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# Dietary silicon interacts with oestrogen to influence bone health: Evidence from the Aberdeen Prospective Osteoporosis Screening Study

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ABSTRACT

*Background:* Silicon (Si), as $Si(OH)_4$, is derived mainly from plant-based foods. Dietary Si is associated with bone mineral density (BMD) in premenopausal but not postmenopausal women.
*Objective:* To examine the association between Si intake and markers of bone health in middle-aged women and to test for interaction with oestrogen status.
*Methods:* Femoral neck (FN) and lumbar spine (LS) BMD, urinary markers of bone resorption (free pyridinoline and deoxypyridinoline cross-links relative to creatinine, fPYD/Cr and fDPD/Cr) and serum markers of bone formation (N-terminal propeptide of type 1 collagen, P1NP) were measured in a cohort of 3198 women aged 50–62 years ($n = 1170$ current HRT users, $n = 1018$ never used HRT). Dietary Si, bioavailable Si and dietary confounders were estimated by food frequency questionnaire.
*Results:* Mean FN BMD was 2% lower ($p < 0.005$) in the lowest quartile (Q1) compared to the top quartile of energy-adjusted Si intake (Q4) (mean (SD) Q1, 16 (4.0) mg/d; Q4, 31.5 (7.3) mg/d). Energy-adjusted Si intake was associated with FN BMD for oestrogen-replete women only (late premenopausal women ($r = +0.21$, $p = 0.03$); women on HRT [$r = +0.09$, $p < 0.001$]). There was an interaction between oestrogen status and quartile of energy-adjusted Si intake on FN BMD, which was significant after adjustment for confounders ($F = 3.3$, $p = 0.020$), and stronger for bioavailable Si ($F = 5.0$. $p = 0.002$). Quartile of energy-adjusted dietary Si intake was negatively associated with fDPD/Cr and fPYD/Cr ($p < 0.001$) and positively with P1NP ($p < 0.05$).
*Conclusions:* This study suggests that oestrogen status is important for Si metabolism in bone health. Further work is required to elucidate the mechanism.

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## Introduction

The relationship between intake of fruit and vegetables and bone health has been gaining interest over the last decade [1–4]. The hypothesis that fruit and vegetables may influence bone health because of their acid-balancing properties (which can neutralise the net endogenous acid production (NEAP) of the Western diet) has been questioned [5]. Fruit and vegetables contain a number of components (for example vitamins: vitamin C, folate, and vitamin K, minerals: magnesium [Mg], potassium [K], and other bioactive constituents such as flavonoids) that could plausibly influence bone health [6]. The mineral silicon (Si) is a common component of the diet found mainly in plant-based foods (cereal grains and some fruits and vegetables), drinking water (especially mineral water) and some alcoholic beverages, notably beer [7]. Currently there are no recommended intakes for dietary Si and its role in human health is unclear. Animal and cellular studies have shown that Si is required for normal development of bone and connective tissues [8–11] but there are few studies examining dietary Si in humans. Osteoporotic subjects who used the dietary Si supplement, monomethyl silane triol, showed an increase in vertebral bone volume [12] and an increase in femoral and vertebral bone mineral density (BMD) [13]. Bone formation (measured by N-terminal propeptide of type 1 collagen, P1NP) was apparently higher in women after one year supplementation with choline-stabilised orthosilicic acid at daily Si doses of 6 mg and 12 mg. In the Framingham offspring cohort an association between dietary Si and hip BMD was found in premenopausal women, which was less strong in men [14]. The study also found no association between dietary Si intake and BMD for postmenopausal women, which suggests that oestrogen (specifically estradiol) may be involved in the relationship between Si and bone health.

Absorption of Si depends on the food source, with Si in green beans and drinking water being particularly well-absorbed (>50%

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of the total Si content in the food) in contrast to bananas, where Si is poorly absorbed (2% of the total Si content in bananas) [15]. Adjustment for absorption may better reflect the bioavailability of Si. Single nutrient studies may be criticised because the nutrient being tested could be a marker for other nutrients found in similar dietary sources. Foods that are rich in Si are also a good source of the minerals K and Mg, both of which could potentially influence bone health.

The primary aim of this study was to examine the association between dietary Si intake and markers of bone health in early postmenopausal women (both HRT users and non users) in the UK and to determine whether there was an interaction with oestrogen status. We also tested for the association with bioavailable Si (*i.e.* following adjustment for absorption) as an *a priori* planned analysis, and for confounding by other common plant containing minerals, Mg and K.

## Methods

### *Study population*

Subjects were taken from the Aberdeen Prospective Osteoporosis Screening Study (APOSS), involving 5119 women aged 45–54 y, that took place between 1990 and 1993 with further assessment for 3883 of the women between 1998 and 2000 [16] when the women were aged 50–62 y. The women were weighed on both occasions wearing light clothing and no shoes on a set of balance scales (Seca, Hamburg, Germany) calibrated to 0.5 kg. Height was measured with a stadiometer (Holtain Ltd, Crymych, United Kingdom). Information on health, smoking, menopausal status, use of HRT and other medication was also collected. Physical activity level (PAL) was assessed using questions from the Scottish Heart Health Study and MONICA [17] which estimates the numbers of hours in a day doing heavy, moderate, or light activities and sleeping/resting. PAL is the ratio of energy expenditure divided by the basal metabolic rate (BMR), which is calculated from Schofield equations [18]. For women aged 30–59 y, the equation is BMR (MJ/d) = 0.034 × weight (kg) + 3.538. For women aged 60–74 y, the equation is BMR (MJ/d) = 0.0386 × weight (kg) + 2.875. None of the women were taking other treatments for osteoporosis (*e.g.* bisphosphonates) at the time of this study.

