± SE. Nutrients and food groups were adjusted for age, sex, and total energy intake. Energy intake was adjusted for age and sex.

†From analysis of covariance.

Table 3. Multivariable-Adjusted Odds Ratios and 95% Confidence Intervals for Kidney Stones Across the Tertiles of Dietary Acid Load Indices

	PRAL				NEAP			
Model	T1 (n = 143)	T2 (n = 144)	T3 (n = 143)	*P* value*	T1 (n = 143)	T2 (n = 143)	T3 (n = 144)	*P* value*
Crude	1 (reference)	1.27 (0.80-2.02)	1.61 (1.01-2.57)	.045	1 (reference)	1.86 (1.16-2.98)	2.00 (1.25-3.20)	.004
Model 1	1 (reference)	1.28 (0.80-2.07)	1.47 (0.91-2.39)	.115	1 (reference)	1.83 (1.13-2.96)	1.83 (1.13-2.97)	.014
Model 2	1 (reference)	1.18 (0.73-1.93)	1.46 (0.89-2.39)	.133	1 (reference)	1.79 (1.09-2.93)	1.89 (1.15-3.10)	.012
Model 3	1 (reference)	1.18 (0.73-1.93)	1.45 (0.89-2.38)	.136	1 (reference)	1.80 (1.10-2.95)	1.88 (1.14-3.09)	.013
Model 4	1 (reference)	1.21 (0.72-2.04)	1.49 (0.88-2.51)	.138	1 (reference)	1.95 (1.15-3.30)	2.06 (1.21-3.52)	.010
Model 5	1 (reference)	1.20 (0.73-1.96)	1.40 (0.84-2.32)	.194	1 (reference)	1.81 (1.10-2.97)	1.81 (1.09-3.01)	.020
Model 6	1 (reference)	1.14 (0.69-1.86)	1.53 (0.93-2.52)	.096	1 (reference)	1.72 (1.05-2.83)	1.99 (1.21-3.30)	.007

Model 1, adjusted for education; Model 2, further adjustment was made for calcium supplement consumption, smoking, hypertension, and diabetes mellitus; Model 3, dietary potassium intake was additionally adjusted; Model 4, adjusted for education, calcium supplement consumption, smoking, hypertension and diabetes mellitus, and protein intake; Model 6, adjusted for education, calcium supplement consumption, smoking, hypertension and diabetes mellitus, and sodium intake; NEAP, net endogenous acid production; PRAL, potential renal acid load.

*P-value was calculated from Mantel-Haenszel χ^2 test.

cases. Controls had greater intake of protein, calcium, magnesium, phosphorus, vegetables, and dairy, but lower intake of sodium, fruits, soft drinks, and SSB, compared with cases. NEAP and PRAL scores were significantly higher in controls versus cases.

Table 2 details the dietary intake of participants across the tertiles of dietary acid load indices. Higher scores of both PRAL and NEAP were significantly associated with lower intake of carbohydrate, magnesium, potassium, vitamin C, fruits, vegetables, nuts, and tea, but higher intakes of protein, cholesterol, phosphorus, sodium, meats, and whole and refined grains. Calcium and dairy product consumption were not significantly different across the tertiles of PRAL, but displayed a decreasing trend across the tertiles of NEAP. Higher PRAL scores were associated with lower intake of nuts, but no difference was found across the tertiles of NEAP. Consumption of fat, fiber, and legumes was similar across the tertiles of both PRAL and NEAP.

Crude and multivariable-adjusted ORs (95% CIs) for calcium oxalate kidney stones are demonstrated in Table 3 and they are stratified by sex in Table 4. In the crude model, individuals in the third tertile of PRAL had a 61.0% (95% CI 1.01-2.57, $P = .045$) greater risk for kidney stones, in comparison with those in the first tertile. However, adjustment for education weakened this association to the point it was no longer significant (OR = 1.47, 95% CI 0.91-2.39, $P = .115$). Further controlling for calcium supplement consumption, smoking, hypertension and diabetes mellitus, and dietary potassium or protein intake did not yield any substantial effect on the association. We also compared the dietary intake of cases with controls according to the tertiles of dietary acid load indices (Supplementary Table 1), which indicated that the dietary intake of cases and controls across the tertiles of PRAL was comparable, and the trends of changes were similar for most nutrients.

