Nutritional influences on bone mineral density: a cross-sectional study in premenopausal women[1-3]

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ABSTRACT   The association between current and past dietary intake and bone mineral density (BMD) was investigated in 994 healthy premenopausal women aged 45–49 y. BMD was measured with dual-energy X-ray absorptiometry (DXA). Dietary intake was assessed with a food-frequency questionnaire (FFQ). Energy-adjusted nutrient intakes were grouped into quartiles and mean BMD at the lumbar spine (LS), femoral neck (FN), femoral trochanter (FT), and femoral Wards (FW) were calculated. With higher intakes of zinc, magnesium, potassium, and fiber, LS BMD was significantly higher ($P < 0.05$–0.006), and a significant difference in LS BMD was also found between the lowest and highest quartiles for these nutrients and vitamin C intake ($P < 0.05$–0.01). These results remained significant after adjustment for important confounding factors. LS BMD and FT BMD were lower in women reporting a low intake of milk and fruit in early adulthood than in women with a medium or high intake ($P < 0.01$). High, long-term intakes of these nutrients may be important to bone health, possibly because of their beneficial effect on acid-base balance. *Am J Clin Nutr* 1997;65:1831–9.

KEY WORDS   Bone mineral density, food-frequency questionnaire, energy-adjusted nutrient intakes, confounding variables, potassium, magnesium, vitamin C, acid-base balance, premenopausal women

INTRODUCTION

The influence of nutrient intake on bone mineral density (BMD) is still largely undefined. There is some evidence to suggest that calcium intake may be important during skeletal growth and peak bone mass development (1–3), and calcium supplementation has been shown to reduce bone loss in women who are ≥ 5 y postmenopausal (4, 5). However, few studies have examined the effect of other nutrients on bone mass. Excessive intakes of protein, sodium, and caffeine are known to affect calcium metabolism (6–8) but have not been shown to affect BMD adversely. The effect of intakes of other nutrients such as fiber, minerals, and antioxidant vitamins have received little attention. Studies reported to date on dietary influences on bone health have tended to use older, less reliable BMD measuring techniques (9–11) and 24-h recall as a measure of dietary intake (9, 10, 12), and have inadequately adjusted for total energy intake (13–15) or other important confounding variables such as weight, height, smoking, socioeconomic status, and physical activity level (PAL) (15, 16).

The purpose of this study was to investigate the association between dietary intake and BMD with maximum accuracy and reliability. This was achieved by using the most up-to-date technique for bone mineral measurement, dual-energy X-ray absorptiometry (DXA); by using a purpose-designed food-frequency questionnaire (FFQ) for assessing usual dietary intake in a population; and by carefully controlling for important confounding variables, including total energy intake.

SUBJECTS AND METHODS

Selection of subjects

Women for this study were a subset of those who had participated in the Osteoporosis Screening Program (Aberdeen, Scotland) during 1990–1992, details of which were reported previously (17). Briefly, women were drawn at random from the Community Health Index and invited to receive a BMD scan. The overall response rate to the screening program was 75%; with one reminder letter (18). Women who had taken any medication, who suffered from any condition likely to affect their bone metabolism, or who were classified as perimenopausal (absence of regular periods in the previous 6 mo) were excluded from the study. From a total of 3000 women scanned, 1230 were eligible for the study. The main reasons for exclusion were uncertainty of menstrual status and peri- or postmenopausal state.

The study was approved by the Joint Ethical Committee of Grampian Health Board and the University of Aberdeen.

Study design

The study was designed such that dietary data were collected after women had received their BMD scan. To control as much as possible for the seasonal variation that may occur with such a study design, women who had been scanned during the

autumn and winter seasons (between October and March; winter group) or spring and summer seasons (between April and September; summer group) completed their FFQs during these seasons.

Women were divided into "time groups" according to the length of time between the BMD scan and dietary assessment: group 1, 20–24 mo; group 2, 13–16 mo; group 3, 10–12 mo; and group 4, 3–9 mo. BMD and nutrient intakes for the winter and summer groups and the four time groups were then examined to see whether groups should be analyzed separately or together. No significant differences were found in nutrient intake or BMD between either seasonal or time groups. Furthermore, the effect of nutrient intakes on BMD for these groups were investigated but no significant differences between the slope or intercept of the regression lines were found. All groups were therefore analyzed together.

Anthropometric and BMD measurements

The weight of each woman (in light clothing and without shoes) was recorded by using a set of balance scales (Seca, Hamburg, Germany) calibrated to 0.05 kg, and height was measured with a stadiometer (Holtain Ltd, Crymych, United Kingdom). BMD was assessed with DXA (model XR-26; Norland, Madison, WI) at the lumbar spine (lumbar vertebrae 2–4; LS) and left femur [femoral neck (FN), greater trochanter (FT), and Ward's area (FW)] by an experienced radiographer. The DXA machine was fully calibrated and the standard technique for measurement was adhered to strictly (19). The CV of this technique in our laboratory was 0.9% for LS, 2.7% for FN, 3.2% for FT, and 4.5% for FW.

