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Dietary Calcium Intake in Young Bangladeshi Female Garment Factory Workers: Associations with Serum Parathyroid Hormone Concentrations

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ABSTRACT

The association of low calcium intake with high serum PTH concentrations, increased bone resorption and low bone mass has been studied extensively in elderly women but studies examined the association among young women of childbearing age are scanty. The present study was designed to evaluate the dietary intake of calcium and its association with intact parathyroid hormone (S-iPTH) in young female garment factory worker with hypovitaminosis D. A total of 198 apparently healthy subjects (aged 18-36 years) were randomly selected. Anthropometric and background information was collected. Three days dietary records were used to estimate habitual calcium intake. Serum concentrations of 25-hydroxyvitamin D (S-25OHD) and S-iPTH were measured. The dietary calcium intake was uniformly low. About 60% of subjects failed to meet the lowest level (400 mg day^{-1}) of Recommended Dietary Allowances (RDA) of calcium for Indian adult women. S-iPTH showed a significant inverse relationship with dietary calcium intake ($r = -0.60$, $p<0.001$). We observed that dietary calcium intake and S-25OHD together explained 46% of the variation in S-iPTH and 39% of the total variability in S-iPTH was explained by the variation in dietary calcium intake alone. Low calcium intake could reduce the bone accretion and may increase the risk of hyperparathyroidism, osteoporosis and fracture burden in these subjects.

Key words: Bangladesh, calcium intake, parathyroid hormone, women

INTRODUCTION

Adequate calcium intake is very important for optimal bone health. Calcium intake has been identified as one of the several factors that affect high peak bone mass which is inversely associated with bone fragility (Matkovic, 1992; Heaney *et al*., 2000). A number of studies indicated that calcium, vitamin D and PTH participate in bone metabolism.

Our earlier studies have shown a high prevalence of vitamin D insufficiency and low dietary intake of calcium among different groups of Bangladeshi women (Jahan and Hossain, 1998; Islam *et al*., 2002, 2006, 2008). Due to exclusive homebound lifestyle or covered-up style traditional dresses, reduction in endogenous synthesis of vitamin D together with inadequate dietary intake of calcium, Bangladeshi women may be especially prone to osteoporosis. However, so far, no data are available on bone health and the prevalence of osteoporosis and fracture in the population

of Bangladesh. With increasing trend of life expectancy and a high prevalence of nutritional deficiencies in low income women, the increase in morbidity due to osteoporosis might strain the health care system of Bangladesh in the near future.

The export-based garment industries in Bangladesh are dominated by female labour and have employed more than 4.0 million young women of low socio-economic standing. Due to rapid growth of garment industries in Bangladesh the number women workers is increasing everyday. These young women typically work dawn to dusk in an over-crowded, congested and poorly ventilated sub-standard environment. We reported a high prevalence of vitamin D insufficiency and uniformly low bone mass in the subjects of the present study (Islam *et al*., 2008).

We know calcium plays an important role in bone health and similarly bones play a key role in calcium homeostasis and maintain normal blood calcium levels. In fact, there is no controversy regarding calcium-bone relationship. Unfortunately, calcium intake among Bangladeshi women and other South Asian populations is typically well below the present recommendation (Jahan and Hossain, 1998; Islam *et al*., 2002; Harinarayan *et al*., 2007). As the data are scanty on calcium intake and bone metabolism among South Asians so that question remain whether this lower intake is adequate or inadequate in terms of bone health.

The wide variation in S-PTH concentrations could be linked to different levels of dietary intake of calcium since serum calcium regulates PTH release and other associated factors as well (Heaney, 2002). There is no universally accepted cut-off value for S-PTH available to identify the demarcation line that may be detrimental. However, the elevated S-PTH concentrations were found to be associated with increased bone loss in older women (Lips, 2001). The association of reduced intake of calcium with high concentrations of serum PTH, increased bone resorption and low bone mass was reported also in elderly subjects (Jorde *et al*., 2000; Nakamura *et al*., 2009). The beneficial effect of calcium intake on S-PTH concentration was shown in one study which indicated a lower bone resorption in response to higher calcium intake (Karkkainen *et al*., 1998). In fact, a number of studies have been carried out on the relationship between S-PTH concentration, S-25OHD concentration and calcium intake in elderly women but data are scarce in young women.