Written informed consent was obtained for all the women and the study was approved by Grampian Research Ethics Committee.

### *Dietary intakes*

Dietary silicon intake was assessed at the second visit by a validated semi-quantitative food frequency questionnaire (FFQ) [19] using the UK silicon food database [7]. 'Bioavailable' Si intake was estimated by weighing the Si content of the food according to how well it is absorbed from the food matrix *i.e.* based on the measured increase in mean urinary Si in human subjects following ingestion of particular foods [15,20]. Other nutrients were calculated using the UK Composition of Foods version 5 [21]. A sub-group of women (n = 898) also completed the same FFQ at baseline. For the majority of women there was little change in nutrient intake, although mean energy intake had decreased from 8.1 ± 1.2 MJ to 7.7 ± 1.1 MJ a day (mean ± SD) [19]. Nineteen subjects were excluded because of extreme daily energy intakes (<3.2 MJ and >18 MJ). The net endogenous acid excretion of the diet was estimated using the intakes of potassium (converted from mg to mEq/d) and protein (in g/d) obtained from the FFQ in the equation: estimated NEAP = 54.5 × [protein / potassium] − 10.2 [22,23].

### *Bone mineral density*

Bone mineral density (BMD) of the left femoral neck (FN) and lumbar spine (L2–4) was measured by dual energy X-ray absorptiometry (DXA) using Norland scanners (Cooper Surgical Inc, Trumbull, CT). At the earlier visit (1990–93) the women were measured on the same scanner (XR26); at the current visit (1998–2000), the majority of women were scanned using a Norland XR26 but 12% of women had measurements on the XR36. The *in vivo* precision (coefficient of variation, root mean squared %) for the XR26 scanner in our hands was 2.0% for the lumbar spine (LS), and 2.3% for the femoral neck (FN); and for the XR36 the corresponding values were 1.2% (LS) and 2.3% (FN). Comparison between the XR26 and XR36 using 50 phantom spine measurements for each machine showed that the XR36 consistently gave slightly higher readings (1.258%) and a correction factor was applied using regression analysis to bring these in line with the XR26 measurements [16].

### *Markers of bone metabolism*

A fasted second morning void urine sample was used for the analysis of free pyridinoline cross-links (fPYD) and free deoxypyridinoline cross-links (fDPD) by high-performance liquid chromatography (HPLC), and the results were expressed relative to creatinine (Cr) [24]. Serum samples were analysed for the N-terminal propeptide of type 1 collagen (P1NP) by an enzyme chemiluminescence immunoassay (ECLIA) (Roche Products Ltd, Penzberg, Germany) and for the intact 84 amino acid chain of PTH by an immunoradiometric method (Nichols Institute Diagnostics, San Juan, Capistrano, USA). Serum 25-hydroxyvitamin D [25(OH)D] was measured by HPLC.

### *Statistical analysis*

Analyses were performed using SPSS version 17.0 (SPSS Inc., Chicago). Dietary intakes of nutrients (Si and other nutrients) were adjusted for dietary energy intake by the residual method [25]. Stepwise multiple linear regression analysis was used to determine independent predictors of bone markers and BMD, including energy-adjusted dietary Si as a continuous variable. Dummy variables for menopausal status (premenopausal, perimenopausal and postmenopausal), non use and past use of HRT were used to account for HRT use (with current HRT use as the reference). One-way ANOVA (with Scheffe post-hoc test) and ANCOVA (adjusting for confounding variables age, weight, height, current smoking, physical activity level, social deprivation index, alcohol intake (as quartiles) and dietary confounders, which included dietary vitamin D [with supplements that were regularly taken such as cod liver oil, multivitamins], dietary calcium, dietary magnesium and dietary potassium) were used to examine differences in characteristics and bone health indices between different quartiles of energy-adjusted dietary Si intake. An additional adjustment was made for vitamin D status (as 25-hydroxyvitamin D [25OHD]). As 25(OH)D has a significant seasonal relationship in this population and the subjects were seen throughout the year, the adjustment was done for the analysis of the bone turnover markers only, as they can respond within a season. Chi squared tests were used for the categorical variables. Interaction between oestrogen status and quartile of energy adjusted Si intake was tested by including the multiplicative term of the two variables in addition to the separate variables. For these latter analyses, perimenopausal women were excluded because of the well known variability in circulating estradiol during the perimenopausal period [26].

## Results

In this population of perimenopausal and early postmenopausal women, mean (SD) daily dietary Si intake was 23.3 (7.5) mg ranging from a low of 5.7 to a high of 59.4 mg. Intakes were positively skewed and median intake (interquartile range) was 22.1 (9.1) mg. The main sources of silicon in this population were fruit and vegetables (26%), cereals and cereal products (including bread) (30%), and tea and coffee (18%); with biscuits/cakes adding an additional 8%. These contributed to 82% of the total silicon in the diet with dairy, alcoholic drinks, fish, meat and meat products making minor contributions.

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