Dietary magnesium and potassium intakes and circulating magnesium are associated with heel bone ultrasound attenuation and osteoporotic fracture risk in the EPIC-Norfolk cohort study[1,2]

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ABSTRACT

Background: In our aging population, maintenance of bone health is critical to reduce the risk of osteoporosis and potentially debilitating consequences of fractures in older individuals. Among modifiable lifestyle and dietary factors, dietary magnesium and potassium intakes are postulated to influence bone quality and osteoporosis, principally via calcium-dependent alteration of bone structure and turnover.

Objective: We investigated the influence of dietary magnesium and potassium intakes, as well as circulating magnesium, on bone density status and fracture risk in an adult population in the United Kingdom.

Design: A random subset of 4000 individuals from the European Prospective Investigation into Cancer and Nutrition–Norfolk cohort of 25,639 men and women with baseline data was used for bone density cross-sectional analyses and combined with fracture cases (n = 1502) for fracture case-cohort longitudinal analyses (mean follow-up 13.4 y). Relevant biological, lifestyle, and dietary covariates were used in multivariate regression analyses to determine associations between dietary magnesium and potassium intakes and calcaneal broadband ultrasound attenuation (BUA), as well as in Prentice-weighted Cox regression to determine associated risk of fracture. Separate analyses, excluding dietary covariates, investigated associations of BUA and fractures with serum magnesium concentration.

Results: Statistically significant positive trends in calcaneal BUA for women (n = 1360) but not men (n = 968) were apparent across increasing quintiles of magnesium plus potassium (Mg+K) z score intake (P = 0.03) or potassium intake alone (P = 0.04). Reduced hip fracture risk in both men (n = 1958) and women (n = 2755) was evident for individuals in specific Mg+K z score intake quintiles compared with the lowest. Statistically significant trends in fracture risk in men across serum magnesium concentration groups were apparent for spine fractures (P = 0.02) and total hip, spine, and wrist fractures (P = 0.02). None of these individual statistically significant associations remained after adjustment for multiple testing.

Conclusions: These findings enhance the limited literature studying the association of magnesium and potassium with bone density and demonstrate that further investigation is warranted into the mechanisms involved and the potential protective role against osteoporosis. *Am J Clin Nutr* 2015;102:376–84.

Keywords: dietary minerals, fractures, osteoporosis, bone density, quantitative ultrasound, epidemiology

INTRODUCTION

A multitude of factors are known to influence bone health, including modifiable factors such as diet, physical activity, and smoking but also age, sex, and genetics (1). Osteoporosis, characterized by bone loss due to deterioration of bone microarchitecture and consequent increased risk of fracture, is particularly associated with age and thus represents a major public health concern for our aging population (2). Calcium and vitamin D have traditionally been the primary nutritional candidates for osteoporosis prevention and maintenance of bone health (3), but magnesium intake has also been linked with bone mass and magnesium deficiency with osteoporosis (4–8). Magnesium is a major component of bone, with 67% of total body magnesium found there (9). Animal studies have suggested a number of mechanisms for involvement of magnesium in bone metabolism, including nitric oxide–dependent effects on osteoblast activity and osteoclast number (10); influence of magnesium on hydroxyapatite crystal formation and consequent bone stiffness (11); regulation of calcium homeostasis through parathyroid hormone, 1,25-dihydroxyvitamin D, and magnesium-dependent calcium channels (9); and altered inflammatory cytokine release (12). Similarly, epidemiologic studies have associated lower dietary potassium intake with poorer bone density (5, 6, 8, 13). Increasing potassium intake increases urinary retention, reducing loss of calcium and thus creating a more positive calcium balance and inhibiting bone resorption; urinary loss of phosphorus also decreases, which inhibits renal synthesis of 1,25-dihydroxyvitamin D and cuts intestinal

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[2] Supplemental Tables 1–5 are available from the "Supplemental data" link in the online posting of the article and from the same link in the online table of contents at http://ajcn.nutrition.org.

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absorption of calcium, stopping the positive calcium balance persisting (14). Occurrence of this stabilization has recently been disputed, although potassium source differences may be the cause of the discrepancy between studies (15).

When considering the dietary association of magnesium and potassium with bone health, it is most appropriate to study these minerals concurrently because they are frequently consumed together from intact or moderately altered plant or animal tissues (16). Metabolism of magnesium and potassium is linked because magnesium is required for effective Na^+/K^+-ATPase pump function (17), magnesium and potassium have additive effects in preventing an increase in the endogenous sodium potassium pump inhibitor (16), and both have direct and indirect effects on calcium homeostasis (9, 18). Previous studies of the association of dietary magnesium and potassium with bone health have had limited generalizability because of their focus on discrete population groups, such as narrow age range groups of relatively old (5, 19) or young individuals (20), restrictions to premenopausal (8, 20) or postmenopausal women only (13), and non–United Kingdom residents (5, 7, 20). Indeed, the most recent and comprehensive study, with a large cohort size and longitudinal analysis of fracture risk, was also limited to women only (7). The current study therefore aimed to explore potential associations of dietary magnesium and potassium intakes and circulating magnesium with bone density status, as well as risk of incident osteoporotic fractures in a general population of men and women in the United Kingdom, by using a measure of broadband ultrasound attenuation (BUA)[5] of the calcaneus and records of incident fractures of the hip, spine, and wrist.