Usual dietary intake

Usual dietary intake (over the previous 12 mo) was assessed with an FFQ that had been designed and validated previously against 7-d weighed-food records from the study population (20). The FFQ, instruction sheet, and covering letter were sent to each woman and if the questionnaire had not been returned in the prepaid envelope within 3 wk, a reminder letter was sent. A final response rate of 82% (n = 1008) was achieved. Information on anthropometric data, BMD measurements, and smoking history was available on all women who did not return their FFQs.

The questionnaires were coded and analyzed by using the Rowett Research Institute Nutritional Analysis Program (RONA) based on McCance and Widdowson's food-composition tables and supplements (21). Consumption of vitamin and mineral preparations was included in the assessment of mean daily nutrient intakes. Fourteen women who were determined to have answered the FFQ incorrectly (by SAN) were excluded (energy intake < 4.0 MJ, n = 13; > 21 MJ, n = 1). Other women with low energy intakes but who apparently completed the FFQ correctly were retained so as not to unduly bias the data. Therefore, results are given for 994 women.

Past dietary intake

For assessment of past dietary intake, two age categories were chosen as important stages in skeletal growth: childhood (≤ 12 y) and early adulthood (20–30 y). Women were asked how much milk they had consumed and the frequency of consumption of fruit and vegetables (excluding potatoes). Past

dietary intake was determined for the purpose of categorizing women as low, medium, or high consumers of the key foods. Categorization of intakes was as follows: milk—low (< 284 mL/d), medium (284–568 mL/d), and high (> 568 mL/d); fruit and vegetables—low (1–4 times/d ≤ 2 d/wk), medium (1–4 times/d 3–4 d/wk), and high (1–4 times/d ≥ 5 d/wk).

Smoking, social status, and physical activity level

A questionnaire was used to collect information on present smoking habits (number of cigarettes smoked per day and duration of smoking) and educational attainment, as a marker of socioeconomic status. PAL was assessed from the womens' responses to questions concerning work and leisure activities: examples of light, moderate, and active pursuits were provided and women estimated the number of hours per day spent in each category of activity and the number of hours they slept. PALs were calculated by using the equations of James and Schofield (22): light, 1.56; moderate, 1.64; and active, 1.82.

Statistical analysis

The Statistical Program for Social Sciences (SPSSPC+ 1988, SPSS Inc, Chicago) was used for data analysis. Because the intakes of most nutrients are highly correlated with total energy intake, the study of their respective relation with disease requires adjustment for total energy intake. Energy-adjusted intakes were computed by using the residual method of Willett (23).

Descriptive statistics (means ± SDs, medians, and ranges) were determined for all variables; when the distributions were found to be skewed, log transformations (ln) were used to normalize the data before parametric analysis. Pearson correlation and partial correlation coefficients—adjusted for age, weight, height, physical activity, socioeconomic status, and smoking—were calculated for each nutrient at each BMD site. For multiple comparisons with correlations and partial correlations, a Bonferroni correction was made and although P values at the 5% level are shown, they may be of little biological significance. Pearson correlations between the different nutrients were also determined to assess whether one nutrient may have acted as a surrogate for another.

To determine whether any of the nutrients were independent predictors of bone mass at the LS and proximal femur sites, age, weight, height, smoking, social status, and physical activity were entered into a stepwise-multiple-regression model together with intakes of the important macro- and micronutrients (protein, fiber, calcium, potassium, phosphorus, sodium, magnesium, zinc, vitamin C, and vitamin D).

In addition to treating nutrients as continuous variables, quartiles of intake were also analyzed because this is a particularly appropriate way to analyze nutrient intakes from FFQs. Intakes were grouped into quartiles and the mean BMD at each site was calculated. Differences in BMD between quartiles of intake were assessed by using a multiple-range test [one-way analysis of variance (ANOVA) with a Scheffé range test], which is based on the 95% CI limits of each estimate and is more informative than the simple t test because it identifies which of the quartiles differ from each other. Analysis of covariance (ANCOVA) was also used to assess differences after adjustment for the confounding variables age, weight, height, PAL, smoking, and socioeconomic status. The associ-

ation between past dietary intake and BMD was undertaken by using ANOVA. To examine whether past intake of calcium was associated with current energy, calcium, and milk intakes, the chi-square test was used.

RESULTS

Responders and nonresponders

No differences were found for age, weight, height, and BMD between responders and nonresponders. Nonresponders smoked for more years than did the responders ($P < 0.01$).

Descriptive statistics

Anthropometric data and BMD measurements are shown in **Table 1**. Values had an approximately normal distribution. Sixty-eight percent of women were within the accepted limits of ideal body weight [a body mass index (BMI; in kg/m^2) of 20–25], with 6% being underweight and 26% overweight.

The mean daily intake of nutrients (including vitamin and mineral supplements) are shown in **Table 2**. Intakes of energy and fat were remarkably similar to the estimated average requirement (EAR) and reference nutrient intake (RNI) for the UK population aged 19–50 y (24). Intakes of other nutrients including protein, phosphorus, calcium, sodium, fiber, and vitamin C were higher than the RNI. Ninety percent of women had calcium intakes greater than the RNI (700 mg/d), but when analyzed differently, only 50% of the women had intakes greater than the USA recommended dietary allowance (RDA) of 1000 mg (24). Alcohol was consumed by 77% of the women (mean intake: 7 g). The mean energy equivalent (energy intake/calculated basal metabolic rate) was 1.42.

