Article

Dietary Acid, Age, and Serum Bicarbonate Levels among Adults in the United States

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Summary

Background and objectives Greater dietary acid has been associated with lower serum bicarbonate levels in patients with CKD. Whether this association extends to the general population and if it is modified by age are unknown.

Design, setting, participants, & measurements This study examined the association of the dietary acid load, estimated by net endogenous acid production, with serum bicarbonate levels in adult participants in the National Health and Nutrition Examination Survey 1999–2004.

Results The mean serum bicarbonate was 24.9 mEq/L (SEM=0.1), and the mean estimated net endogenous acid production was 57.4 mEq/d (SEM=0.4). Serum bicarbonate was linearly associated with age, such that the oldest participants had the highest serum bicarbonate levels. After multivariable adjustment, participants in the highest quartile of net endogenous acid production had 0.40 mEq/L (95% confidence interval, −0.55 to −0.26) lower serum bicarbonate and a 33% (95% confidence interval, 3 to 72) higher likelihood of acidosis compared with those participants in the lowest quartile. There was a significant interaction by age of the association of net endogenous acid production with serum bicarbonate (P=0.005). Among participants 20–39, 40–59, and ≥60 years old, those participants in the highest net endogenous acid production quartile had 0.26 (95% confidence interval, −0.49 to −0.03), 0.60 (95% confidence interval, −0.92 to −0.29), and 0.49 (95% confidence interval, −0.84 to −0.14) mEq/L lower serum bicarbonate, respectively, compared with participants in the lowest quartile.

Conclusion Greater dietary acid is associated with lower serum bicarbonate in the general US population, and the magnitude of this association is greater among middle-aged and elderly persons than younger adults.

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Introduction

Acid–base homeostasis is tightly controlled in normal humans, and even minimal disturbances can have deleterious effects. Chronic metabolic acidosis as a manifestation of CKD is believed to have adverse affects on bone metabolism and skeletal muscle breakdown (1). In addition, reduced albumin synthesis, insulin resistance, and progression of kidney disease have all been linked with acidosis (2,3).

A low-level metabolic acidosis may exist even in individuals without kidney disease (4). This subclinical acidosis, mediated in part by the effects of aging and the Western diet, may have important sequelae on bone, skeletal muscle, and insulin resistance (5–7). Oral alkali administered to postmenopausal women has reduced bone resorption, improved bone mineral density, and decreased urinary nitrogen excretion, suggesting that acidosis has clinical sequelae even in this population (8–10).

In normal subjects, endogenous acid production, which varies according to the composition of the diet, affects steady state acid–base balance in metabolic studies (11). The magnitude of the net acid load is determined by the balance of acid precursors and base precursors in the diet. This dietary stimulus for acid production can vary as much as 10-fold between individuals consuming diets of differing compositions (11). Net endogenous acid production (NEAP) may be estimated using the dietary protein and potassium contents as indices of acid and base precursors, respectively. In CKD patients, higher NEAP has been associated with lower serum bicarbonate (12). However, the effect of the dietary acid load on acid–base status has not been examined in the general population. Because acidosis is associated with adverse outcomes on a population-wide level, even in people without kidney disease (6,7,13–15), determining the extent to which dietary acid may be a modifiable contributor to low-grade acidosis could have important public health implications. In addition, because older persons have lower capacity to excrete an acid load (16), this effect might be modified by age.

We hypothesized that dietary acid would be associated with acid–base status in the general population and that this effect would be greater in older than younger persons. We tested these hypotheses in adult participants ages 20 years and older with data on serum bicarbonate and dietary intake in the National

Health and Nutrition Examination Survey (NHANES) 1999–2004.

Materials and Methods
Study Population
NHANES 1999–2004 was a nationally representative survey of the noninstitutionalized civilian population in the United States (17). A stratified, multistage, probability sampling design was used to select participants. The NHANES protocol was approved by the National Center for Health Statistics ethics review board, and written informed consent was obtained from all participants. Overall, 13,274 adults≥20 years of age completed the interview and examination components and had available serum bicarbonate data. We excluded participants who had a history of emphysema or chronic bronchitis by self-report (n=969), were pregnant at the time of examination (n=680), had an estimated GFR (eGFR)<15 ml/min per 1.73 m² (n=36), had reported use of carbonic anhydrase inhibitors (n=4), had invalid dietary data (n=531), or were missing covariate data (n=1273). Thus, 9781 participants were available for analysis.

Data Collection
Information on household income, education, smoking, comorbidities, and medication use in the previous month was obtained by self-report. Race/ethnicity was self-identified. Smoking was classified as never, former, or current smoker. Low socioeconomic status was defined as less than 100% of the poverty index. Data on dietary intake were obtained from a 24-hour dietary recall questionnaire. The diet-dependent net acid load was estimated as NEAP (mEq/d)= [54.5×protein (g/d)/potassium (mEq/d)]−10.2 (18). Hypertension was defined as systolic BP≥140 mmHg, diastolic BP≥90 mmHg, physician diagnosis, and/or antihypertensive medication use (19). Diabetes mellitus was defined as a physician diagnosis while not pregnant or the current use of insulin or oral hypoglycemic medications. Cardiovascular disease (CVD) was defined by self-report of a physician diagnosis of congestive heart failure, coronary heart disease, angina, myocardial infarction, or stroke.

Serum chemistry values were measured using the Hitachi 917 Multichannel Analyzer (Roche Diagnostics, Indianapolis, IN) in 1999–2001 and the Beckman Synchron LX20 (Beckman Coulter Inc., Brea, CA) in 2002–2004. Serum albumin was measured by the bromocresol purple method. Serum creatinine was measured by a modified kinetic Jaffé reaction. Values from 1999 to 2000 were calibrated to the Cleveland Clinic laboratory standard by multiplying by 1.013 and then adding 0.147. Correction of values from 2001 to 2004 was not necessary. eGFR was calculated using the CKD Epidemiology Collaboration Equation (20).

Outcome Variables
Serum bicarbonate was measured in two laboratories by the phosphoenolpyruvate carboxylase method from 1999 to 2001 and with a pH-sensitive electrode from 2002 to 2004. Because the mean serum bicarbonate was 1.105±0.178 mEq/L higher (P<0.001) among all NHANES participants in 2003–2004 compared with 1999–2002, serum bicarbonate levels in 1999–2002 were adjusted by adding 1.105 mEq/L, which has been done previously (13). Acidosis was defined as

serum bicarbonate<23 mEq/L. Because Kidney Disease Outcomes Quality Initiative recommends maintaining the serum bicarbonate≥22 mEq/L in patients with CKD, we also repeated our analyses when defining acidosis as serum bicarbonate<22 mEq/L.