METHODS

The European Prospective Investigation into Cancer and Nutrition (EPIC)–Norfolk cohort analyzed in this study is part of EPIC, a global collaboration involving 10 countries developed primarily to examine the association between diet and cancer, with additional health outcomes also examined in EPIC-Norfolk. This cohort has been described in detail previously (21), but in brief, the Norfolk cohort consisted of 25,639 men and women aged 40–79 y living in the general community who participated in a baseline health check between 1993 and 1997. A second health check was attended by 15,786 participants, aged 42–82 y, between 1997 and 2000, when quantitative ultrasound measurements of the calcaneus (heel bone) were performed according to standardized protocols by using a contact ultrasound bone analyzer device (McCue Ultrasonics). Quantitative ultrasound represents a cheaper, more rapid, and easier method of assessing bone density status in general practice compared with the gold standard of dual X-ray absorptiometry, and has been shown capable of predicting fracture risk (22). Measurements of BUA (dB/MHz) from each foot were taken at least in duplicate, and the mean of both feet was recorded, as described previously (22).

The data set analyzed here includes 4000 randomly selected participants with baseline health check data, plus a group of 1502 participants with fractures, representing all hip, spine, and wrist fracture cases in the cohort up to 31 March 2009. Some overlap exists between the random subcohort and the fracture cases, and thus the fracture case cohort contains 5319 unique individuals (4713 participants had complete data for diet and fracture analyses; 3469 for serum and fracture analyses). Ultrasound data were available for 2341 individuals (2328 participants had complete data for diet and ultrasound analyses; 1726 for serum and ultrasound analyses).

The Norfolk District Health Authority Ethics Committee approved all procedures involving human subjects, and written informed consent was provided by all participants according to the Declaration of Helsinki.

Height and weight were measured according to standard protocols (21) at both health checks, conducted at a clinic or the participant's general practitioner's practice. Height was determined to the nearest millimeter by using a freestanding stadiometer. Weight was recorded to the nearest 0.2 kg with the participant wearing light clothing and no shoes. BMI (in kg/m^2) was calculated from these measurements.

Participants also completed a self-administered health and lifestyle questionnaire at both health checks. This included smoking status, categorized as current, former, or never; family history of osteoporosis, categorized as yes or no; menopausal status (women only), categorized as premenopausal, perimenopausal (<1 y), perimenopausal (1–5 y), or postmenopausal; and hormone replacement therapy (HRT) status (women only), categorized as current, former, or never users. A short physical activity questionnaire was used to assess typical physical activity over the previous 12 mo. Physical activity levels were then categorized into inactive, moderately inactive, moderately active, and active categories by a method validated against heart rate monitoring data (21, 23).

Dietary intake of each participant was assessed by using a 7-d food diary (24), with each participant recording all food and drink consumed within a 7-d period, as well as the portion sizes. This method has been validated previously, proving more accurate in estimating dietary nutrient intake than food-frequency questionnaires (FFQs) (21, 25). Detail of the Data Into Nutrients for Epidemiologic Research software used to record and translate the dietary information provided by the 7-d food diaries into nutrient quantities is reported elsewhere (26). All data entries were checked by nutritionists trained in use of the system (27).

Serum magnesium concentration was determined by using blood sampled by peripheral venipuncture during the baseline health check. Samples were stored in liquid nitrogen at −196°C until analyzed by Quotient Bioresearch, by using an Olympus AU640 Chemistry Immuno Analyzer to perform a xylidyl blue–based colorimetric assay.

Fracture incidence data were collected by questionnaire at baseline and follow-up health checks. In addition, the East Norfolk Health Authority database, which records all hospital contact that Norfolk residents have in England and Wales, was available to EPIC researchers for data linkage (28). This enabled the incidence of osteoporotic fractures occurring in the cohort, up to the end of March 2009, to be determined by retrieving data with the National Health Service numbers of EPIC participants

[5] Abbreviations used: BMD, bone mineral density; BUA, broadband ultrasound attenuation; EPIC, European Prospective Investigation into Cancer and Nutrition; FFQ, food-frequency questionnaire; HRT, hormone replacement therapy.

and the International Classification of Diseases Ninth and Tenth Revision diagnostic codes for osteoporotic fractures by site (hip, spine, and wrist).

Statistical analyses were performed by using STATA statistical software (version 12; Stata Corp.). All analyses were stratified by sex because significant differences in age-related changes in bone between men and women have been reported for this population, with a much greater magnitude of deterioration evident in women (22). Hypotheses and covariates included in regression models were well defined a priori by using evidence from previous research, and thus P values ≤ 0.05 were considered statistically significant in individual analyses. The individual hypotheses tested in this study have been grouped into families of tests (**Supplemental Table 1**), allowing the significance of individual P values to be determined in comparison to a Bonferroni-generated family-wise critical P value.

Because of the high degree of collinearity between magnesium and potassium dietary intakes (Pearson $r = 0.84$ and $r = 0.82$ in men and women, both $P < 0.001$) and thus the potential for statistical issues and any independent effects to be diminished, a summation of magnesium and potassium intake was used as the main exposure; however, because the amounts of each mineral consumed varied widely, both minerals were standardized before summation, resulting in a magnesium plus potassium (Mg+K) z score intake variable (5).