Correlations between nondietary factors and BMD

Correlations among age, weight, height, PAL, and BMD are shown in **Table 3**. Weight and height were significantly correlated with the four BMD sites, although coefficients were higher for weight. No significant correlations were found for age or PAL. LS BMD was significantly lower ($P < 0.02$) in smokers (1.04 ± 0.16 g/cm^2; $n = 224$) than in nonsmokers (1.07 ± 0.16 g/cm^2; $n = 770$). No significant correlations were found between BMD and socioeconomic status.

TABLE 1
Anthropometric data and bone mineral density (BMD) measurements of 994 women[1]

	$\bar{x} \pm$ SD	Median	Range
Age (y)	47.1 ± 1.43	47.0	44.0–50.0
Weight (kg)	64.1 ± 11.2	62.0	37.0–124.0
Height (m)	1.614 ± 0.058	1.616	1.159–1.784
BMI (kg/m^2)	24.6 ± 4.1	23.8	15.8–44.0
LS BMD (g/cm^2)	1.062 ± 0.162	1.049	0.622–1.958
FN BMD (g/cm^2)	0.886 ± 0.122	0.876	0.588–1.600
FT BMD (g/cm^2)	0.718 ± 0.120	0.694	0.425–1.878
FW BMD (g/cm^2)	0.841 ± 0.157	0.829	0.467–1.467

[1] LS, lumbar spine (vertebrae L2–4); FN, femoral neck; FT, femoral trochanter; FW, femoral Ward's area.

TABLE 2
Mean daily nutrient intake of 994 women

Nutrient	$\bar{x} \pm$ SD	Median	Range	RNI or EAR[1]
Energy (kJ)	8130 ± 2290	7820	4870–16820	8.1
EI:BMR[2]	1.43 ± 0.41	1.37	1.01–3.00	—
Protein (g)	81 ± 22	78	20–231	45
Fat (g)	74 ± 29	70	18–206	73
Carbohydrate (g)	243 ± 72	236	75–564	193
Fiber (g)	15 ± 6	15	4–47	12
Alcohol (g)	7 ± 8	5	0–120	—
Calcium (mg)	1060 ± 344	1007	174–3066	700
Sodium (mg)	2630 ± 862	2510	559–7535	1600
Potassium (mg)	3320 ± 807	3238	1475–6897	3500
Phosphorus (mg)	1467 ± 400	1423	539–3390	541
Iron (mg)		12.1		
	13.0 ± 4.8		2.3–36.3	11.4
Magnesium (mg)	311 ± 85	301	109–638	270
Zinc (mg)	10.0 ± 2.9	7.0	2.4–23.7	—
Vitamin D (μg)	3.5 ± 2.3	3.03	0.2–29.5	8–10
Vitamin C (mg)	126 ± 96	106	16–1164	40

[1] RNI, reference nutrient intake; EAR, estimated average requirement (for energy only).

[2] Energy intake/calculated basal metabolic rate.

Correlations and partial correlations between nutrient intake and BMD

Correlation and partial correlation coefficients between energy-adjusted nutrient intakes and BMD are shown in **Table 4**. The simple correlation coefficients show many significant relations, but after adjustment for age, weight, height, physical activity, smoking, and social status only potassium intake remained significantly correlated with BMD at all sites. Magnesium intake was significant at the LS as were vitamin C and alcohol intake. Correlation coefficients between nutrients showed highly significant relations between magnesium and potassium, magnesium and zinc, magnesium and fiber, and potassium and zinc ($r > 0.80$, $P < 0.001$).

Nutrients as independent predictors of bone mass

Lumbar spine

At the LS, alcohol and magnesium intakes were independent predictors of bone mass after weight and height with the equation as follows:

$$LS \quad BMD = 0.209 + 0.0044 \text{ wt (kg)}$$

$$+ 0.3122 \text{ ht (m)} + 0.0014 \text{ alcohol intake (g)}$$

$$+ 0.00018 \text{ magnesium intake (mg)} \quad (1)$$

TABLE 3
Pearson correlation coefficients between nondietary factors and bone mineral density (BMD)[1]

	Age	Weight	Height	Physical activity
LS BMD	−0.04	0.36[2]	0.26[2]	0.04
FN BMD	−0.02	0.41[2]	0.25[2]	0.01
FT BMD	0.03	0.39[2]	0.20[2]	−0.03
FW BMD	−0.05	0.49[2]	0.21[2]	0.01

[1] LS, lumbar spine (vertebrae L2–4); FN, femoral neck; FT, femoral trochanter; FW, femoral Ward's area.

[2] $P < 0.001$.