Statistical Analyses
All analyses used NHANES-appropriate sampling weights and accounted for the complex multistage cluster design using the survey command in Stata 11.1 (Stata Corporation, College Station, TX). The distributions of participant characteristics were examined by quartiles of NEAP. Linear and logistic regression models were created to examine the associations of NEAP with serum bicarbonate and acidosis, respectively. NEAP was analyzed as a continuous variable and within quartiles to examine nonlinear associations with either outcome. Models were created examining associations in the overall cohort without adjustment for additional covariates and then including age, sex, and race/ethnicity as covariates. Multivariable models were then created to include other potential confounders of the association of NEAP with acid–base status as covariates, including poverty and education (as markers of socioeconomic status), body mass index categories, smoking status, diagnosis of diabetes mellitus, hypertension, CVD, diuretic use, eGFR categories, log-transformed urine albumin–creatinine ratio, serum albumin, and log-transformed C-reactive protein. To determine whether participants with CKD drove our results, we repeated these analyses after excluding those participants with eGFR<60 ml/min per 1.73 m². Effect modification by age was tested by including a multiplicative interaction term in the models. Age-stratified analyses were then performed to separately examine associations of NEAP with serum bicarbonate and acidosis within age categories. A P value<0.05 was considered statistically significant.

Sensitivity Analyses
Because NEAP does not account for variations in the sulfur content of animal versus vegetable protein, we repeated our analyses using animal protein-derived endogenous acid production (ADEAP), which we computed by replacing total dietary protein in the NEAP calculation with protein derived from meat, poultry, or fish consumption defined using US Department of Agriculture Food Codes 20000000–28500000. We also examined the associations of dietary protein and potassium separately with serum bicarbonate using linear regression models that included dietary energy as a covariate (21). To determine if residual confounding caused by imprecise ascertainment of obesity affected our results, we repeated our analyses and included percent total body fat as a covariate among the subgroup of 9499 participants who underwent whole-body dual-energy x-ray absorptiometry. Details of the dual-energy x-ray absorptiometry protocol and data validation are available (http://www.cdc.gov/nchs/nhanes/dxx/dxa.htm) (22–24).

Results
Participant Characteristics
The mean age was 45.7 years (SEM=0.3), and the mean serum bicarbonate was 24.9 mEq/L (SEM=0.1). The mean

NEAP was 57.4 mEq/d (SEM=0.4). The median dietary protein and potassium intakes were 73 g/d (interquartile range [IQR]=52–100) and 63 mEq/d (IQR=45–86), respectively. Median estimated protein intake per kilogram body weight was 0.93 g/kg per day (IQR=0.66–1.30).

Participants with higher NEAP were younger, were more likely to be men, were more likely to be Mexican Americans or non-Hispanic blacks, were less likely to have hypertension, diabetes, or CVD, were less likely to use diuretics, and had higher eGFR (Table 1). They were more likely to be obese and current smokers, had lower dietary potassium and higher dietary protein intake, and had higher C-reactive protein levels. There was a progressive trend of lower NEAP among older participants (Figure 1).

Association of NEAP with Serum Bicarbonate

In unadjusted analysis, a 1 SD higher NEAP was associated with 0.22 mEq/L (95% confidence interval [95% CI], −0.28 to −0.16) lower serum bicarbonate (Table 2). Multivariable adjustment somewhat attenuated this association (−0.15 mEq/L [95% CI, −0.20 to −0.10] per 1 SD higher NEAP). Compared with participants in the lowest quartile of NEAP, participants in the highest quartile had 0.59 mEq/L (95% CI, −0.76 to −0.43) and 0.40 mEq/L (95%

Table 1.   Participant characteristics by quartiles of net endogenous acid production

Characteristic	Net Endogenous Acid Production (mEq/d)				P
	<39.3	39.3–52.9	53.0–69.9	≥70.0	
Number	2477	2403	2411	2490	
Age (yr)	50.7 (0.45)	48.4 (0.46)	44.0 (0.42)	39.8 (0.41)	<0.001
Women (%)	58.6 (1.1)	49.0 (1.3)	45.9 (1.5)	43.6 (1.2)	<0.001
Race/ethnicity (%)					<0.001
Non-Hispanic white	78.1 (1.7)	76.8 (1.5)	71.9 (2.0)	64.1 (2.3)	
Mexican American	5.4 (0.7)	7.9 (1.1)	7.8 (1.0)	8.7 (1.4)	
Non-Hispanic black	7.3 (0.9)	7.0 (0.7)	9.4 (1.0)	17.2 (1.7)	
Other	9.2 (1.4)	8.3 (0.9)	10.8 (1.5)	10.0 (1.3)	
Body mass index (kg/m^2; %)					<0.001
<18.5 (underweight)	2.2 (0.4)	1.4 (0.3)	0.9 (0.2)	1.5 (0.3)	
18.5–24.9 (normal)	37.5 (1.3)	33.4 (1.4)	30.5 (1.5)	29.3 (1.1)	
25–29.9 (overweight)	35.3 (1.2)	37.3 (1.3)	35.5 (1.1)	33.1 (1.4)	
≥30 (obese)	24.8 (1.1)	27.7 (1.3)	33.0 (1.8)	35.9 (1.0)	
Poverty (<100% poverty index; %)	11.2 (0.9)	11.5 (1.0)	11.1 (0.9)	16.5 (1.1)	<0.001
Less than high school diploma (%)	18.4 (1.3)	17.5 (1.2)	16.7 (0.9)	21.6 (1.3)	0.11
Smoking (%)					<0.001
Never	50.7 (1.5)	50.7 (1.8)	51.5 (1.5)	50.9 (1.9)	
Former	28.7 (1.3)	27.8 (1.5)	23.4 (1.3)	19.9 (1.1)	
Current	20.6 (1.2)	21.5 (1.2)	25.1 (1.4)	29.2 (1.7)	
Dietary protein (g/d)	60.6 (0.9)	81.3 (1.2)	92.7 (1.2)	99.3 (1.1)	<0.001
Dietary potassium (mEq/d)	82.0 (1.2)	78.4 (1.2)	71.1 (1.0)	54.6 (0.7)	<0.001
Diuretic use (%)	11.5 (0.8)	10.5 (1.0)	8.7 (0.8)	8.7 (0.8)	0.001
Hypertension (%)	43.5 (1.7)	42.1 (1.7)	36.1 (1.5)	34.9 (1.3)	<0.001
Cardiovascular disease (%)	10.6 (0.8)	7.9 (0.9)	7.6 (0.7)	6.0 (0.6)	<0.001
Diabetes mellitus (%)	7.7 (0.5)	6.7 (0.7)	6.8 (0.7)	6.2 (0.6)	0.03
Estimated GFR (ml/min per 1.73 m^2; %)					<0.001
≥120	6.8 (0.6)	8.1 (0.7)	10.1 (0.7)	16.6 (1.2)	
90–119	43.5 (1.7)	46.7 (1.3)	52.4 (1.3)	50.9 (1.6)	
60–89	40.5 (1.5)	38.9 (1.5)	33.4 (1.4)	28.6 (1.6)	
45–59	6.7 (0.6)	4.5 (0.5)	2.8 (0.3)	2.8 (0.3)	
30–44	1.9 (0.3)	1.4 (0.2)	0.9 (0.2)	1.0 (0.2)	
15–29	0.6 (0.1)	0.3 (0.1)	0.3 (0.1)	0.07 (0.04)	
Urine albumin–creatinine ratio (mg/g; %)					0.36
<30	90.8 (0.8)	90.5 (0.9)	92.9 (0.7)	91.1 (0.8)	
30–300	8.3 (0.8)	8.3 (0.9)	6.1 (0.6)	7.6 (0.7)	
>300	1.0 (0.2)	1.2 (0.3)	1.0 (0.2)	1.3 (0.3)	
Serum albumin (g/dl)	4.36 (1.2)	4.38 (1.0)	4.37 (1.2)	4.37 (1.1)	0.48
C-reactive protein (mg/dl; %)					0.02
<0.36	71.1 (1.1)	71.2 (1.2)	69.7 (1.3)	66.8 (1.2)	
0.36–0.99	19.7 (0.8)	20.4 (1.1)	20.8 (1.0)	24.0 (1.1)	
≥1	9.2 (0.8)	8.4 (0.7)	9.5 (0.9)	9.2 (0.7)	