Little is known on dietary habits and calcium intake of female garment factory workers in Bangladesh. The aim of the present study was to evaluate dietary calcium intake and to find out its association with S-iPTH in a representative sample of low income young women with exclusive indoor lifestyle.

MATERIALS AND METHODS

Study setting and subjects: This study was carried out in an export-oriented garment factory situated in Mirpur in the city of Dhaka. The factory is located in a modern multi-storied building with better working facilities belonging to Standard Group Bangladesh, who maintain a high quality working environment for their workers. A total of 198 subjects aged 18-36 years were randomly selected from the garment factory. The subjects of this study were mainly young women from low-income rural families who migrated to the city for a job at least two years before. They live in low-cost accommodations with no indoor supply of water, no sanitary latrines and densely-packed sleeping arrangements. They work long hours (dawn to dusk), 7 d/week. The eligibility criteria to include the subjects in this study was no history of serious medical conditions, no history of medication known to affect bone metabolism, no current pregnancies, no lactation within the previous three years and lived in the city for at least two years. Our first approach was to contact the Chief Medical Officer of the Standard Group, to explain the purpose of our study and to ask for

their co-operation. We contacted the subjects through the Chief Medical Officer and explained to them the objectives of the study in an understandable way and asked for their written consent to participate. The subjects received a reasonable compensation for their loss of one day's work to participate in the study. On weekly working days, a small group of about ten subjects were brought in a minibus to the Centre for Nuclear Medicine and Ultrasound, Sir Salimullah Medical College, Mitford, Dhaka, where the background information, anthropometric measurements and blood samples were collected sequentially. The time subjects spent outside daily was recorded. The study was approved by the ethics committee of the Faculty of Agriculture and Forestry, University of Helsinki. During the field study in Bangladesh, we also followed the ethical guidelines of the University of Dhaka.

Anthropometric measurements and other data: We measured height, weight, biceps, triceps skinfolds, mid-upper arm circumference (MUAC); head, waist and hip circumference. Standing height was measured with a wall-mounted scale to the nearest 0.5 cm. Body weight was measured without shoes and with light clothing on a portable weighing scale to the nearest 0.5 kg. We used the classifications of Body Mass Index (BMI) [weight (kg)/height (m^2)] recommended by the World Health Organisation (WHO, 1995). A Harpenden calliper was used to measure skinfold thicknesses (SFT) and a measuring tape was used for the measurements of MUAC, waist and hip circumference. A complete questionnaire was developed, regarding necessary information such as name, age, address, education level, parity, total physical activity level (factory-based and leisure-based activity), out-door clothing, frequency and duration of sunshine exposure, use of sunlight protection cream, medical history, use of vitamin D and calcium supplements within three months etc.

Dietary information: Information regarding food consumption was collected for three days. The consumption of each food item was closely monitored and recorded by using food record method. The subjects were asked to maintain their normal dietary practice and the fieldworkers were present at the garments factory during their meals. The amount food was measured using measuring spoons, cups, glasses and plates of different sizes, collected from the Institute of Nutrition and Food Science (INFS), Dhaka University (DU) which were used earlier in Nutritional Surveys of Bangladesh. The subjects were requested to measure their food by themselves in front of the female fieldworkers using the standardized measures provided. The fieldworker wrote down all the information about foods and beverage taken. Subjects were instructed to report the food taken between meals and to describe the portions in terms of measures provided which were also recorded by the fieldworkers during following visits. The recorded food items were coded and weight of the food item was measured in grams using Food Code List. The equivalent weight of the raw food was calculated using a conversion table for Bangladeshi foods formulated at the INFS (Ali and Pramanik, 1991). The average daily energy, calcium and other nutrient intakes were calculated using computerised version of the Bangladesh food composition database developed by the INFS, DU. The intake of betel leaves (Piper betel) and Areca nut (Areca catechu) and $Ca(OH)_2$ were not included in the present study because the present food composition database does not contain these food items.

Laboratory measurements: Fasting blood samples were collected between 8:30 and 10:00 a.m. using disposable syringes. Serum was separated and collected in tubes and preserved at -20°C.