Table 4. Multivariable Adjusted Odds Ratios and 95% Confidence Intervals for Kidney Stones Across the Tertiles of Dietary Acid Load Indices Stratified by Sex

	PRAL				NEAP			
Model	T1	T2	T3	*P* value*	T1	T2	T3	*P* value*
Male (n)	99	107	126		100	105	127	
Crude	1 (reference)	1.17 (0.69-1.96)	1.78 (1.08-2.92)	.020	1 (reference)	1.18 (0.69-1.99)	2.13 (1.30-3.51)	.002
Model 1	1 (reference)	1.10 (0.65-1.86)	1.75 (1.06-2.89)	.023	1 (reference)	1.11 (0.65-1.89)	2.09 (1.27-3.46)	.003
Model 2	1 (reference)	1.03 (0.61-1.76)	1.68 (1.02-2.78)	.035	1 (reference)	1.06 (0.62-1.82)	1.98 (1.19-3.29)	.006
Model 3	1 (reference)	1.03 (0.60-1.75)	1.67 (1.01-2.77)	.036	1 (reference)	1.06 (0.62-1.81)	1.97 (1.19-3.28)	.006
Model 4	1 (reference)	1.05 (0.55-2.01)	1.36 (0.57-3.25)	.465	1 (reference)	1.12 (0.57-2.19)	2.05 (0.82-5.13)	.105
Female	44	37	17		43	38	17	
Crude	1 (reference)	2.44 (1.11-5.38)	2.10 (0.90-4.90)	.046	1 (reference)	3.04 (1.33-6.95)	1.66 (0.74-3.73)	.114
Model 1	1 (reference)	2.47 (1.12-5.47)	2.06 (0.88-4.82)	.051	1 (reference)	3.10 (1.35-7.12)	1.62 (0.71-3.67)	.135
Model 2	1 (reference)	2.58 (1.16-5.73)	2.12 (0.90-4.98)	.044	1 (reference)	3.24 (1.40-7.49)	1.66 (0.73-3.78)	.111
Model 3	1 (reference)	2.57 (1.14-5.79)	2.02 (0.85-4.81)	.058	1 (reference)	3.10 (1.32-7.26)	1.65 (0.72-3.80)	.124
Model 4	1 (reference)	3.12 (1.09-8.89)	2.79 (0.60-12.92)	.150	1 (reference)	3.18 (1.05-9.61)	1.66 (0.34-8.12)	.453

BMI, body mass index; Model 1, adjusted for energy, age, and sex; Model 2, additional adjustment was made for alcohol; Model 3, further adjustment for BMI; Model 4, protein and potassium intakes were additionally adjusted; NEAP, net endogenous acid production; PRAL, potential renal acid load.

*P-value was calculated from Mantel-Haenszel χ^2 test.

Regarding the NEAP, in the crude model, the risk of stone formation in the highest tertile was 2 times higher compared with the first tertile (OR = 2.0, 95% CI 1.25-3.20). Adjustment for education in model 1 weakened the association, but remained statistically significant (OR = 1.83, 95% CI 1.13-2.97, $P = .014$). Further control for calcium supplement consumption, smoking, and hypertension and diabetes mellitus slightly changed the association. In model 3, adjustment for dietary potassium intake strengthened the association (OR = 2.06, 95% CI = 1.21-3.52, $P = .010$), and adjustment for dietary protein intake in model 4 did not ameliorate the association (OR = 1.81, 95% CI 1.09-3.01, $P = .021$).

Discussion

In the current case-control study, we found that higher dietary acid load, as measured by NEAP, but not PRAL, was associated with increasing the risk for calcium oxalate stones, approximately, 2-fold. Moreover, this association was independent from dietary potassium or protein intake.

The association between lower urinary pH and the risk of kidney stone formation has been well established[27]; however, to our knowledge, and in spite of the effect of dietary acid load on urinary pH,[28] information on the association between dietary acid load and the risk of kidney stone is pauce. In a large prospective examination of 3 cohort studies (the Health Professionals Follow-Up Study, the Nurses' Health Study I, and the Nurses' Health Study II), there was a 41% higher risk of calcium oxalate stone incidence for those participants in the top quintile of protein to potassium ratio.[12] Furthermore, this association remained significant even after further adjustment for animal protein and potassium intake. Similarly, our results highlighted that association between NEAP and kidney stone was independent from protein and potassium intake. Two other case-control studies, among Italian populations, consistently suggested that stone formers had greater dietary acid load, compared with controls.[19,20] However, in our study, we observed that PRAL could not predict the risk of calcium oxalate stone after adjustment for potential confounders.

Although in the current study dietary acid load was greater in controls in comparison with cases, the risk of calcium oxalate stone was higher in cases. This difference might be related to potassium and protein intakes. Indeed, while cases and controls in our study had similar potassium intake, protein intake was greater in controls than cases. This difference in protein intake was attributed to differences in dairy product intake, which was consumed to a greater extent by controls versus cases. Dairy products represent good sources of magnesium and calcium, which can decrease the risk of calcium oxalate stone formation; however, they are not considered in the NEAP calculation. In addition, although fruit intake was greater in cases compared with controls, it did not lead to differences in fiber intake between cases and controls as a protective factor against kidney stones. Therefore, it is conceivable that protein sources likely play a varying and important role in predicting the risk of calcium oxalate stone formation.

In the present study, we also observed that cases were less educated than controls, which is consistent with literature. This might be explained by the relatively lower awareness of less educated people with respect to kidney stone prevention and the differences in lifestyle factors, other than dietary intake. In addition, individuals with different educational levels, and therefore, socioeconomic statuses, may experience different levels of psychological stress which is closely correlated with kidney stones.[29] However, due to a lack of data in this regard, we could not evaluate this association; therefore, it would be pragmatic for further research to acutely consider the potential impact of socioeconomic status, concomitant to other confounding variables.