Data are expressed as mean (SEM) or percent (SEM).
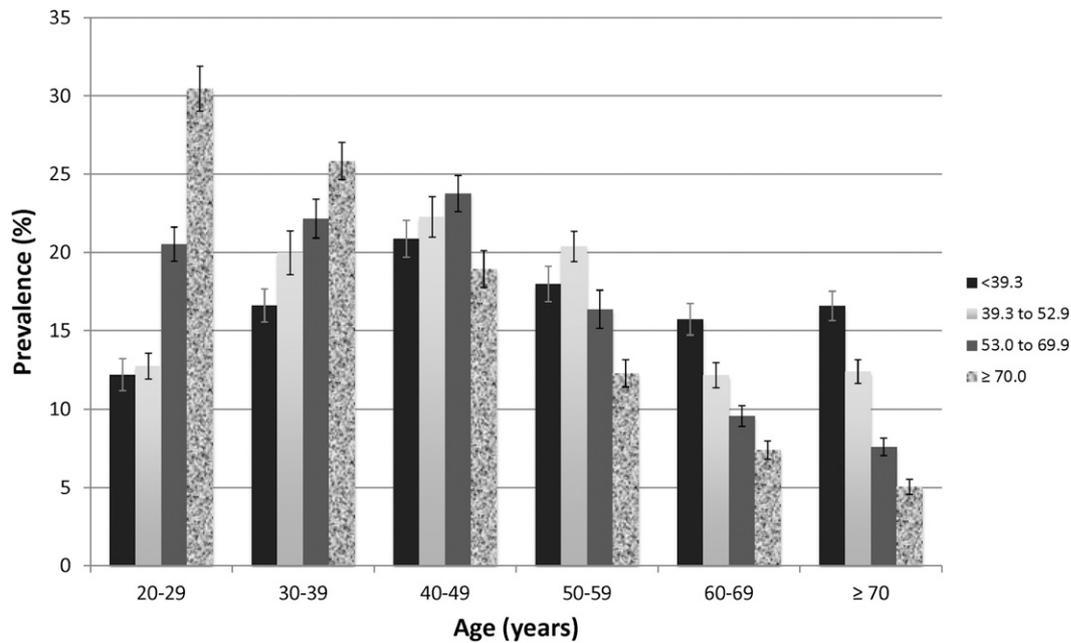
Figure 1. | **Association of estimated net endogenous acid production (NEAP; mEq/d) with age in 9781 participants of the National Health and Nutrition Examination Survey 1999–2004.** Each bar represents an NEAP quartile. Error bars represent SEMs.

Table 2. Association of net endogenous acid production with serum bicarbonate in 9781 participants of the National Health and Nutrition Examination Survey 1999–2004

Net Endogenous Acid Production	Coefficient (95% Confidence Interval)		
	Model 1	Model 2	Model 3
Continuous[a]	−0.22 (−0.28 to −0.16)[b]	−0.20 (−0.26 to −0.14)[b]	−0.15 (−0.20 to −0.10)[b]
<39.3 mEq/d	Reference	Reference	Reference
39.3–52.9 mEq/d	−0.12 (−0.27 to −0.04)	−0.13 (−0.28 to −0.01)	−0.11 (−0.26 to −0.03)[b]
53.0–69.9 mEq/d	−0.35 (−0.55 to −0.16)[b]	−0.33 (−0.52 to −0.14)[b]	−0.24 (−0.42 to −0.07)[b]
≥70.0 mEq/d	−0.59 (−0.76 to −0.43)[b]	−0.53 (−0.70 to −0.36)[b]	−0.40 (−0.55 to −0.26)[b]
P for trend	<0.001	<0.001	<0.001

Model 1: unadjusted. Model 2: adjusted for age, sex, and race/ethnicity. Model 3: model 2 adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated GFR, log-transformed urine albumin–creatinine ratio, serum albumin, and log-transformed C-reactive protein.
[a]Per SD higher net endogenous acid production (SD=26.4 mEq/d).
[b]$P<0.05$.

CI, −0.55 to −0.26) lower serum bicarbonate in unadjusted and multivariable-adjusted analyses, respectively.

We defined acidosis as a serum bicarbonate<23 mEq/L. A 1 SD higher NEAP was associated with a 1.09 (95% CI, 1.00 to 1.21) greater likelihood of acidosis after multivariate adjustment (Table 3). Participants in the highest NEAP quartile, compared with the lowest NEAP quartile, had a multivariable-adjusted odds ratio for acidosis of 1.33 (95% CI, 1.03 to 1.72). Our results were unchanged after excluding participants with eGFR<60 ml/min per 1.73 m² (Supplemental Tables 1 and 2). Similar results were found defining acidosis as serum bicarbonate<22 mEq/L (Supplemental Table 3).

Interaction of Age with NEAP and Serum Bicarbonate

In the fully adjusted model, a 10 year greater age was associated with a 0.24 mEq/L (95% CI, 0.18 to 0.31) higher serum bicarbonate. We next examined the distribution of serum bicarbonate across age groups (Figure 2). Serum bicarbonate was linearly associated with age, such that bicarbonate levels were highest among the oldest participants. Additional examination stratified by NEAP quartiles showed that the magnitude of this association was dependent on NEAP. The largest increase in serum bicarbonate across age groups was in the lowest NEAP quartile, whereas the smallest difference was seen in the highest quartile.
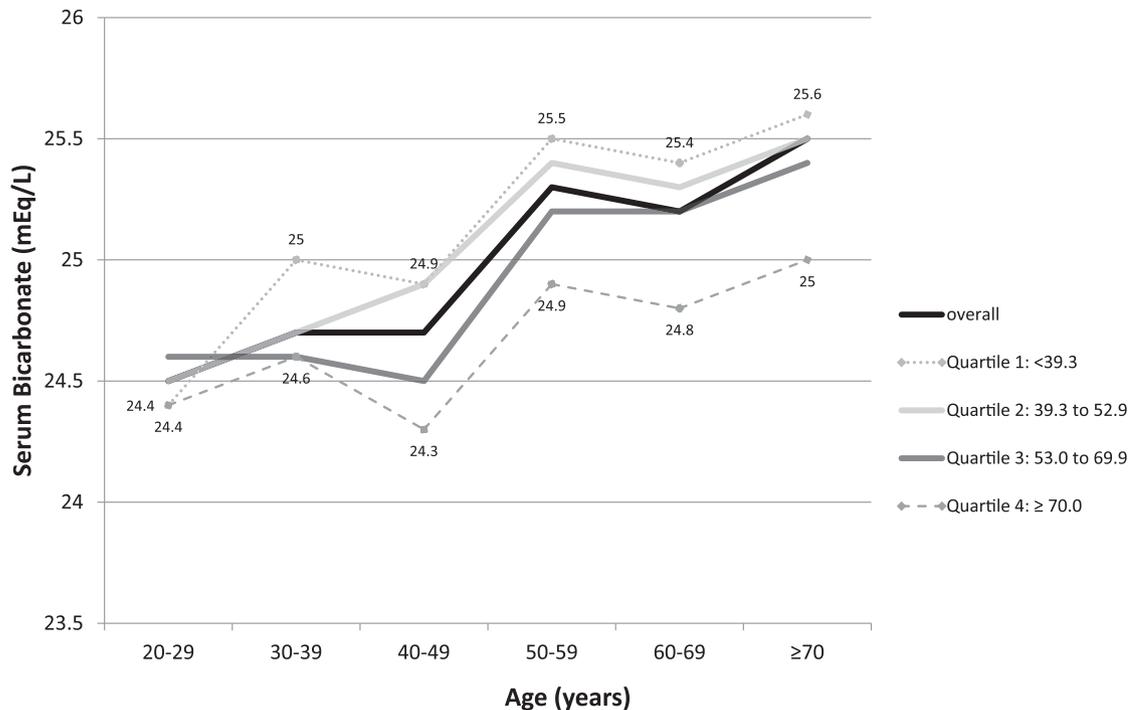
Table 3.   Odds ratio of acidosis by net endogenous acid production in 9781 participants of the National Health and Nutrition Examination Survey 1999–2004