Finally, the serum sample was transported to Helsinki in a special type of container with dry ice and preserved in the freezing room of the Division of Nutrition, Department of Food and Environmental Sciences (Nutrition), University of Helsinki at -20°C temperature until analysed. The S-iPTH concentration was measured with a commercial two-site immunoenzymometric assay (OCTEIA, IDS), with 10-65 ng L^{-1} as a reference range. Intra-and inter-assay CV for iPTH were 3.8 and 5.7%, respectively. We defined secondary hyperparathyroidism as S-iPTH levels >65 ng L^{-1}. S-25OHD was measured from all fasting samples with an OCTEIA immunoenzymometric assay (IDS, Boldon, UK). The intra-and inter-assay CV were 4.9 and 6.8%, respectively. Reproducibility was ensured by adhering to the Vitamin D External Quality Assessment Scheme, DEQAS. Standardized Concentrations of S-25OHD were provided. The laboratory analyses were carried out at the Department of Food and Environmental Sciences, University of Helsinki.

Statistical analyses: The statistical analysis was completed using SPSS version 12.0 (SPSS Inc, Chicago, IL, USA). Data are presented as means ± SDs. Logarithmic transformations (ln) were used when necessary to normalize the distribution. Group comparison was made by the t-test. Correlations between variables were estimated by using Pearson's coefficient of correlation. The association between S-iPTH concentrations and dietary calcium intake were studied both by linear and non-linear regression models. The determinants S-iPTH concentrations were studied in the regression models. Age, BMI, dietary calcium intake and S-25OHD were used in the regression models to study the determinants of S-iPTH concentration and p values <0.05 were considered significant.

RESULTS

Basic characteristics of the subjects: A summary of the descriptive, physical, biochemical characteristics and the information on the prevalence of vitamin D insufficiency, bone mineral density and bone mineral contents of the study population have been published elsewhere (Islam *et al*., 2008). The subjects were mainly young women and more than 73% of them were between 18-25 years old. We identified 4 levels of education (category 1 = illiterate, category 2 = primary level, category 3 = secondary level and category 4 = above secondary level) among subjects. The distribution of the subjects in different levels of education were nearly 10% in category 1, 62% in category 2, 20% in category 3 whereas only 8% of the subjects had category 4 level of education. No subject reported any supplementation of vitamin D or calcium. The mean physical activity level of the subjects showed that a total of 13.2 h/day were occupied with some physical activity of which factory-based physical activity attributed to about 80.5% and less than 20% was home-based leisure and other physical activity. The mean monthly income of the subjects was USD 42.8.

Dietary intake: The diet composition and food records displayed a lacked variety of food consumed by the subjects. The diet of subjects mostly based on staples (rice and bread), with a small amount of animal products (such as meat, fish, eggs), milk and dairy products, fruits and vegetables. We observed that milk and dairy products (one of the major sources of dietary calcium) and fruits were absent in the diets of large proportion of the subjects (57%). The mean daily calcium intake was low and mainly contributed by cereal products (Fig. 1). Approximately 75% of the subjects consumed on average less than 600 mg day^{-1} (the current recommendation of calcium by the Indian Council of Medical Research, ICMR) and more than 13% of the subjects consumed less than 200 mg day^{-1}. More than 90% of the subjects failed to meet the recent dietary reference intake for calcium