TABLE 4
Pearson correlation coefficients and partial correlation coefficients between energy-adjusted intakes of nutrients and bone mineral density (BMD)[1]

	Lumbar spine	Femoral neck	Femoral trochanter	Femoral Ward's area
Energy (kJ)	0.02	0.03	0.04	0.01
Protein (g)	0.03	0.02	0.04	0.02
Calcium (mg)	0.06[2] [0.03]	0.03	0.04	0.01
Fiber (g)	0.08[3] [0.03]	0.04	0.04	0.03
Potassium (mg)	0.11[4] [0.07][2]	0.09[3] [0.06][2]	0.09[4] [0.06][2]	0.08[3] [0.06][2]
Magnesium (mg)	0.10[4] [0.06][2]	0.05	0.04	0.04
Zinc (mg)	0.05	0.03	0.05	0.03
Vitamin C (mg)	0.10[4] [0.07][2]	0.03	0.05	0.05
Alcohol (g)	0.11[4] [0.08][3]	0.02	0.05	0.01

[1] Partial correlation coefficients (adjusted for age, weight, height, physical activity, smoking, and social status) in brackets.
[2] $P < 0.05$.
[3] $P < 0.01$.
[4] $P < 0.001$.

where adjusted $R2 = 0.14$, $n = 994$, and $P < 0.001$. Partial correlation coefficients were significant at $P < 0.0001$, $P < 0.001$, $P < 0.017$, and $P < 0.028$, respectively. Vitamin C and potassium just failed to be incorporated into the equation with partial correlation coefficients of $P < 0.055$ and $P < 0.061$, respectively.

Proximal femur

For FN BMD and FW BMD there were no dietary predictors of bone mass. For FT BMD, potassium and vitamin C were almost significant ($P < 0.067$ and $P < 0.068$, respectively).

Analysis by quartiles of intake

LS BMD increased significantly across the quartiles of calcium intake ($P < 0.05$) and a significant difference was found between the lowest and highest quartile of calcium intake ($P < 0.02$, ANOVA); however, the increase was not significant by ANCOVA. BMD increased significantly at all four sites across the quartiles for zinc intake ($P < 0.05$ to < 0.006), and there was a significant difference between the lowest and highest quartiles at the LS, FN, and FT sites ($P < 0.01$); the difference remained significant after ANCOVA at the LS ($P < 0.05$). Across the quartiles for magnesium and potassium intakes, BMD at the LS, FN, and FT sites also increased significantly ($P < 0.01–0.004$), and there was a significant difference in LS BMD between the lowest and highest quartiles for magnesium intake ($P < 0.01$; **Figures 1** and **2**), remaining significant after appropriate adjustment ($P < 0.02$). There were significant diffferences in potassium intake between the lowest and highest quartiles at the LS ($P < 0.007$), FN ($P < 0.009$), and FT ($P < 0.009$)sites, but these differences were no longer significant after adjustment for the confounding variables ($P < 0.06$; **Figures 3** and **4**).

The trend in BMD across the quartiles of vitamin C intake was nonlinear. BMD was higher in the third quartile and was significantly different from the lowest quartile at all four sites even after adjustment for the confounding factors ($P < 0.02–0.002$; **Figures 5** and **6**). The mean LS BMD also increased significantly with increasing fiber intakes ($P < 0.01$), and a significant difference was found between the lowest and highest quartiles ($P < 0.04$), which was almost significant after appropriate adjustment ($P < 0.08$).

Because of the strong correlations among zinc, potassium, magnesium, and fiber, analysis by quartile of intake was further analyzed with each of these nutrients entered as covariates. For magnesium intake, associations did not change significantly after adjustment for potassium, zinc, or fiber. However, for zinc, potassium, and fiber, adjustment for magnesium intake slightly weakened the quartile of intake relation.

The mean LS BMD increased significantly across the quartiles of alcohol intake ($P < 0.01$), and a significant difference was found between the lowest and highest quartiles, which remained significant after appropriate adjustment ($P < 0.04$). No significant differences were found between quartiles of intake and any of the other nutrients investigated.

Association between past dietary intake and BMD

There were significant differences in LS BMD between women who reported a low intake of milk in their childhood and early adulthood and those who reported a medium or high intake ($P < 0.05$ and < 0.01, respectively). BMD also differed significantly at the FN and FT sites in women who had reported
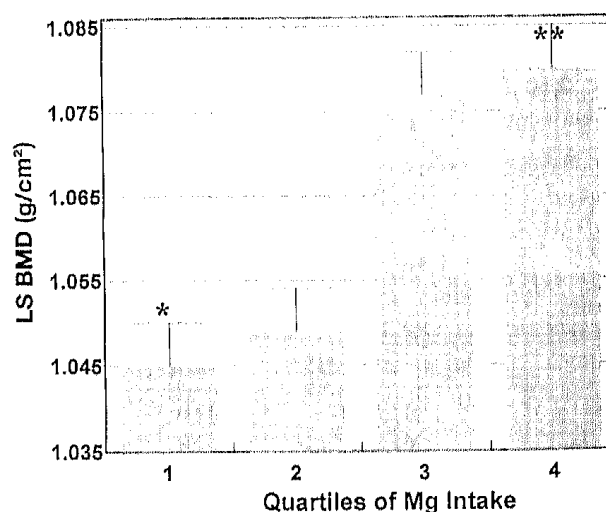