Several mechanisms may explain the positive association between dietary acid load and kidney stones formation. First, a diet low in potential acid load is high in fruits and vegetables, which contains high amounts of water, and therefore can increase urine volume and decrease relative supersaturation for calcium oxalate.[12] Second, a chronic low grade acid load may putatively lead to higher urinary calcium excretion (possibly through bone buffering), lower urinary citrate excretion, and lower urinary pH, which would increase the risk of stone formation.[13,14] Third, dietary potassium, as a component of dietary acid load indices, can influence the reabsorption of bicarbonate in the proximal tubule, alter proton transport and renal ammonia production in the distal tubule, and consequently affect acidification.[30] Furthermore, a diet with low potential of acid load contains higher amounts of plant foods which have various salts of organic anions (malate, citrate, galacturonate, tartrate), and can exert alkalinizing effects through bicarbonate generation and thereby decrease the risk of crystal formation.[31] There may be also some nutraceutical effects of foods (such as cranberries) which should be considered. Indeed, consistently, several studies have elucidated that a plant-based diet like the dietary approach to stop hypertension and Mediterranean eating styles are associated with decreased risk of kidney stones.[6-9]

Despite representing a novel addition to the literature, our study has several limitations that should be considered. First, we did not have information regarding urinary composition; therefore, we could not validate the measures of dietary acid load (PRAL and NEAP) using the urinary pH. Nevertheless, there are several studies suggesting that urinary pH is correlated with dietary acid load indices.[12,28] Second, our study design does not allow causal inferences to be made; however, assuming a causal relationship based on our results, dietary interventions aimed at decreasing potential acid load would considerably decrease the risk of kidney stones, and have a profound impact on the

economic and societal burden of kidney stones. Third, case-control studies are inevitably prone to selection and recall bias; thus, further prospective cohort studies are required to confirm the veracity of these results. Fourth, we used a validated FFQ; however, it has some associated measurement errors[32,33] which can result in misclassification of participants and an inaccurate estimation of the association.[34] It is also worth mentioning that FFQ cannot provide precise estimation of salt and water intake, and we did not measure sodium excretion in 24-hour urinary sample or water consumption in a day, which might impact the risk of stone formation. Although sodium restriction has been frequently recommended in patients with kidney stones, adherence is generally poor,[35] and therefore, it is unlikely that it would considerably change our findings. In support of this assertion, we made further adjustment for sodium intake, estimated through FFQ, across the tertiles of PRAL and NEAP, and observed no substantial change in our findings. Regarding water intake, we assumed that it is likely similar between cases and controls, since our cases were newly diagnosed kidney stone patients, and therefore, since we assessed their dietary intakes within the preceding year, current dietary changes could not be considerably different in comparison with controls. In addition, given that water derived from foods is the first determinant of water intake,[36] when we considered liquid food volumes (including dairy, tea, coffee, soft drinks, and SSB), it was not substantially different between cases and controls (1333 vs. 1233 g/d, respectively). Therefore, it is unlikely that these 2 factors confounded our results. Finally, did not consider the role of hereditary factors in dietary acid load-kidney stones relations. Besides these limitations, our study has a number of strengths. Indeed, to the authors' knowledge, this is the first study, in this context, to have been conducted in a Middle-Eastern country, with a markedly different dietary pattern and lifestyle to Western dietary habits. In addition, the adequate sample size of this study allowed us to control a number of relevant confounding factors.

Overall, in the current case-control study, we found that a diet high in potential acid load, determined by NEAP, was associated with an increased risk of calcium oxalate stone. However, PRAL was only significantly associated with increased risk of calcium oxalate stones in the crude model. In addition, the association between NEAP and calcium oxalate stones was independent from dietary protein or potassium intake. Longitudinal studies are warranted to confirm the veracity of our findings in different populations. In addition, due to nutraceutical effect of cranberries on the kidneys, future studies are needed to consider such foods, as well.

Practical Application

Estimating dietary acid load using PRAL and NEAP formulas cannot consistently predict the risk of calcium oxalate kidney stone. Our results suggest that NEAP might be superior to the PRAL to predict the risk of kidney stone formation. Nonetheless, due to lack of epidemiological studies in this regard, further studies should be conducted to confirm the validity of our findings.

CRediT Authorship Contribution Statement

Behnood Abbasi: Writing - Review & Editing, Conceptualization, Methodology, Supervision. Fahimeh Haghighatdoost: Writing - Original Draft Preparation, Formal analysis, Data Curation. Reyhaneh Sadeghian: Writing - Original Draft preparation, Provision of study materials. Cain C. T. Clark: Writing - Review & Editing.

Acknowledgment

We want to acknowledge all the participants and the financial support provided to us by Science and Research Branch of Islamic Azad University.

Supplementary Data

Supplementary data related to this article can be found at https://doi.org/10.1053/j.jrn.2020.08.012.

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