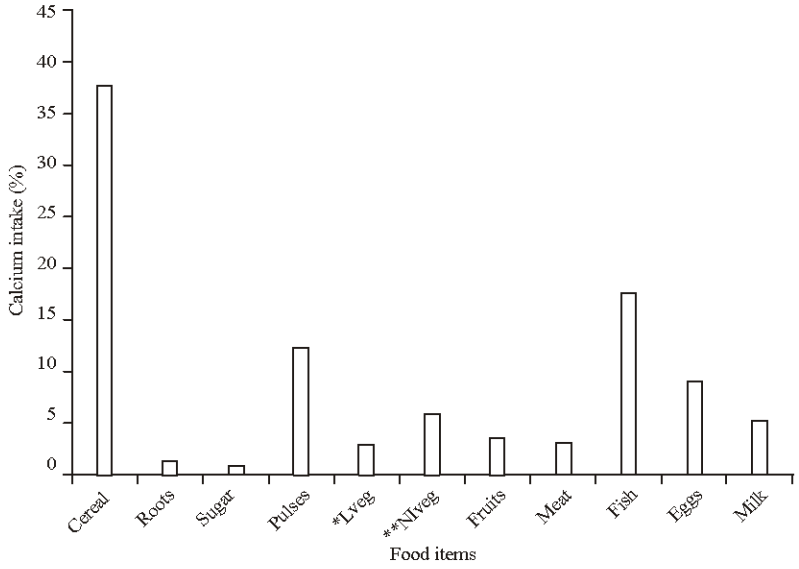

Fig. 1: The proportional contribution of different food groups to the total mean daily calcium intake in the subjects, *Leafy vegetables, ** Non-leafy vegetables

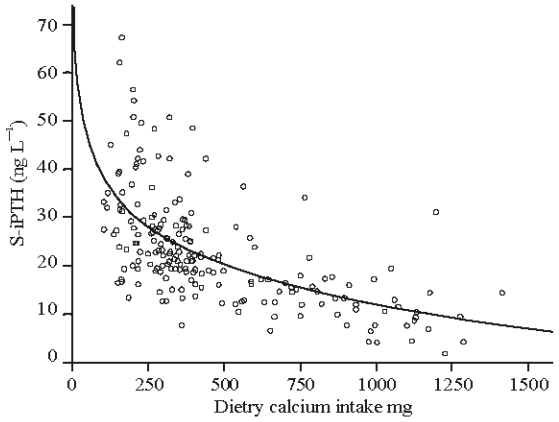

Fig. 2: Association between dietary calcium intake and serum intact parathyroid hormone concentrations in female garment factory workers (n = 198) in Bangladesh

proposed by the Institute of Medicine, USA (Ross *et al*., 2011). Daily intake of energy and nutrients is given in Table 1. The energy intake of the subjects was found to cover only 69.5% of the recommended level. The dietary source of energy was about 17% from proteins, 6% from fats and 75% from carbohydrates. In the present study, we observed that dietary calcium intake correlated with protein intake ($r = 0.19$, $p<0.05$).

Serum 25OHD, dietary calcium intake and iPTH concentration The high prevalence of vitamin D inadequacy in these subjects has been described elsewhere (Islam *et al*., 2008). Dietary calcium intake correlated inversely with S-iPTH (r and -0.60, $p<0.001$). Association between dietary calcium intake and S-iPTH concentrations is presented in Fig. 2. In linear regression analysis, dietary calcium intake (ln transformed) alone explained 39% of the variance in S-iPTH ($r = 0.64$, $r^2 = 0.39$, S-iPTH = 93-12*lnCa-intake, $p<0.0005$). Dietary calcium intake and

Table 1: Daily intake of energy and nutrients by the subjects in relation to RDA* for Indian adult female (Rao, 2010)

	Total intake			
Energy and nutrients	Mean	SD	RDA	RDA (%)
Energy intake (kcal)	1547.0	489.0	2225.0	69.5
Carbohydrate (g)	289.2	94.0	300.0	96.4
Protein (g)	53.6	20.8	55.0	97.5
Fat (g)	9.9	7.8	20.0	49.5
Iron (mg)	25.1	14.1	30.0	83.6
Vitamin C (mg)	65.2	35.6	40.0	163.0
Calcium (mg)	465.0	299.0	600.0	75.5
Calcium density (Ca mg/1000 kcal)	321	227	-	-

*Recommended Dietary Allowances

S-25OHD together explained 46% of the variation in S-iPTH ($r = 0.67$, $r^2 = 0.46$, $p<0.0005$, S-iPTH = 124-9*lnS-25OHD-11*lnCa-intake).

DISCUSSION

The high prevalence of energy and nutrient deficiencies has been identified as an important concern for public health in Bangladesh. Several investigators identified the women, children and elderly as the most vulnerable groups. We observed energy intake in the subjects of the present study was much lower than recommended levels and similar to earlier reported studies in Bangladesh (Torres *et al.*, 1990; Ahmed *et al.*, 1998; Islam *et al.*, 2004; Kabir *et al.*, 2010) but physical activity level of the subjects of the present study was much higher compared with subjects of those studies. As far as we know, the present study is the first study focused on the habitual dietary intake and its effect on S-PTH concentrations in young low income Bangladeshi women.