FIGURE 1. Mean ($\pm$ SEM) increase in lumbar spine bone mineral density (LS BMD) with quartiles of energy-adjusted magnesium intake. The test for linearity among all quartiles was significant, $P < 0.004$ (F test). *Significantly different from quartile 4, $P < 0.01$ (ANOVA). **Significantly different from quartile 1, $P < 0.02$ (ANCOVA; adjusted for age, weight, height, physical activity, smoking, and social status).
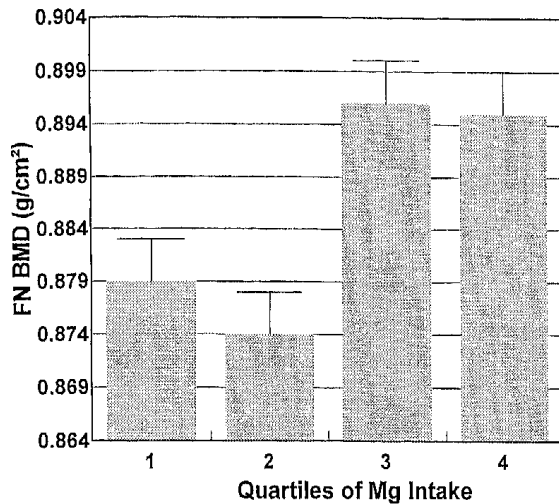
FIGURE 2. Mean (± SEM) increase in femoral neck bone mineral density (FN BMD) with quartiles of energy-adjusted magnesium intake. The test for linearity among all quartiles was significant, $P < 0.008$ (F test). There were no significant differences between the lowest and highest quartiles.

FIGURE 4. Mean (± SEM) increase in femoral neck bone mineral density (FN BMD) with quartiles of energy-adjusted potassium intake. The test for linearity among all quartiles was significant, $P < 0.01$ (F test). *Significantly different from quartile 4, $P < 0.009$ (ANOVA). **Significantly different from quartile 4, $P < 0.06$ (ANCOVA; adjusted for age, weight, height, physical activity, smoking and social status).

a low intake of milk in early adulthood ($P < 0.02$; **Table 5**). Differences in BMD at the other sites were not significant. After adjustment for the confounding variables, BMD remained significantly different at the LS site according to milk intake in early adulthood ($P < 0.03$). Reported high intakes of milk during both childhood and early adulthood were significantly and positively related to current energy, calcium, and milk intakes ($P < 0.0001$, chi-square test). However, adjustment for current calcium intake did not alter significantly the relation between past milk consumption and bone mass.

BMD was significantly lower at the four sites (LS, FN, FT, and FW) in women who reported a low intake of fruit in early adulthood than in those who reported a medium or high intake ($P < 0.01$; Table 5), which remained significant at the LS and FT sites after appropriate adjustment ($P < 0.01$).
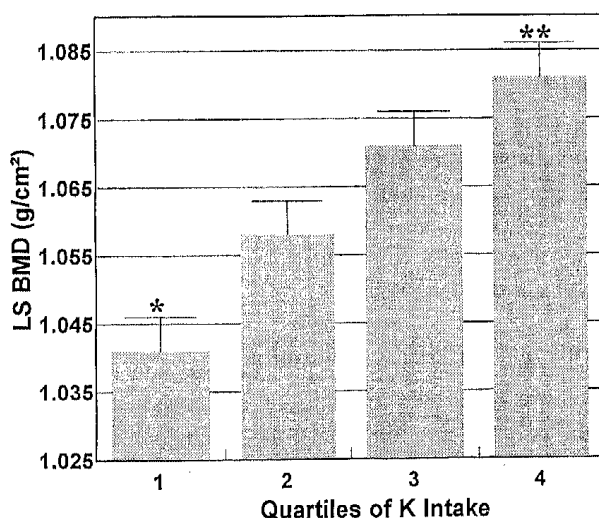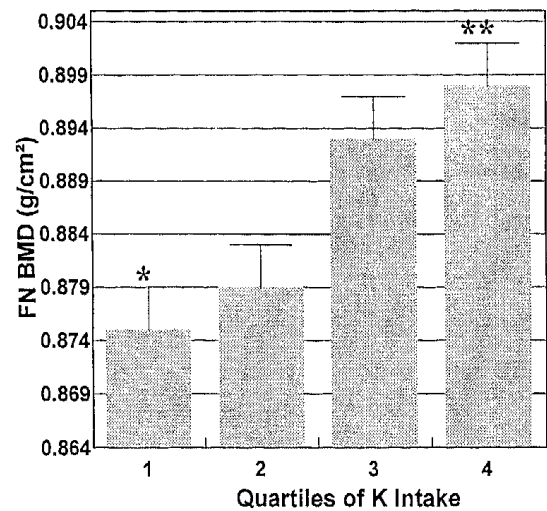
DISCUSSION