Net Endogenous Acid Production	Odds Ratio (95% Confidence Interval)		
	Model 1	Model 2	Model 3
Continuous[a]	1.17 (1.08 to 1.28)[b]	1.15 (1.05 to 1.26)[b]	1.09 (1.00 to 1.21)
<39.3 mEq/d	Reference	Reference	Reference
39.3–52.9 mEq/d	1.11 (0.90 to 1.37)	1.12 (0.92 to 1.38)	1.11 (0.90 to 1.38)
53.0–69.9 mEq/d	1.38 (1.08 to 1.78)[b]	1.34 (1.05 to 1.73)[b]	1.25 (0.97 to 1.60)
≥70.0 mEq/d	1.61 (1.26 to 2.05)[b]	1.51 (1.16 to 1.95)[b]	1.33 (1.03 to 1.72)[b]
P for trend	<0.001	0.002	0.03

Acidosis defined as serum bicarbonate<23 mEq/L. Model 1: unadjusted. Model 2: adjusted for age, sex, and race/ethnicity. Model 3: model 2 adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated GFR, log-transformed urine albumin–creatinine ratio, serum albumin, and log-transformed C-reactive protein.
[a]Per SD higher net endogenous acid production (SD=26.4 mEq/d).
[b]$P<0.05$.

Figure 2. | Mean serum bicarbonate levels across age categories, overall, and within quartiles of estimated net endogenous acid production (NEAP; mEq/d) in 9781 participants of the National Health and Nutrition Examination Survey 1999–2004. Individual lines represent the overall population and individual NEAP quartiles. Within each age category, the mean serum bicarbonate is listed for the highest and lowest quartile of NEAP.

We found a significant interaction by age of the association of NEAP with serum bicarbonate ($P=0.005$). Participants were then stratified by age into three categories: 20–39, 40–59, and ≥60 years old (Table 4). Among participants in the youngest age group, NEAP as a continuous variable was not significantly associated with lower serum bicarbonate after multivariable adjustment. Compared with participants in the lowest NEAP quartile, participants in the highest NEAP quartile had 0.26 mEq/L (95% CI, −0.49 to −0.03) lower serum bicarbonate. Among participants 40–59 and ≥60 years old, NEAP was significantly associated with serum bicarbonate as both a continuous variable and within quartiles. In these age groups, after multivariable adjustment, participants in the highest NEAP quartile had 0.60 (95% CI, −0.92 to −0.29) and 0.49 (95% CI, −0.84 to −0.14) mEq/L lower serum bicarbonate, respectively, compared with those participants in the lowest quartile.

Table 4. Association of net endogenous acid production with serum bicarbonate within age categories

Net Endogenous Acid Production	Coefficient (95% Confidence Interval)		
	Model 1	Model 2	Model 3
Age 20–39 yr (*n*=3299)			
Continuous[a]	−0.05 (−0.14 to 0.03)	−0.12 (−0.20 to −0.03)[b]	−0.07 (−0.16 to 0.01)
<39.3 mEq/d	Reference	Reference	Reference
39.3–52.9 mEq/d	−0.10 (−0.42 to 0.21)	−0.17 (−0.49 to 0.14)	−0.12 (−0.42 to 0.18)
53.0–69.9 mEq/d	−0.14 (−0.48 to 0.20)	−0.27 (−0.56 to 0.02)	−0.15 (−0.40 to 0.11)
≥70.0 mEq/d	−0.25 (−0.51 to 0.02)	−0.40 (−0.64 to −0.17)[b]	−0.26 (−0.49 to −0.03)[b]
P for trend	0.05	0.001	0.03
Age 40–59 yr (*n*=3139)			
Continuous[a]	−0.29 (−0.39 to −0.19)[b]	−0.33 (−0.43 to −0.23)[b]	−0.26 (−0.37 to −0.16)[b]
<39.3 mEq/d	Reference	Reference	Reference
39.3–52.9 mEq/d	−0.03 (−0.34 to 0.28)	−0.13 (-0.43 to 0.17)	−0.11 (−0.40 to 0.18)
53.0–69.9 mEq/d	−0.36 (−0.65 to −0.08)[b]	−0.42 (−0.69 to −0.14)[b]	−0.35 (−0.63 to −0.07)[b]
≥70.0 mEq/d	−0.64 (−0.96 to −0.32)[b]	−0.76 (−1.08 to −0.43)[b]	−0.60 (−0.92 to −0.29)[b]
P for trend	<0.001	<0.001	<0.001
Age≥60 yr (*n*=3343)			
Continuous[a]	−0.21 (−0.35 to −0.07)[b]	−0.20 (−0.34 to −0.06)[b]	−0.17 (−0.30 to −0.04)[b]
<39.3 mEq/d	Reference	Reference	Reference
39.3–52.9 mEq/d	−0.11 (−0.38 to 0.16)	−0.10 (−0.37 to 0.18)	−0.11 (−0.39 to 0.17)
53.0–69.9 mEq/d	−0.22 (−0.50 to 0.06)	−0.18 (−0.45 to 0.09)	−0.15 (−0.40 to 0.09)
≥70.0 mEq/d	−0.58 (−0.93 to −0.23)[b]	−0.55 (−0.90 to −0.19)[b]	−0.49 (−0.84 to −0.14)[b]
P for trend	0.002	0.004	0.008

Model 1: unadjusted. Model 2: adjusted for age, sex, and race/ethnicity. Model 3: model 2 adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated GFR, log-transformed urine albumin–creatinine ratio, serum albumin, and log-transformed C-reactive protein. *P* for interaction by age=0.005.
[a]Per SD higher net endogenous acid production (SD=26.4 mEq/d).
[b]*P*<0.05.