Univariate linear regression was used to estimate the association of selected biological, lifestyle, and dietary factors with sex-specific quintiles of dietary magnesium, potassium, or Mg+K z score intake. Multivariable regression with ANCOVA was used to investigate differences in calcaneal BUA across sex-specific quintiles of dietary magnesium, potassium, or Mg+K z score intake. An adjusted model was tested, correcting for the potential effects of biological (age, BMI, family history of osteoporosis, menopausal status, HRT status, and corticosteroid use), lifestyle (smoking status and physical activity), and dietary factors [calcium intake (29, 30), total energy intake (31), and calcium and vitamin D supplement use, previously shown to influence bone ultrasound measurements in this population (22, 32)]. Participants were excluded from analyses if they had missing values for any variables included in the multivariate model ($n = 1672$, 41.8%). In a similar way, differences in calcaneal BUA across sex-specific groups of serum magnesium concentration were investigated by using the same covariates but excluding dietary factors in the adjusted model. Published guidance suggests 0.7–1.0 mmol/L should be used as a normal reference range (33). Concentration groups were categorized as $<$0.7 mmol/L (group 1, deficient), 0.7–0.8 mmol/L (group 2), 0.8–0.9 mmol/L (group 3), 0.9–1.0 mmol/L (group 4), and $>$1.0 mmol/L (group 5, excess).

Prentice-weighted Cox regression was used to investigate associations between the incidence of fractures and sex-specific quintiles of dietary magnesium, potassium, or Mg+K z score intake. An unadjusted model without covariates was tested followed by a model adjusting for the aforementioned variables. The full case-cohort data set described above, including the random subset and all fracture cases, was used for these analyses. Participants were excluded from analyses if they had missing values for any variables included in the adjusted model. For analysis of site-specific fracture risk (hip, spine, or wrist), other fracture data were excluded from the analysis unless contained in the subcohort to retain a distinct control group. Total risk of hip, spine, or wrist fracture was calculated as the risk of an individual having one of these types of fracture. This total does not include multiple fractures, and therefore the site-specific fracture incidences described may not sum to the total. The association between incidence of fractures and sex-specific groups of serum magnesium concentration was investigated by using the same covariates but excluding dietary factors in the adjusted model.

RESULTS

Descriptive statistics

Selected characteristics are summarized in **Table 1**, for men and women, as means ± SDs or frequency and percentage for categorical variables. There were 968 men and 1360 women in the ultrasound cohort population with information for all selected variables; in the fracture case cohort, there were data for 1958 men and 2755 women. Age was 63.0 ± 9.3 y for men and 61.7 ± 9.2 y for women in the ultrasound cohort; in the fracture case cohort, age at baseline was 59.7 ± 9.6 y for men and 59.8 ± 9.5 y for women. BMI was 26.9 ± 3.4 for men and 26.5 ± 4.4 for women in the ultrasound cohort; in the fracture case cohort, BMI at baseline was 26.5 ± 3.3 for men and 26.2 ± 4.3 for women. Total daily energy intake was 2263 ± 478 kcal for men and 1732 ± 374 kcal for women in the ultrasound cohort; in the fracture case cohort, intake at baseline was 2239 ± 514 kcal for men and 1683 ± 385 kcal for women. Magnesium intake was 329 ± 92 mg/d for men and 277 ± 72 mg/d for women in the ultrasound cohort; in the fracture case cohort, intake at baseline was 321 ± 92 mg/d for men and 265 ± 73 mg/d for women; these values are slightly higher than the United Kingdom Reference Nutrient Intake of 300 mg and 270 mg (34), respectively. Calcium intake was 925 ± 282 mg/d for men and 782 ± 247 mg/d for women in the ultrasound cohort; in the fracture case cohort, intake at baseline was 914 ± 296 mg/d for men and 762 ± 253 mg/d for women; these values are also higher than the United Kingdom Reference Nutrient Intake of 700 mg for all adults $>$19 y (34). Calcium supplements were used by 1.5% of men and 7.2% of women in the ultrasound cohort and by 1.3% of men and 5.6% of women in the fracture case cohort. Potassium intake was 3525 ± 803 mg/d for men and 3070 ± 662 mg/d for women in the ultrasound cohort; in the fracture case cohort, mean intake at baseline was 3445 ± 815 mg/d for men and 2969 ± 690 mg/d for women. Potassium intake for women in this cohort is therefore lower than the United Kingdom Reference Nutrient Intake of 3500 mg for all adults $>$18 y (34). Serum magnesium concentration was 0.81 ± 0.12 mmol/L for men ($n = 1006$) and 0.79 ± 0.13 mmol/L for women ($n = 720$). Vitamin D supplements were used by 23.6% of men and 34.6% of women in the ultrasound cohort and by 22.0% of men and 31.8% of women in the fracture case cohort.

Current smokers represented 7.9% of men and 9.8% of women in the ultrasound cohort, and the proportion of never smokers was higher for women than for men (58.6% compared with 36.6%); in the fracture case cohort, current smokers and never smokers represented 12.2% and 32.6% of men and 12.5% and 55.2% of

TABLE 1
Selected characteristics of the EPIC-Norfolk cohort population stratified by sex for the ultrasound cohort group (n = 2328) and the fracture case-cohort group (n = 4713)[1]