General

We investigated the association between nutrient intake and BMD, with adjustment for many important confounding factors, including total energy intake. This study was the first cross-sectional, population-based study to report the association between dietary intake and bone mass at the clinically important sites of the LS and FN in women near peak bone mass by using a purpose-designed tool for assessment of dietary intake, namely the FFQ. In a previous population-based study, dietary intake was assessed by using 24-h recall and BMD was measured with single-photon absorptiometry (9). Only one other recently published study in selected volunteers adjusted for total energy intake in the analysis of diet and bone
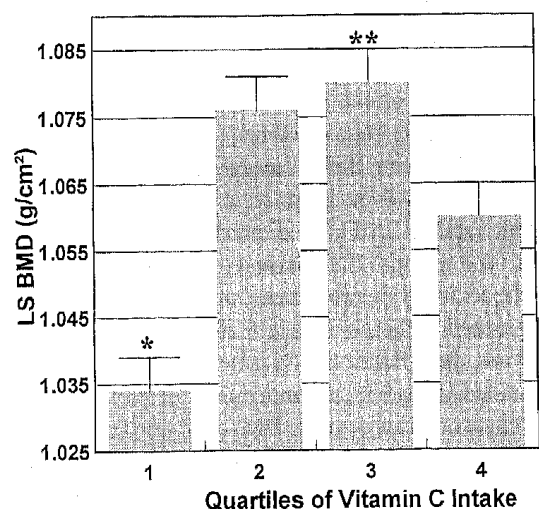

FIGURE 3. Mean (± SEM) increase in lumbar spine bone mineral density (LS BMD) with quartiles of energy-adjusted potassium intake. The test for linearity among all quartiles was significant, $P < 0.005$ (F test). *Significantly different from quartile 4, $P < 0.007$ (ANOVA). **Significantly different from quartile 1, $P < 0.06$ (ANCOVA; adjusted for age, weight, height, physical activity, smoking, and social status).

FIGURE 5. Mean (± SEM) increase in lumbar spine bone mineral density (LS BMD) with quartiles of energy-adjusted vitamin C intake. The test for linearity among all quartiles was not significant. *Significantly different from quartile 3, $P < 0.001$ (ANOVA). **Significantly different from quartile 1, $P < 0.002$ (ANCOVA; adjusted for age, weight, height, physical activity, smoking, and social status).

FIGURE 6. Mean (± SEM) increase in femoral neck bone mineral density (FN BMD) with quartiles of energy adjusted for vitamin C intake. The test for linearity among all quartiles was not significant. *Significantly different from quartile 3, $P < 0.005$ (ANOVA). **Significantly different from quartile 1, $P < 0.01$ (ANCOVA; adjusted for age, weight, height, physical activity, smoking, and social status).
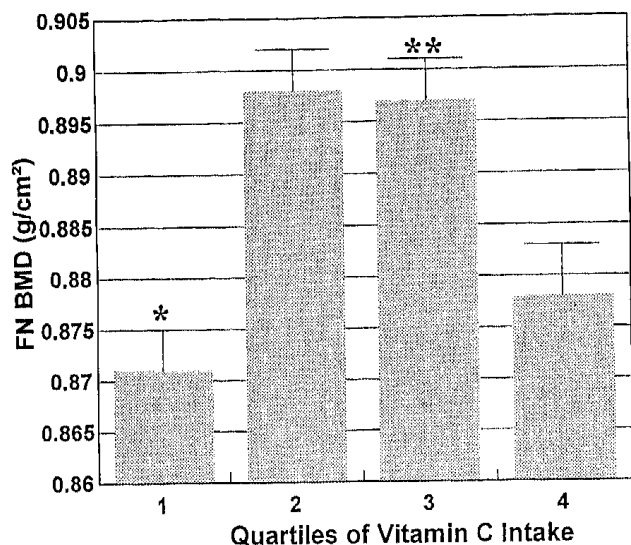
mass relations (25). The importance of such adjustment has been stressed by Willett (23) and by Prentice et al (26) in relation to adequate correction for body size in BMD measurements.

Subjects were selected for this study from women who had participated in the Osteoporosis Screening Program (17). A random cross-section of the population was achieved because the response rate was good (82%), and, on the basis of information from nonparticipants, the only significant difference between the two groups was in duration of smoking (years).

The strict exclusion criteria ensured that the data for analysis were derived only from women who were classified as "healthy" with regard to BMD and bone metabolism, and who were as close to peak bone mass as possible within the confines of the overall study design. Women who had not had regular periods within the previous 6 mo were excluded from the study and thus the cohort was unlikely to have included women with ovarian failure.

Data on dietary intake were collected after the BMD scan, which may not have been ideal because there were differences in time span between BMD measurements and assessment of dietary intake. However, there were no significant differences in nutrient intake, BMD, or dietary effects on BMD for the different time groups. Furthermore, because current diet generally reflects usual dietary habits, at least in recent years (27), it was assumed with confidence that the women with a 24-mo gap between their BMD measurement and dietary assessment still provided an adequate indication of their diet for that period. A seasonal variation in BMD and nutrient intake, which may occur, was partly controlled for by matching the season (winter or summer) of the BMD measurement with assessment of dietary intake. Thus, analysis of both the time and seasonal groups as a whole appeared justified.

The FFQ was generally well answered and the energy equivalent (energy intake/calculated basal metabolic rate) was well within the range of that established for satisfactory completion (28). One of the main limitations of this study, however, was

that the design was cross-sectional. Thus, we can state that there was a relation between nutrition and bone mass but we could not make any conclusions about the influence of nutrition on bone health.