We similarly found significant effect modification by age using acidosis as the outcome (*P*=0.02). There was no statistically significant association of NEAP with acidosis in the youngest age group (Table 5). There was a significant association of higher NEAP with greater likelihood of acidosis among participants 40–59 years old; there was a similar trend among those participants≥60 years of age, but it did not reach statistical significance.

Sensitivity Analyses

The distribution of ADEAP quartiles across age categories was similar to the distribution of NEAP quartiles (Supplemental Figure 1). Higher ADEAP was associated with lower serum bicarbonate (Supplemental Table 4), and there was a significant interaction by age of the association of ADEAP with serum bicarbonate (*P* for interaction=0.01) (Supplemental Table 3). Higher dietary protein (Supplemental Table 6) and lower dietary potassium (Supplemental Table 7) were each associated with lower serum bicarbonate, although the associations seemed more robust for potassium intake. Adjustment for percent total body fat did not affect our results (Supplemental Tables 8 and 9).

Discussion

The principle objective of this study was to investigate the association of dietary acid with metabolic acidosis in the general population and examine the possibility of effect modification by age of this association. We found that a higher dietary acid load, which was measured by NEAP, was associated with lower serum bicarbonate and a higher likelihood of acidosis in a nationally representative sample of Americans. In addition, the magnitude of these associations was greater in middle-aged and elderly participants than younger participants. This result suggests that the deleterious effects of dietary acid may be most prominent among older individuals.

Previous physiologic studies in normal adults have shown that differences in dietary acid affect extracellular pH and serum bicarbonate (8,11). Despite these variations in diet, renal acid excretion maintains acid–base homeostasis in normal persons, but this capacity declines with age (25). Our results extend these findings to indicate that dietary acid plays a role in determining acid–base status on a population-wide level and that this effect is most pronounced among older persons.

This finding could have important clinical implications. Even among individuals without kidney disease, a low-grade acidosis contributes to bone breakdown and possibly loss of muscle mass (9,10), and low-normal serum bicarbonate levels have been associated with lesser muscle strength and cardiovascular fitness, greater insulin resistance, and faster progression of kidney disease (6,7,13,26). Interventions to treat acidosis, including dietary modifications (27,28), may be most efficacious among older persons, in whom the capacity to excrete the daily acid load from modern Western diets is diminished.

Table 5.   Odds ratio of acidosis by net endogenous acid production within age categories

Net Endogenous Acid Production	Odds Ratio (95% Confidence Interval)		
	Model 1	Model 2	Model 3
Age 20–39 yr (*n*=3299)			
Continuous[a]	1.02 (0.92 to 1.13)	1.06 (0.96 to 1.19)	1.03 (0.92 to 1.15)
<39.3 mEq/d	Reference	Reference	Reference
39.3–52.9 mEq/d	0.97 (0.63 to 1.50)	1.02 (0.66 to 1.58)	0.96 (0.61 to 1.52)
53.0–69.9 mEq/d	1.15 (0.77 to 1.72)	1.28 (0.87 to 1.87)	1.14 (0.80 to 1.62)
≥70.0 mEq/d	1.16 (0.83 to 1.61)	1.31 (0.96 to 1.79)	1.04 (0.84 to 1.56)
P for trend	0.25	0.08	0.28
Age 40–59 yr (*n*=3139)			
Continuous[a]	1.27 (1.10 to 1.48)[b]	1.31 (1.13 to 1.52)[b]	1.22 (1.03 to 1.45)[b]
<39.3 mEq/d	Reference	Reference	Reference
39.3–52.9 mEq/d	1.19 (0.81 to 1.73)	1.25 (0.86 to 1.80)	1.27 (0.88 to 1.82)
53.0–69.9 mEq/d	1.39 (0.98 to 1.95)	1.42 (1.01 to 2.01)[b]	1.38 (0.94 to 2.04)
≥70.0 mEq/d	1.78 (1.07 to 2.94)[b]	1.91 (1.18 to 3.08)[b]	1.65 (0.98 to 2.78)
P for trend	0.02	0.01	0.06
Age≥60 yr (*n*=3343)			
Continuous[a]	1.13 (0.96 to 1.34)	1.12 (0.94 to 1.34)	1.11 (0.93 to 1.33)
<39.3 mEq/d	Reference	Reference	Reference
39.3–52.9 mEq/d	1.08 (0.71 to 1.63)	1.06 (0.69 to 1.63)	1.09 (0.69 to 1.72)
53.0–69.9 mEq/d	1.27 (0.84 to 1.91)	1.24 (0.81 to 1.88)	1.22 (0.81 to 1.84)
≥70.0 mEq/d	1.47 (0.97 to 2.25)	1.46 (0.93 to 2.28)	1.44 (0.91 to 2.29)
P for trend	0.05	0.08	0.09

Model 1: unadjusted. Model 2: adjusted for age, sex, and race/ethnicity. Model 3: model 2 adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated GFR, log-transformed urine albumin–creatinine ratio, serum albumin, and log-transformed C-reactive protein. *P* for interaction by age=0.02.
[a]Per SD higher net endogenous acid production (SD=26.4 mEq/d).
[b]$P<0.05$.

We also observed an unexpected association between serum bicarbonate and age. Contrary to previous literature suggesting that older people have lower serum bicarbonate because of an age-related decline in renal acid excretion (29,30), we found that older age was associated with higher serum bicarbonate. This finding seems partly caused by lower dietary acid intake among older persons, suggesting that NEAP may be equally important as the age-related decline in acid excretion as a determinant of serum bicarbonate. However, even when comparing older and younger Americans within the same dietary acid quartile, bicarbonate levels were higher among the former group. This association persisted after accounting for possible explanatory factors, including diuretic use. One possible explanation is that of a secular trend, in which additional factors that lower serum bicarbonate exist among younger but not older persons. We speculate that such factors could relate to diet or the worsening epidemic of obesity. Over time, these individuals would develop lower serum bicarbonate and worsening low-grade metabolic acidosis if our understanding of the physiology relating aging and renal acid excretory capacity remains correct. Compared with the current cohort of older Americans, however, these younger individuals have lower bicarbonate levels. Therefore, the association of older age with higher serum bicarbonate would not be causal but rather an artifact of the cross-sectional nature of our analyses. It should be noted, however, that the previous reports of declining bicarbonate levels with age also resulted from cross-sectional studies (29,30). In contrast to those data, which come from participants of small clinical and metabolic studies, our findings are from a large, nationally representative dataset.

It remains possible that a more precise measurement of dietary acid intake might account for all of the increase in serum bicarbonate that was seen across age categories. There may also be unaccounted factors related to aging that result in higher bicarbonate levels. Tubular sodium reabsorption is impaired in the elderly (31); resultant mild volume depletion, if present, would increase bicarbonate reabsorption. Subclinical changes in respiratory status, such as mild CO_2 retention, could also occur with aging. If such factors were more common among elderly than middle-aged participants, they could also mask the effect of dietary acid to a greater extent in the elderly. This result could explain why the associations of NEAP with serum bicarbonate and acidosis seemed somewhat stronger among middle-aged adults. Longitudinal population-based data are needed to address this question.