	Ultrasound cohort			Fracture case-cohort		
Selected characteristics	Men (n = 968)	Women (n = 1360)	P value	Men (n = 1958)	Women (n = 2755)	P value
Age, y	63.0 ± 9.3[2]	61.7 ± 9.2	<0.001	59.7 ± 9.6	59.8 ± 9.5	0.809
BMI, kg/m^2	26.9 ± 3.4	26.5 ± 4.4	0.039	26.5 ± 3.3	26.2 ± 4.3	0.004
Magnesium intake, mg/d	329 ± 92	277 ± 72	<0.001	321 ± 92	265 ± 73	<0.001
Potassium intake, mg/d	3525 ± 803	3070 ± 662	<0.001	3445 ± 815	2969 ± 690	<0.001
Calcium intake, mg/d	925 ± 282	782 ± 247	<0.001	914 ± 296	762 ± 253	<0.001
Calcium supplement use, n (%)	14 (1.5)	98 (7.2)	<0.001	25 (1.3)	155 (5.6)	<0.001
Vitamin D supplement use, n (%)	228 (23.6)	471 (34.6)	<0.001	430 (22.0)	875 (31.8)	<0.001
Total energy intake, kcal/d	2263 ± 478	1732 ± 374	<0.001	2239 ± 514	1683 ± 385	<0.001
Serum Mg, mmol/L	0.81 ± 0.12[3]	0.79 ± 0.13[4]	0.003	0.81 ± 0.12[5]	0.79 ± 0.13[6]	0.001
BUA, dB/MHz	89.6 ± 17.4	72.1 ± 16.5	<0.001			
Smoking, n (%)			<0.001			<0.001
Current	76 (7.9)	133 (9.8)		238 (12.2)	343 (12.5)	
Former	538 (55.6)	430 (31.6)		1082 (55.3)	890 (32.3)	
Never	354 (36.6)	797 (58.6)		638 (32.6)	1522 (55.2)	
Physical activity, n (%)			<0.001			<0.001
Inactive	275 (28.4)	342 (25.1)		614 (31.4)	908 (33.0)	
Moderately inactive	236 (24.4)	462 (34.0)		472 (24.1)	877 (31.8)	
Moderately active	248 (25.6)	333 (24.5)		436 (22.3)	577 (20.9)	
Active	209 (21.6)	223 (16.4)		436 (22.3)	393 (14.3)	
Family history of osteoporosis, n (%)			0.001			<0.001
No	937 (96.8)	1277 (93.9)		1900 (97.0)	2601 (94.4)	
Yes	31 (3.2)	83 (6.1)		58 (3.0)	154 (5.6)	
Corticosteroid use, n (%)			0.391			0.243
Current or former (>3 mo)	43 (4.4)	71 (5.2)		50 (2.6)	97 (3.5)	
Never (<3 mo)	925 (95.6)	1289 (94.8)		1908 (97.5)	2658 (96.5)	
Menopausal status, n (%)						
Premenopausal		86 (6.3)			414 (15.0)	
Perimenopausal (<1 y)		47 (3.5)			127 (4.6)	
Perimenopausal (1–5 y)		246 (18.1)			448 (16.3)	
Postmenopausal		981 (72.1)			1766 (64.1)	
HRT, n (%)						
Current		288 (21.2)			472 (17.1)	
Former		222 (16.3)			324 (11.8)	
Never		850 (62.5)			1959 (71.1)	

[1]Ultrasound group characteristics at second health check (time of ultrasound). Fracture group characteristics at first health check or time of consent. P values are for differences between men and women for each applicable variable, according to the t test for continuous or χ^2 for categorical variables. BUA, broadband ultrasound attenuation; EPIC, European Prospective Investigation into Cancer and Nutrition; HRT, hormone replacement therapy.
[2]Mean ± SD (all such values).
[3]n = 720.
[4]n = 1006.
[5]n = 1460.
[6]n = 2009.

women, respectively. There was a broad spread of physical activity levels across the 4 categories (inactive, moderately inactive, moderately active, or active) for both men and women, although there was a higher proportion of women classified as inactive or moderately inactive than men (59.1% compared with 52.8% in the ultrasound cohort; 64.8% compared with 55.5% in the fracture case cohort). Family history of osteoporosis in the ultrasound cohort was 3.2% in men and 6.1% in women; in the fracture case cohort, it was 3.0% in men and 5.6% in women. Most women (72.1% ultrasound cohort; 64.1% fracture case cohort) were postmenopausal, and 37.5% in the ultrasound cohort and 28.9% in the fracture case cohort were current or former users of HRT. Current or former users of corticosteroids for ≥3 mo accounted for 4.4% of men and 5.2% of women in the ultrasound cohort; in the fracture case cohort, it was 2.6% of men and 3.5% of women.

Associations between dietary magnesium and potassium intake and bone density

Mean calcaneal BUA values stratified by quintiles of dietary magnesium, potassium, or Mg+K z score intake are shown in **Figure 1**, stratified by sex. Data are presented for the fully adjusted model. In men, no linear trends in fully adjusted BUA were apparent across quintiles of magnesium, potassium, or Mg+K z score intake. In women, statistically significant linear trends were apparent across quintiles of potassium and Mg+K z score intake, but not magnesium intake alone, for fully adjusted