Association between dietary intake and bone mass

Analysis of the association between present and past dietary intakes and BMD resulted in findings that, in part, support the work of others, but that also suggest several associations that have not been reported previously. Associations between nutrients and BMD tended to be more marked for the LS, which may be explained by the predominance of cancellous bone at this site. The surface area of this type of bone is greater than that of cortical bone, even though it occupies only 20% of the skeletal mass in the healthy adult skeleton, and bone remodeling is known to occur at an annual rate of 25% (compared with 2–3% in cortical bone) (29).

Calcium

The study did not find a significant relation between current calcium intake and BMD. These findings agree with those of some (13, 14, 16) but not all (15, 30–33) studies. However, in many of these studies calcium intakes were assessed by using a questionnaire that only asked questions about calcium-containing foods (15, 33) without any adjustment for total energy intake. Thus, the results observed may have been due to a higher energy intake (34) and possibly greater PALs.

The present investigation did show significant differences in BMD with different intakes of milk in earlier life. Because ≈40–60% of the calcium intake in UK children is believed to be from milk (35), these data appear to suggest a positive effect of calcium intake in early life on bone mass. These findings support those of previous studies that investigated past calcium intake in premenopausal women (36, 37) and past milk consumption in postmenopausal (38) and middle-aged and elderly (39, 40) women, and highlight further the importance of adequate calcium nutrition during the period of bone mass accretion.

It is important to note, however, that these past intake data are subject to recall bias and thus the findings should be interpreted with caution. Individuals were asked to consider how much of a particular food they consumed 30 y ago and there is no method available to establish the validity of the data collected. Past intakes of milk were found to be positively correlated with present intakes, and thus women may have answered the questions based on their present consumption. Although this is a common phenomenon (41, 42), there are data that suggest that answers to recall questions agree better with actual past intakes than do questions about current dietary intakes (43, 44).

Minerals and antioxidant vitamins

A significant association was found between intakes of potassium, magnesium, zinc, vitamin C, and fiber and BMD. These results remained significant for most of the nutrients after adjustment for many of the important confounding factors. Differences in BMD between quartiles for zinc, fiber, and vitamin C were particularly evident in the lowest quartile, possibly suggesting a threshold effect. Intakes of vitamin C were very high in this group of women compared with the Scottish average (45), with 90% of the women having intakes above the RNI (40 mg/d). However, intakes were not especially

TABLE 5
Difference in bone mineral density (BMD) by past intake of milk and fruit in early adulthood (age 20–30 y)[1]

	Past intake of milk			Past intake of fruit		
	Low	Medium	High	Low	Medium	High
	g/cm^2			g/cm^2		
LS BMD	$1.04 \pm 0.15^{2,3}$	1.08 ± 0.17	1.07 ± 0.17	$1.03 \pm 0.16^{2,3}$	1.07 ± 0.16	1.08 ± 0.17
	(1.03–1.06)	(1.06–1.09)	(1.04–1.10)	(1.01–1.05)	(1.05–1.08)	(1.06–1.10)
FN BMD	0.87 ± 0.12^{2}	0.89 ± 0.13	0.90 ± 0.13	0.86 ± 0.12^{2}	0.89 ± 0.13	0.89 ± 0.12
	(0.86–0.88)	(0.88–0.90)	0.882–0.92	(0.85–0.88)	(0.88–0.90)	(0.88–0.91)
FT BMD	0.71 ± 0.11^{2}	0.72 ± 0.13	0.73 ± 0.11	$0.70 \pm 0.13^{2,4}$	0.72 ± 0.13	0.73 ± 0.11
	(0.70–0.72)	(0.71–0.74)	(0.71–0.75)	(0.68–0.72)	(0.71–0.73)	(0.72–0.74)
FW BMD	0.83 ± 0.15	0.85 ± 0.16	0.84 ± 0.17	0.82 ± 0.15^{2}	0.85 ± 0.16	0.86 ± 0.15
	(0.82–0.85)	(0.83–0.86)	(0.82–0.87)	(0.80–0.84)	(0.83–0.86)	(0.84–0.87)

[1] $\bar{x} \pm$ SD; 95% CIs in parentheses. LS, lumbar spine; FN, femoral neck; FT, femoral trochanter; FW, femoral Ward's area.
[2-4] Significantly different from medium and high intake (adjusted for age, weight, height, physical activity, smoking, and social status): [2] $P < 0.01$ (ANOVA), [3] $P < 0.03$ (ANCOVA), [4] $P < 0.01$ (ANCOVA).

high when compared with those in other countries (46, 47) and thus are not necessarily a reflection of overreporting.

Associations between vitamin C intake and BMD have only been reported previously in one study of postmenopausal women (10), and may be explained by the requirement of vitamin C in the process of collagen hydroxylation (48).

Several studies have noted a positive relation between magnesium intake and BMD. Angus et al (16) found the dietary intake of magnesium to be positively correlated with and a significant predictor of forearm BMC in premenopausal women. Mean serum magnesium was also positively correlated with forearm bone mass. Dietary intakes of magnesium have also been found to be significantly reduced (49) and serum magnesium low (50) in postmenopausal osteoporotic women. Magnesium supplementation has been shown to prevent fracture, increase bone mass, and arrest bone loss in postmenopausal women (51). Furthermore, cancellous bone from osteoporotic humans has been shown to have low magnesium contents (52).