Serum bicarbonate levels among 20–29 year olds did not differ by dietary acid level. This finding suggests that young adults handle even large acid loads within the range of usual dietary intake without perturbing acid–base status. Our data indicate that this capacity already begins to diminish by the fourth decade of life, which is consistent with previous reports (25,30). It should be noted, however, that acid retention may occur even without apparent perturbation of the serum bicarbonate or

extracellular pH. A study of patients with mildly reduced eGFR and normal serum bicarbonate found that changes in urine net acid excretion may be seen even without a change in blood acid–base parameters (32). Therefore, high dietary acid could produce the sequelae of a subclinical acidosis in these younger adults even without observable differences in plasma acid–base parameters.

Intracellular buffering would blunt the effect of NEAP on extracellular buffers, and buffering capacity likely varies directly with the serum bicarbonate (33). Thus, the change in bicarbonate might particularly underestimate the effects of NEAP among older persons, who had the highest bicarbonate levels in our sample. Conversely, people with CKD may have reduced buffering capacity and would experience a relatively greater decrease in serum bicarbonate for any level of NEAP (32).

Several limitations of our analyses should be noted. Calculation of NEAP may have been affected by the limitations of dietary assessment using a single 24-hour recall period. Dietary acid load was not directly measured, and data on renal net acid excretion were not available. However, we used a validated formula to estimate NEAP that was derived using measurements of renal net acid excretion. Although this calculation does not account for differences in the sulfur content of protein or base precursors not associated with potassium, our results were substantively unchanged after replacing total protein with animal-derived sources of protein, which could be a more specific measure of dietary acid precursors. Participants may have underreported dietary protein intake; alternatively, even modestly elevated protein intake may be associated with observable differences in serum bicarbonate levels. We speculated regarding effects of dietary acid on outcomes, such as muscle catabolism and bone disease, based on differences in serum bicarbonate. We believed such speculation was reasonable and should be tested in future studies. We had only a single value of serum bicarbonate available and did not have measurements of serum pH or pCO_2. Sample handling, including processing delays and exposure of serum to air, may affect bicarbonate levels (34–36). We could not account for this possibility, which would likely introduce nondifferential misclassification and bias our results to the null. Finally, we cannot rule out residual confounding caused by undetected kidney disease among older individuals, because eGFR is an imprecise measurement of kidney function. We addressed this possibility by restricting our analyses to participants with eGFR≥60 ml/min per 1.73 m^2, which did not change the results. In addition, if older age was simply a marker for reduced kidney function, our findings would still be clinically relevant, because older persons constitute an expanding proportion of the population with kidney disease.

In summary, greater dietary acid is associated with lower serum bicarbonate and a higher prevalence of acidosis in the general US population, and this association is greater in older than younger Americans. In contrast to previous reports, serum bicarbonate levels are highest among older Americans. Longitudinal studies are needed to prospectively determine the association of the dietary acid load with acid–base status in free-living individuals. In the interim, clinicians should be aware that older persons may be most susceptible to the potential deleterious effects of diets high in acid-forming potential.

Disclosures
None.

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Supplementary Tables and Figures

Table S1. Association of Net Endogenous Acid Production with Serum Bicarbonate in Participants with Estimated Glomerular Filtration Rate ≥ 60 mL/min/1.73m^2

Table S2. Odds Ratio of Acidosis by Net Endogenous Acid Production in Participants with Estimated Glomerular Filtration Rate ≥ 60 mL/min/1.73m^2

Table S3. Odds Ratio of Acidosis (defined as serum bicarbonate <22 mEq/L) by Net Endogenous Acid Production

Table S4. Association of Animal Protein-Derived Endogenous Acid Production with Serum Bicarbonate

Table S5. Association of Animal Protein-Derived Endogenous Acid Production with Serum Bicarbonate within Age Categories

Table S6. Association of Dietary Protein with Serum Bicarbonate

Table S7. Association of Dietary Potassium with Serum Bicarbonate

Table S8. Association of Net Endogenous Acid Production with Serum Bicarbonate After Adjustment for Total Body Fat in 9,499 Participants of NHANES 1999-2004 Who Underwent Dual Energy X-ray Absorptiometry

Table S9. Association of Net Endogenous Acid Production with Serum Bicarbonate within Age Categories After Adjustment for Total Body Fat in 9,499 Participants of NHANES 1999-2004 Who Underwent Dual Energy X-ray Absorptiometry

Figure S1. Association of estimated animal protein-derived endogenous acid production (ADEAP, mEq/day) with age in 9,781 participants of NHANES 1999-2004.

Table S1. Association of Net Endogenous Acid Production with Serum Bicarbonate in 8,909 Participants with Estimated Glomerular Filtration Rate ≥ 60 mL/min/1.73m²

	Coefficient (95% CI)		
	Model 1	Model 2	Model 3
NEAP			
Continuous*	**-0.22 (-0.28 to -0.16)**	**-0.20 (-0.27 to -0.14)**	**-0.15 (-0.21 to -0.10)**
< 39.3 mEq/d	[Ref]	[Ref]	[Ref]
39.3 – 52.9 mEq/d	-0.14 (-0.29 to 0.02)	**-0.17 (-0.32 to -0.02)**	-0.13 (-0.28 to 0.01)
53.0 – 69.9 mEq/d	**-0.39 (-0.59 to -0.19)**	**-0.38 (-0.58 to -0.18)**	**-0.29 (-0.47 to -0.10)**
≥ 70.0 mEq/d	**-0.60 (-0.77 to -0.43)**	**-0.55 (-0.72 to -0.38)**	**-0.43 (-0.57 to -0.28)**
P for trend	<0.001	<0.001	<0.001

Abbreviations: CI, confidence interval. NEAP, Net Endogenous Acid Production, **Bold** values indicate p<0.05.
Model 1: unadjusted
Model 2: adjusted for age, sex, and race/ethnicity
Model 3: Model 2 + adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated glomerular filtration rate, log-transformed urine albumin-creatinine ratio, serum albumin and log-transformed C-reactive protein.
*Per standard deviation higher NEAP (SD = 26.5 mEq/d)

Table S2. Odds Ratio of Acidosis by Net Endogenous Acid Production in 8,909 Participants with Estimated Glomerular Filtration Rate ≥ 60 mL/min/1.73m^2

	OR (95% CI)		
	Model 1	Model 2	Model 3
NEAP			
Continuous*	**1.18 (1.08 – 1.29)**	**1.15 (1.05 – 1.27)**	1.10 (1.00 – 1.22)
< 39.3 mEq/d	[Ref]	[Ref]	[Ref]
39.3 – 52.9 mEq/d	1.14 (0.93 – 1.40)	1.16 (0.95 – 1.42)	1.13 (0.92 – 1.40)
53.0 – 69.9 mEq/d	**1.46 (1.14 – 1.87)**	**1.43 (1.11 – 1.83)**	**1.30 (1.02 – 1.66)**
≥ 70.0 mEq/d	**1.66 (1.29 – 2.13)**	**1.54 (1.19 – 2.01)**	**1.36 (1.05 – 1.75)**
P for trend	<0.001	0.002	0.02

Abbreviations: OR, odds ratio; CI, confidence interval. **Bold** values indicate p<0.05.

Acidosis defined as serum bicarbonate <23 mEq/L.