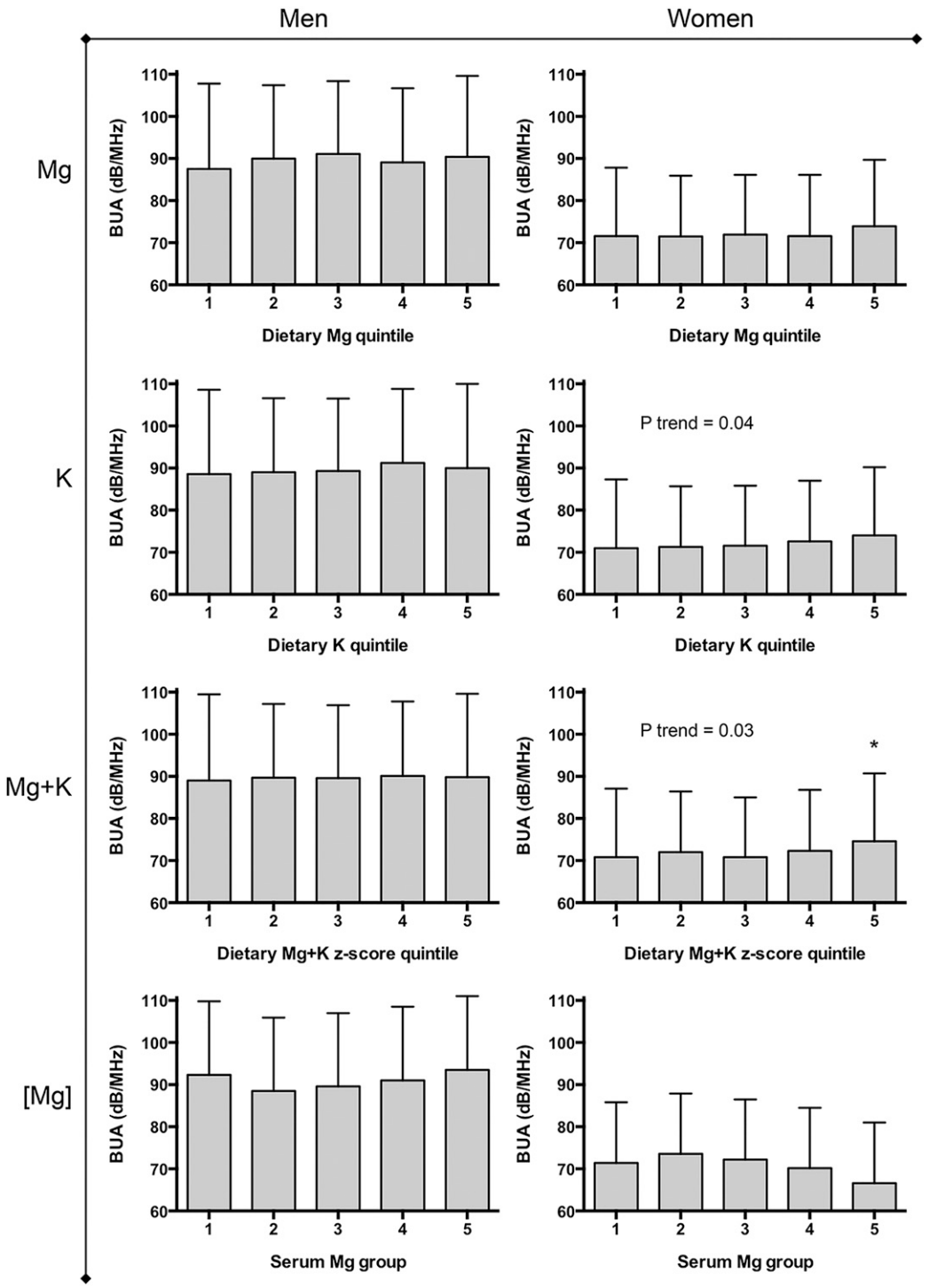

FIGURE 1 Fully adjusted calcaneal BUA of the EPIC-Norfolk cohort population (968 men and 1360 women) stratified by sex and quintiles of magnesium or potassium dietary intake, *z* score quintiles of dietary Mg+K intake, or serum magnesium concentration groups (720 men and 1006 women). Adjusted for age, BMI, smoking status, physical activity, family history of osteoporosis, menopausal and HRT status in women, corticosteroid use, calcium intake and supplement use (excluding serum Mg model), vitamin D supplement use (excluding serum Mg model), and total energy intake (excluding serum Mg model). Values are presented as means ± SDs. Mg intake (mean ± SD; mg/d) by Mg quintiles. Men*:* mean, 329 ± 32; Q1, 218 ± 31; Q2, 277 ± 12; Q3, 319 ± 13; Q4, 366 ± 16; Q5, 466 ± 73. Women*:* mean, 277 ± 72; Q1, 189 ± 26; Q2, 237 ± 10; Q3, 270 ± 10; Q4, 307 ± 12; Q5, 383 ± 58. K intake (mean ± SD; mg/d) by K quintiles. Men*:* mean, 3525 ± 803; Q1, 2505 ± 344; Q2, 3099 ± 125; Q3, 3478 ± 101; Q4, 3854 ± 122; Q5, 4697 ± 603. Women*:* mean, 3070 ± 662; Q1, 2196 ± 287; Q2, 2721 ± 99; Q3, 3038 ± 90; Q4, 3367 ± 106; Q5, 4030 ± 429. Mg intake (mean ± SD; mg/d) by Mg+K *z* score quintiles. Men*:* mean, 329 ± 92; Q1, 221 ± 35; Q2, 279 ± 22; Q3, 321 ± 29; Q4, 364 ± 29; Q5, 460 ± 78. Women*:* mean, 277 ± 72; Q1, 192 ± 29; Q2, 238 ± 19; Q3, 271 ± 21; Q4, 306 ± 24; Q5, 378 ± 61. K intake (mean ± SD; mg/d) by Mg+K *z* score quintiles. Men*:* mean, 3525 ± 803; Q1, 2539 ± 375; Q2, 3117 ± 218; Q3, 3489 ± 229; Q4, 3857 ± 270; Q5, 4630 ± 668. Women*:* mean, 3070 ± 662; Q1, 2217 ± 309; Q2, 2753 ± 177; Q3, 3047 ± 205; Q4, 3351 ± 230; Q5, 3983 ± 479. Serum Mg concentration groups: <0.7 mmol/L (group 1), 0.7–0.8 mmol/L (group 2), 0.8–0.9 mmol/L (group 3), 0.9–1.0 mmol/L (group 4), and >1.0 mmol/L (group 5). *$P \leq 0.05$ vs. quintile 1, according to ANCOVA (not statistically significant after multiple testing adjustment). BUA, broadband ultrasound attenuation; EPIC, European Prospective Investigation into Cancer and Nutrition; HRT, hormone replacement therapy; Q, quintile.