Zinc intakes in these women were likely to have been underestimated because the data available on the zinc contents of foods were incomplete. However, zinc intake was positively associated with BMD at the LS site and other evidence of a positive association between dietary intake of zinc and bone mass in pre- (16) and postmenopausal women (10) has been reported. Reduced serum concentrations (53), increased zinc excretion (54), and lower skeletal concentrations (55) have been reported in osteoporotic patients, although others did not find this association (56). Although zinc is known to be essential for growth in humans and animals, its role in osteoporosis is unclear. In small animals zinc stimulates bone growth and bone mineralization (57), and it has been shown to have a stimulatory effect on bone formation in tissue culture (58). More recent data suggest that it is a highly potent and selective inhibitor of osteoclastic bone resorption even at very low concentrations (59).

Several trace elements (including zinc, copper, and manganese) are known to be essential for organic bone matrix synthesis, but few studies have investigated the importance of these trace elements on bone health. Strause et al (60) showed that bone loss in calcium-supplemented older postmenopausal women can be further arrested by concomitant increases in trace mineral intake. Copper and manganese intakes were not assessed in this cross-sectional study, but the effect of trace mineral intake on bone health requires further attention.

Few studies have found an association between dietary intake of potassium and BMD, although a paper published recently found a weak but positive relation between energy-adjusted intake of potassium and total-body bone mass (25). Potassium bicarbonate has been shown to decrease urinary calcium excretion, and in turn improve calcium balance in healthy premenopausal (61–63) and postmenopausal women (64). More recently, potassium bicarbonate has been shown to decrease bone resorption and increase the rate of bone formation in postmenopausal women (65).

These findings and the possible potential benefit of dietary potassium on bone mass seem to support the theory that the skeleton is a reservoir of labile base (in the form of alkaline salts of calcium) that can be mobilized for the defense of blood pH and plasma bicarbonate concentrations. Wachman and Bernstein (66) first proposed the role of the skeleton in acid-base homeostasis in adults. The progressive decline in bone mass that occurs with age, which is ultimately expressed as osteoporosis, may result in part from the lifelong mobilization of skeletal salts to balance endogenous acid generated from acid-producing foods such as meat, compared with a high intake of foods that are alkaline-forming such as fruit and vegetables (64, 67). In studies of bone in vitro, extracellular acidification increases the activity of osteoclasts (cells that mediate bone resorption) and inhibits the activity of osteoblasts (cells that mediate bone formation) (68).

The relation between bone mass and potassium, magnesium, fiber, and vitamin C in the present study may reflect differences in fruit and vegetable consumption because these nutrients are found in abundance in the two food groups, as is fiber, which also had a positive association with BMD. The results from the current diet were more uniquely supported by the association between bone mass and fruit intake in childhood and early adulthood. Because fruit and vegetables are alkaline-producing foods (ie, excretion of renal base exceeds excretion of acid), high, long-term ingestion may have a beneficial effect on bone health. A positive relation between vegetable consumption and BMD has been reported (69).

Because of the large number of nutrients studied and the possibility that one nutrient may serve as a surrogate for the intake of another, associations between the different nutrients were examined. Highly significant correlations were found between intakes of magnesium and potassium, magnesium and

zinc, and magnesium and fiber. Further analysis by quartile of intake entering each of these nutrients as covariates showed the relation between magnesium and BMD to be unaffected by the other micronutrients. These relations were investigated further by using regression analysis, which found both magnesium and alcohol intakes to be independent predictors of BMD.

Alcohol

Moderate intakes of alcohol were positively correlated with LS BMD and a significant difference was found between the lowest and highest quartiles of alcohol intake, which remained significant after appropriate adjustment. Regression analysis also showed alcohol intake to be an independent predictor of BMD. Such results agree with the findings of others for moderate alcohol intake in postmenopausal women (15, 70). Hansen et al (71) reported a decreased rate of bone loss with moderate alcohol consumption, and a positive association with bone mass was also found in men and women (72). However, in premenopausal women Laitinen et al (70) found the reverse. Different habitual levels of alcohol consumption between populations may account for the different reported effects in the literature. Indeed, in this group of women alcohol consumption was modest (mean intake 7 g/d, equivalent to ≈3–4 glasses of wine/wk).

A suggested mechanism for the effect of alcohol intake on bone mass is the induction of adrenal production of androstenedione and its adrenal conversion to estrone (73). A positive correlation has been reported between alcohol intake and serum estradiol concentration (73). However, the mechanisms involved and the benefits of moderate alcohol intake in premenopausal women are unclear and require further study.

Conclusions

In conclusion, the results of this study suggest that high current intakes of the nutrients potassium, magnesium, vitamin C, fiber, and zinc were associated with a higher bone mass and that a high past consumption of fruit had a positive effect on adult bone mass. These findings appear to indicate that high long-term intakes of nutrients found in abundance in fruit and vegetables may be important to bone health, possibly because of their beneficial effect on acid-base balance.   ▣

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