Model 1: unadjusted

Model 2: adjusted for age, sex, and race/ethnicity

Model 3: Model 2 + adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated glomerular filtration rate, log-transformed urine albumin-creatinine ratio, serum albumin and log-transformed C-reactive protein.

*Per standard deviation higher NEAP (SD = 26.5 mEq/d)

Table S3a. Odds Ratio of Acidosis by Net Endogenous Acid Production in 9,781 Participants of NHANES 1999-2004[†]

	OR (95% CI)		
	Model 1	Model 2	Model 3
NEAP			
Continuous*	**1.19 (1.07 – 1.32)**	**1.18 (1.05 – 1.34)**	**1.15 (1.01 – 1.31)**
< 39.3 mEq/d	[Ref]	[Ref]	[Ref]
39.3 – 52.9 mEq/d	0.92 (0.68 – 1.29)	0.95 (0.69 – 1.29)	0.95 (0.70 – 1.30)
53.0 – 69.9 mEq/d	1.32 (0.95 – 1.82)	1.31 (0.94 – 1.81)	1.24 (0.90 – 1.70)
≥ 70.0 mEq/d	**1.45 (1.10 – 1.90)**	**1.39 (1.02 – 1.89)**	1.28 (0.94 – 1.74)
P for trend	0.002	0.02	0.07

Table S3b. Odds Ratio of Acidosis by Net Endogenous Acid Production in 8,909 Participants with estimated Glomerular Filtration Rate ≥ 60 mL/min/1.73m^{2†}

	OR (95% CI)		
	Model 1	Model 2	Model 3
NEAP			
Continuous*	**1.22 (1.09 – 1.37)**	**1.20 (1.05 – 1.37)**	**1.17 (1.02 – 1.34)**
< 39.3 mEq/d	[Ref]	[Ref]	[Ref]
39.3 – 52.9 mEq/d	0.99 (0.70 – 1.41)	1.01 (0.70 – 1.46)	1.00 (0.69 – 1.43)
53.0 – 69.9 mEq/d	**1.49 (1.03 – 2.14)**	**1.46 (1.01 – 2.12)**	1.36 (0.95 – 1.94)
≥ 70.0 mEq/d	**1.61 (1.18 – 2.19)**	**1.50 (1.06 – 2.13)**	1.37 (0.98 – 1.92)
P for trend	0.001	0.01	0.03

Abbreviations: OR, odds ratio; CI, confidence interval. **Bold** values indicate p<0.05.

[†]**Acidosis defined as serum bicarbonate <22 mEq/L.**

Model 1: unadjusted

Model 2: adjusted for age, sex, and race/ethnicity

Model 3: Model 2 + adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated glomerular filtration rate, log-transformed urine albumin-creatinine ratio, serum albumin and log-transformed C-reactive protein.

*Per standard deviation higher NEAP (SD for Table S3a = 26.4 mEq/d; Table S3b = 26.5 mEq/d)

Table S4. Association of Animal Protein-Derived Endogenous Acid Production with Serum Bicarbonate in 9,781 Participants of NHANES 1999-2004

	Coefficient (95% CI)		
	Model 1	Model 2	Model 3
ADEAP			
Continuous*	**-0.12 (-0.20 to -0.04)**	**-0.14 (-0.23 to -0.06)**	**-0.11 (-0.18 to -0.03)**
< 1.6 mEq/d	[Ref]	[Ref]	[Ref]
1.6 – 15.5 mEq/d	-0.03 (-0.21 to 0.26)	-0.03 (-0.25 to 0.19)	-0.05 (-0.26 to 0.19)
15.6 – 32.9 mEq/d	-0.02 (-0.26 to 0.22)	-0.09 (-0.32 to 0.14)	-0.09 (-0.30 to 0.12)
> 32.9 mEq/d	**-0.31 (-0.56 to -0.07)**	**-0.38 (-0.62 to -0.14)**	**-0.28 (-0.50 to -0.05)**
P for trend	0.01	0.01	0.02

Abbreviation: CI, confidence interval; ADEAP, Animal Protein Derived-Endogenous Acid Production. **Bold** values indicate p<0.05.
Model 1: unadjusted
Model 2: adjusted for age, sex, and race/ethnicity
Model 3: Model 2 + adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated glomerular filtration rate, log-transformed urine albumin-creatinine ratio, serum albumin and log-transformed C-reactive protein.
*Per standard deviation higher ADEAP (SD = 26.2 mEq/d)

Table S5. Association of Animal Protein-Derived Endogenous Acid Production with Serum Bicarbonate within Age Categories

Age 20 to 39 years	Coefficient (95% CI)		
	Model 1	Model 2	Model 3
ADEAP (n = 3299)			
Continuous*	0.02 (-0.10 to 0.13)	-0.06 (-0.18 to 0.05)	-0.04 (-0.14 to 0.07)
< 1.6 mEq/d	[Ref]	[Ref]	[Ref]
1.6 – 15.5 mEq/d	0.04 (-0.34 to 0.42)	-0.03 (-0.39 to 0.32)	-0.03 (-0.35 to 0.28)
15.6 – 32.9 mEq/d	0.13 (-0.24 to 0.49)	-0.05 (-0.39 to 0.28)	-0.02 (-0.31 to 0.27)
> 32.9 mEq/d	-0.05 (-0.39 to 0.29)	-0.27 (-0.60 to 0.05)	-0.17 (-0.46 to 0.11)
P for trend	0.83	0.12	0.24

Age 40 to 59 years	Coefficient (95% CI)		
	Model 1	Model 2	Model 3
ADEAP (n = 3139)			
Continuous*	**-0.19 (-0.30 to -0.09)**	**-0.25 (-0.35 to -0.14)**	**-0.20 (-0.31 to -0.08)**
< 1.6 mEq/d	[Ref]	[Ref]	[Ref]
1.6 – 15.5 mEq/d	0.05 (-0.30 to 0.39)	0.01 (-0.35 to 0.31)	-0.001 (-0.33 to 0.33)
15.6 – 32.9 mEq/d	0.02 (-0.32 to 0.36)	-0.08 (-0.41 to 0.24)	-0.05 (-0.37 to 0.27)
> 32.9 mEq/d	**-0.37 (-0.73 to -0.01)**	-0.52 (-0.87 to 0.17)	**-0.41 (-0.77 to -0.05)**
P for trend	0.04	0.01	0.02

Age ≥ 60 years	Coefficient (95% CI)		
	Model 1	Model 2	Model 3
ADEAP (n = 3343)			
Continuous*	**-0.15 (-0.28 to -0.01)**	-0.14 (-0.28 to 0.003)	-0.10 (-0.24 to 0.03)
< 1.6 mEq/d	[Ref]	[Ref]	[Ref]
1.6 – 15.5 mEq/d	-0.08 (-0.33 to 0.18)	-0.07 (-0.32 to 0.18)	-0.09 (-0.32 to 0.13)
15.6 – 32.9 mEq/d	-0.22 (-0.46 to 0.03)	-0.19 (-0.44 to 0.06)	-0.19 (-0.42 to 0.03)
> 32.9 mEq/d	**-0.35 (-0.66 to -0.04)**	-0.33 (-0.65 to 0.001)	-0.22 (-0.54 to 0.09)
P for trend	0.02	0.04	0.10