BUA ($P = 0.04$, $P = 0.03$, and $P = 0.15$, respectively). Individual statistically significant differences in fully adjusted BUA were also identified for women between quintile 5 and quintile 1 for Mg+K z score intake (74.6 ± 16.1 dB/MHz, $n = 272$ compared with 70.8 ± 16.3 dB/MHz, $n = 272$, a 5.3% difference; $P = 0.02$) but not potassium (74.0 ± 16.2 dB/MHz, $n = 272$ compared with 71.0 ± 16.3 dB/MHz, $n = 272$, a 4.2% difference; $P = 0.05$) or magnesium alone (73.9 ± 15.8 dB/MHz, $n = 272$ compared with 71.6 ± 16.2 dB/MHz, $n = 272$, a 3.3% difference; $P = 0.11$) (see Figure 1). No P values were below the Bonferroni-adjusted family-wise critical value (Supplemental Table 1).

Associations between serum magnesium groups and bone density

Analysis of bone density measures according to serum magnesium concentration groups, adjusting for all covariates previously described, with the exception of dietary factors, showed no statistically significant differences in BUA in either men or women (see Figure 1 and Supplemental Table 1). Furthermore, no correlation was apparent between dietary magnesium intake and serum magnesium concentration for either men ($r = 0.01$, $P = 0.87$, $n = 717$) or women ($r = -0.04$, $P = 0.25$, $n = 1006$).

Associations between dietary magnesium and potassium intake and fracture risk

Between baseline and follow-up, the percentage of men with one or more hip, spine, or wrist fractures was 23.4% lower in quintile 5 than in quintile 1 for magnesium intake quintiles, 18.1% for potassium quintiles, and 10.2% for Mg+K z score quintiles. In women, these figures were 35.9%, 32.1%, and 30.8%, respectively. Risk of hip fracture in men was statistically significantly lower in Mg+K z score quintiles 2 and 5 than in quintile 1 in the fully adjusted model ($P = 0.03$ and $P = 0.02$) (**Figure 2** and **Supplemental Table 2**). The lowest risk of hip fracture in men was evident in Mg+K z score quintile 5 (0.35; 95% CI: 0.14, 0.85). In women, a statistically significant reduced risk of hip fracture was evident in Mg+K z score quintile 4 compared with quintile 1 in the fully adjusted model (0.59; 95% CI: 0.36, 0.97; $P = 0.04$). A reduced risk of spinal fracture in women was evident for dietary magnesium quintile 3 compared with quintile 1 (0.49; 95% CI: 0.25, 0.97; $P = 0.04$) (Figure 2 and **Supplemental Table 3**) but not Mg+K z score or potassium quintiles (Figure 2, Supplemental Table 2, and **Supplemental Table 4**). No P values were below the Bonferroni-adjusted family-wise critical value (Supplemental Table 1).

Associations between serum magnesium groups and fracture risk

Analysis of risk of fracture according to concentration groups of serum magnesium showed a number of statistically significant associations (Figure 2 and **Supplemental Table 5**). In men, there were statistically significant trends in fracture risk across serum concentration groups for spine fractures ($P = 0.02$) and total hip, spine, and wrist fractures ($P = 0.02$) but not for hip ($P = 0.06$) or wrist fractures alone ($P = 0.38$). Hip fracture risk was statistically significantly lower in groups 2 ($P = 0.03$) and 3 ($P < 0.01$) than in group 1 in the fully adjusted model, with the

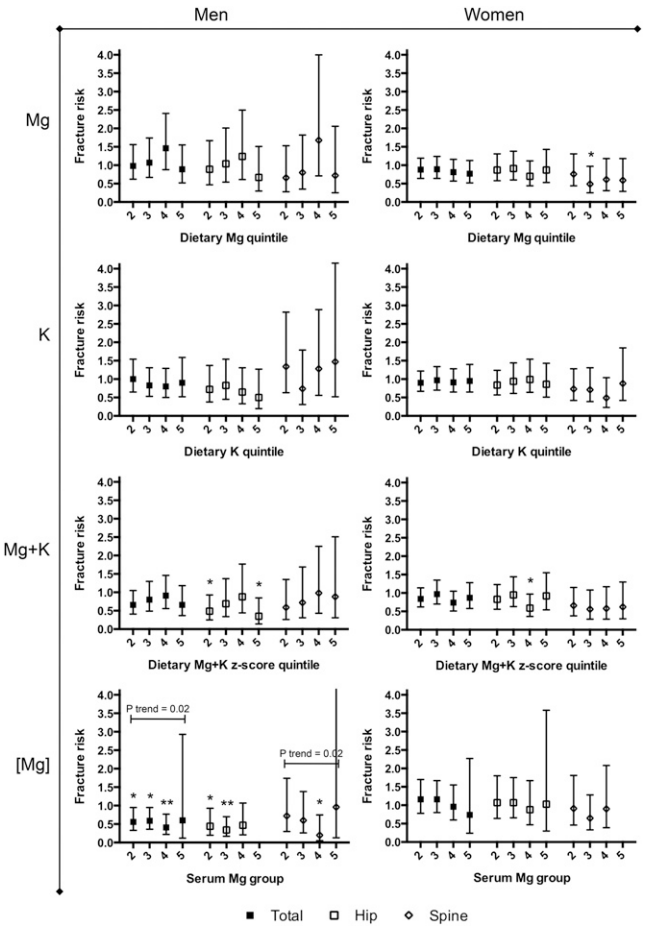

FIGURE 2 Risk of hip, spine, and wrist fractures in the EPIC-Norfolk cohort population (1958 men and 2755 women) at follow-up compared with baseline stratified by sex and quintile of magnesium or potassium dietary intake, z score quintiles of dietary Mg+K intake, or serum magnesium concentration groups (1460 men and 2009 women) (Prentice-weighted Cox proportional hazards ratio and 95% CI of quintiles or groups, with quintile or group 1 as reference). Adjusted for age, BMI, smoking status, physical activity, family history of osteoporosis, menopausal and HRT status in women, corticosteroid use, calcium intake and supplement use (excluding serum Mg model), vitamin D supplement use (excluding serum Mg model), and total energy intake (excluding serum Mg model). Values are presented as means ± SDs. Mg intake (mean ± SD; mg/d) by Mg quintiles. Men: mean, 321 ± 93; Q1, 209 ± 31; Q2, 268 ± 12; Q3, 312 ± 13; Q4, 358 ± 15; Q5, 460 ± 75. Women: mean, 265 ± 73; Q1, 175 ± 25; Q2, 223 ± 10; Q3, 257 ± 9; Q4, 294 ± 13; Q5, 373 ± 59. K intake (mean ± SD; mg/d) by K quintiles. Men: mean, 3449 ± 821; Q1, 2390 ± 356; Q2, 3019 ± 119; Q3, 3405 ± 111; Q4, 3797 ± 126; Q5, 4635 ± 607. Women: mean, 2964 ± 689; Q1, 2065 ± 285; Q2, 2595 ± 102; Q3, 2921 ± 92; Q4, 3268 ± 113; Q5, 3974 ± 448. Mg intake (mean ± SD; mg/d) by Mg+K z score quintiles. Men: mean, 321 ± 93; Q1, 212 ± 35; Q2, 271 ± 23; Q3, 314 ± 28; Q4, 357 ± 28; Q5, 454 ± 80. Women: mean, 265 ± 73; Q1, 178 ± 29; Q2, 225 ± 19; Q3, 257 ± 20; Q4, 294 ± 24; Q5, 368 ± 63. K intake (mean ± SD; mg/d) by Mg+K z score quintiles. Men: mean, 3449 ± 821; Q1, 2422 ± 386; Q2, 3040 ± 212; Q3, 3419 ± 245; Q4, 3788 ± 263; Q5, 4577 ± 663. Women: mean, 2964 ± 687; Q1, 2087 ± 307; Q2, 2618 ± 183; Q3, 2925 ± 189; Q4, 3257 ± 223; Q5, 3935 ± 490. Serum Mg concentration groups: <0.7 mmol/L (group 1), 0.7–0.8 mmol/L (group 2), 0.8–0.9 mmol/L (group 3), 0.9–1.0 mmol/L (group 4), and >1.0 mmol/L (group 5). *$P \leq 0.05$ vs. quintile 1, according to ANCOVA. **$P \leq 0.01$ (not statistically significant after multiple testing adjustment). Insufficient data were available in the highest serum Mg concentration group for some hazards ratio calculations. EPIC, European Prospective Investigation into Cancer and Nutrition; HRT, hormone replacement therapy; Q, quintile.

lowest risk in group 3 (0.34; 95% CI: 0.17, 0.70). Spinal fracture risk was statistically significantly lower (0.20; 95% CI: 0.05, 0.75; $P = 0.02$) in group 4 than in group 1; total risk of hip, spine, and wrist fractures was statistically significantly lower in groups 2 ($P = 0.03$), 3 ($P = 0.03$), and 4 ($P < 0.01$) than in group 1, with the lowest risk in group 4 (0.41; 95% CI: 0.22, 0.77). In women, there were no statistically significant trends for fracture risk across groups of magnesium serum concentration or between specific groups compared with group 1. No P values were below the Bonferroni-adjusted family-wise critical value (Supplemental Table 1).

DISCUSSION

This study has shown statistically significant associations between combined dietary magnesium and potassium intake and a quantitative measure of bone density, with significantly higher calcaneal BUA evident in women in the highest compared with lowest Mg+K z score intake quintiles of these micronutrients, after adjustment for important biological, lifestyle, and other dietary covariates. Furthermore, risk of hip fracture in both women and men was statistically significantly reduced in specific higher Mg+K intake quintiles compared with the lowest. We believe that this study is also the first to show lower total risk of hip, spine, or wrist fracture for men with a clinically normal serum magnesium concentration than for those classed as deficient. However, although each of the described associations was statistically significant individually, no significant associations were evident after adjusting for multiple testing.

The mechanisms by which magnesium and potassium may influence bone metabolism are not fully understood, although a number of theories have been proposed. Insufficient magnesium results in an increased rate of hydroxyapatite formation, resulting in larger crystals and thus lower bone mass and brittle bones, which may be unable to support normal loads. Magnesium also has an effect on osteoblast activity and osteoclast number through a nitric oxide–dependent mechanism (10), and both magnesium and potassium affect bone metabolism through altered calcium homeostasis via influences on calcium transport and urinary retention (9, 10, 14). A number of other studies investigating associations between magnesium and potassium and bone health, either individually or in combination, have demonstrated some degree of improvement with higher intake (4–7, 13, 19, 20), and thus the results presented here largely corroborate these findings. However, a recent US study (7) of postmenopausal women found no difference in relative risk of hip and total fractures across quintiles of magnesium intake. Conversely, high magnesium intake (≥422.5 mg/d) was associated with increased falls and wrist or lower arm fractures (7). By contrast, our analyses show a statistically significant reduction in hip fracture risk with moderately high (206–442 mg/d; quintile 4) combined magnesium and potassium intakes and no significant increases in risk of wrist fracture in either men or women in fully adjusted models, although it is acknowledged that the 95% CIs for wrist fracture risk are wide. Differences between the population groups in the 2 studies with respect to genetics, demographic lifestyle, the range of magnesium intakes, and dietary analysis methods [Orchard et al. (7) used FFQs] may explain the discrepancy (27, 35). Also, Orchard et al. (7) did not present their results adjusted for potassium and energy, although they stated that potassium did not modify the associations between magnesium and fracture risk.