Abbreviation: CI, confidence interval; ADEAP, Animal Protein Derived Endogenous Acid Production. **Bold** values indicate p<0.05.
P for interaction by age = 0.01
Model 1: unadjusted
Model 2: adjusted for age, sex, and race/ethnicity

Model 3: Model 2 + adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated glomerular filtration rate, log-transformed urine albumin-creatinine ratio, serum albumin and log-transformed C-reactive protein.
*Per standard deviation higher ADEAP (SD = 26.2 mEq/d)

Table S6. Association of Dietary Protein with Serum Bicarbonate in 9,781 Participants of NHANES 1999-2004

	Coefficient (95% CI)		
	Model 1	Model 2	Model 3
Dietary Protein			
Continuous*	-0.02 (-0.12 to 0.08)	-0.09 (-0.20 to 0.02)	-0.10 (-0.22 to 0.01)
≤54.5 g/day	[Ref]	[Ref]	[Ref]
54.5 – 76.6 g/day	-0.04 (-0.19 to 0.12)	-0.07 (-0.22 to 0.09)	-0.11 (-0.27 to 0.05)
76.7 – 103.7 g/day	-0.03 (-0.22 to 0.16)	-0.12 (-0.31 to 0.06)	-0.15 (-0.34 to 0.03)
≥103.7 g/day	-0.02 (-0.26 to 0.21)	-0.18 (-0.44 to 0.07)	**-0.23 (-0.50 to -0.04)**
P for trend	0.83	0.12	0.09

Abbreviation: CI, confidence interval, **Bold** values indicate p<0.05.
Model 1: adjusted for dietary energy
Model 2: adjusted for age, sex, race/ethnicity and dietary energy
Model 3: Model 2 + adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated glomerular filtration rate, log-transformed urine albumin-creatinine ratio, serum albumin and log-transformed C-reactive protein.
*Per standard deviation higher dietary protein (SD = 42.4 g/day)

Table S7. Association of Dietary Potassium with Serum Bicarbonate in 9,781 Participants of NHANES 1999-2004

	Coefficient (95% CI)		
	Model 1	Model 2	Model 3
Dietary Potassium			
Continuous*	**0.32 (0.23 to 0.42)**	**0.16 (0.08 to 0.25)**	**0.14 (0.05 to 0.24)**
<47.4 mEq/d	[Ref]	[Ref]	[Ref]
47.4 – 66.7 mEq/d	**0.31 (0.16 to 0.46)**	**0.19 (0.04 to 0.34)**	0.09 (-0.05 to 0.22)
66.8 – 89.4 mEq/d	**0.46 (0.29 to 0.62)**	**0.29 (0.13 to 0.45)**	**0.17 (0.01 to 0.33)**
≥ 89.5 mEq/d	**0.78 (0.51 to 1.04)**	**0.52 (0.24 to 0.79)**	**0.30 (0.05 to 0.55)**
P for trend	<0.001	0.001	0.01

Abbreviation: CI, confidence interval, **Bold** values indicate p<0.05.
Model 1: adjusted for dietary energy
Model 2: adjusted for age, sex, race/ethnicity and dietary energy
Model 3: Model 2 + adjusted for body mass index, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated glomerular filtration rate, log-transformed urine albumin-creatinine ratio, serum albumin and log-transformed C-reactive protein.
*Per standard deviation higher dietary potassium (SD = 32.9 mEq/d)

Table S8. Association of Net Endogenous Acid Production with Serum Bicarbonate After Adjustment for Total Body Fat in 9,499 Participants of NHANES 1999-2004 Who Underwent Dual Energy X-ray Absorptiometry

	Coefficient (95% CI)		
	Model 1	Model 2	Model 3
NEAP			
Continuous*	**-0.15 (-0.21 to -0.10)**	**-0.15 (-0.21 to -0.10)**	**-0.15 (-0.21 to -0.09)**
< 39.3 mEq/d	[Ref]	[Ref]	[Ref]
39.3 – 52.9 mEq/d	-0.10 (-0.25 to 0.04)	-0.12 (-0.27 to 0.02)	-0.11 (-0.26 to 0.04)
53.0 – 69.9 mEq/d	**-0.25 (-0.44 to -0.07)**	**-0.26 (-0.45 to -0.08)**	**-0.25 (-0.44 to -0.06)**
≥ 70.0 mEq/d	**-0.42 (-0.57 to -0.28)**	**-0.43 (-0.58 to -0.28)**	**-0.42 (-0.56 to -0.27)**
P for trend	<0.001	<0.001	<0.001

Abbreviation: CI, confidence interval. NEAP, Net Endogenous Acid Production,
Bold values indicate p<0.05.
Model 1: adjusted for age, sex, race/ethnicity, **body mass index**, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated glomerular filtration rate, log-transformed urine albumin-creatinine ratio, serum albumin and log-transformed C-reactive protein.
Model 2: adjusted for age, sex, race/ethnicity, **% total body fat**, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated glomerular filtration rate, log-transformed urine albumin-creatinine ratio, serum albumin and log-transformed C-reactive protein.
Model 3: Model 1 + adjusted for **% total body fat**
*Per standard deviation higher NEAP (SD =26.5 mEq/d)

Table S9. Association of Net Endogenous Acid Production with Serum Bicarbonate within Age Categories After Adjustment for Total Body Fat in 9,499 Participants of NHANES 1999-2004 Who Underwent Dual Energy X-ray Absorptiometry

	Coefficient (95% CI)		
	20 to 39 years (n=3211)	40 to 59 years (n=3060)	≥ 60 years (n=3228)
NEAP			
Continuous*	-0.07 (-0.15 to 0.02)	**-0.28 (-0.39 to -0.18)**	**-0.17 (-0.30 to -0.04)**
< 39.3 mEq/d	[Ref]	[Ref]	[Ref]
39.3 – 52.9 mEq/d	-0.10 (-0.41 to 0.20)	-0.13 (-0.42 to 0.16)	-0.13 (-0.42 to 0.15)
53.0 – 69.9 mEq/d	-0.14 (-0.42 to 0.14)	**-0.38 (-0.66 to -0.09**)	-0.18 (-0.44 to 0.07)
≥ 70.0 mEq/d	-0.24 (-0.49 to 0.001)	**-0.67 (-0.99 to -0.35)**	**-0.50 (-0.85 to -0.16)**
P for trend	0.05	<0.001	0.01

Abbreviation: CI, confidence interval. NEAP, Net Endogenous Acid Production, **Bold** values indicate p<0.05.

P for interaction by age = 0.004

Models adjusted for age, sex, race/ethnicity, **% total body fat**, poverty, education, smoking status, diuretic use, diagnosis of diabetes mellitus, hypertension, cardiovascular disease, estimated glomerular filtration rate, log-transformed urine albumin-creatinine ratio, serum albumin and log-transformed C-reactive protein.

*Per standard deviation higher NEAP (SD = 26.5 mEq/d)

Figure S1. Association of estimated animal protein-derived endogenous acid production (ADEAP, mEq/day) with age in 9,781 participants of NHANES 1999-2004. Each bar represents an ADEAP quartile. Error bars represent standard errors.