The magnitude of the differences seen here is similar to data published by other authors. For example, fully adjusted BUA was 5.3% greater (+3.8 dB/MHz) in Mg+K z score quintile 5 than in quintile 1 for women. This compares to 3.5% and 3.8% increases in lumbar spine bone mineral density (BMD) for premenopausal women in quartile 4 and quartile 1 of dietary magnesium and potassium intakes, respectively (8). Also similar are results from Ryder et al. (19) and Orchard et al. (7) showing that whole-body BMD was 4.0% greater and 3.0% greater, respectively, for women in magnesium quintile 5 than in quintile 1. Tucker et al. (5) show larger differences in BMD across quartiles of combined magnesium and potassium: quartile 4 compared with quartile 1 for women had 12.8% greater lumbar spine BMD, although the limited number in this group and relatively old age (562 women, 69–97 y) could explain the greater differences seen. In terms of the implications of the magnitude of change seen in the current study, previous published data for this cohort showed that a 5-dB/MHz greater BUA was associated with HRT use and that a 20-dB/MHz decline in BUA approximately doubled fracture risk (36), thus demonstrating the relevance of our observations.

Our findings showed no correlation between dietary magnesium intake and serum magnesium concentration for either men or women. Although supplementation studies with magnesium have demonstrated that serum is a suitable biomarker for diet, other studies such as ours found no relation between dietary and serum magnesium; this is likely a reflection of the tight homeostatic control of this cation in the circulation (37–39). However, although serum magnesium concentration was not associated with calcaneal BUA or risk of hip, spine, or wrist fracture in women, a number of statistically significant associations with fracture risk were evident in men, with those in the healthy normal clinical range, 0.7–1.0 mmol/L (33), showing significantly reduced risk compared with those with suboptimal concentrations.

In the United Kingdom, dietary intake of magnesium is mainly provided by fruit and vegetables, cereals, and beverages; potassium is provided by dietary fruit and vegetables, meat, potato, and savory snacks (40). Accurate estimation of dietary nutrient intake is critical to the findings of this type of study. The method used here of quantitative 7-d food diaries has been validated previously and is expected to have provided more precise dietary intake figures than FFQs or 24-h recall methods (27). Indeed, previous United Kingdom EPIC analyses have shown that correlations between potassium intake estimated from food diary data and 24-h potassium excretion were statistically significantly greater than for FFQ or 24-h recall (41). It is reasonable to assume that this validity would also translate to magnesium. The strong collinearity between dietary intake of magnesium and potassium, a likely consequence of magnesium-rich foods typically also being rich in potassium, makes it difficult to differentiate individual effects of these nutrients on bone density. Other studies have considered this to varying degrees, but an appropriate compromise is achieved by presenting data using standardized magnesium and potassium intakes that have been combined and restandardized (5), and thus the inclusion of this data analysis is a strength of this work. Previous use of this method was confined to analyses of BMD measures alone (5), making our additional longitudinal analysis of fracture risk valuable. Hospital admission

data were used to determine fracture incidence, and it is acknowledged this may underestimate incidence, particularly for spine fractures, and could differ between sexes. We used a subset of the EPIC-Norfolk dietary data and, to reduce the potential for bias, included randomly selected participants from the cohort. Magnesium and potassium dietary data were derived from food intake only and therefore may underestimate total nutrient intakes, although supplements consumed by this cohort provide a relatively small contribution to mineral intakes (42); we included calcium and vitamin D supplement use in our models nevertheless. We acknowledge that mineral contributions of drinking and bottled water may be imprecise due to varying concentrations not detailed sufficiently in food composition tables. Although this observational study cannot show causality in effects, this report is, to our knowledge, the first to provide analysis of bone quality and fracture risk by magnesium serum concentration groups in addition to dietary intake in a general population of both men and women.

In conclusion, this study has positively associated dietary magnesium and potassium intake with a quantitative ultrasound measure of bone density status and reduced fracture risk in a mixed United Kingdom population group of men and pre- and postmenopausal women. These results thus support policies to promote a good-quality diet with sufficient magnesium and potassium intake. Clinically normal serum magnesium concentration, compared with suboptimal concentration, has also been shown to be associated with a reduced risk of incident fracture in men. Further study will be required to determine how generalizable the results of these analyses are and to fully understand the relation between intake of these micronutrients, bone health, and osteoporosis.

The authors' responsibilities were as follows—RPGH and AAW: developed the research question; RPGH: performed the data analyses and drafted the manuscript; RNL and AAW: organized data collection; RNL: implemented the record linkage; MAHL: prepared dietary and supplemental data for statistical analysis; K-TK: was principal investigator of the EPIC-Norfolk Study; AAW: had primary responsibility for the final content; and all authors: were involved in interpreting the data, contributed to the writing of the manuscript, and read and approved the final manuscript. None of the authors had a financial or personal conflict of interest relevant to this research at the time of writing.

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