According to the Food and Nutrition Board for female adults in Asia, the recommend daily allowance (RDA) of calcium is 400-500 mg day^{-1}. The RDA for calcium recommended by the Indian Council of Medical Research (600 mg day^{-1}) is a bit higher than this level (Rao, 2010). The high prevalence of inadequate intake of calcium has been reported earlier in several studies in Bangladesh (Torres *et al.*, 1990; Ahmed *et al.*, 1998; Islam *et al.*, 2003; Islam *et al.*, 2004; Kabir *et al.*, 2010). Our study supported that a low dietary intake of calcium persists in low income Bangladeshi female.

A low level of dietary intake of calcium frequently observed in low income countries where diets are not rich in milk and dairy products. Our study also indicated a low consumption of milk and dairy products in the subjects. However, we observed a small proportion of the participants frequently consumed Areca nuts (*Areca catechu*) and betel leaves (*Piper betel*) with lime [$Ca(OH)_2$] which might contribute to calcium intake. Unfortunately, our food composition database did not contain these items. Further, the high content of phytate in Indian diet has been observed as a barrier for calcium absorption in the gut (Harinarayan *et al.*, 2007).

Parathyroid hormone is responsible for maintaining normal levels of serum calcium and phosphate and PTH is regulated through concentrations of calcitriol and serum calcium. Interestingly we found a strong inverse correlation between dietary calcium intake and S-iPTH suggests that calcium intake in the subjects of the present study was far from sufficient. This observation is an accord with the study by Karkkainen et al. 1998 who found that those with a low calcium intake had higher S-PTH concentrations than those with a high intake in a study in

postmenopausal women. These also comply with the findings of earlier studies which showed that serum PTH concentration is inversely associated with calcium intake (Kinyamu *et al*., 1998; Steingrimsdottir *et al*., 2005; Adami *et al*., 2008; Gunnarsson *et al*., 2009).

In an earlier report in the same subjects we found a strong inverse correlation between S-25OHD and S-PTH (Islam *et al*., 2008). When introducing both calcium intake and S-25OHD in a regression model, we found that they together explained 46% of the variance in S-PTH. Calcium intake alone, however, explained 39% of the variance, indicating that calcium intake is more important than vitamin D status. This has not been found in other studies (Kinyamu *et al*., 1998; Steingrimsdottir *et al*., 2005; Adami *et al*., 2008; Gunnarsson *et al*., 2009). Low calcium intake has been proposed previously to aggravate vitamin D deficiency through increased catabolism of 25-hydroxyvitamin D (Lips *et al*., 1991; Lips, 1998).

It has been shown that Asian women have lower bone mineral density than whites (Marquez *et al*., 2001; Wu *et al*., 2003; Mehta *et al*., 2004). Reports indicated that due to low peak bone mass, early menopause and other underlying causes, osteoporotic fractures occur 10-20 years earlier among the south Asians than among western Caucasians (Shatrugna *et al*., 2005). The observed low calcium intake, coupled with long periods of elevated calcium requirements as well as high S-iPTH concentrations, high prevalence of vitamin D insufficiency and low peak bone mass could lead to a very high risk of osteoporosis in these subjects. The findings of the present study could be of serious concern for the public health of Bangladesh as the subjects represent a large homogeneous group (more than 4 million female workers) as the recent industrial growth in Bangladesh has been driven by a booming garment industry.

Limitations to the study: Earlier studies reported that all dietary assessment methods are imprecise. Calcium and other nutrient intake measured by different dietary assessment methods may have considerable measurement error leading to over-or underestimation. In addition, due to some technical limitations, we could not measure calcium content of Areca nuts (*Areca catechu*), betel leaves (*Piper betel*) and lime [$Ca(OH)_2$] which might contribute to total calcium intake. However, a cross-sectional study like ours is not enough to demonstrate the causality, the relationship between vitamin D status, calcium intake and the interaction between these two variables and the serum PTH concentration is a strong indication of the comparative importance of these nutrients. Therefore, intervention studies in women of different age groups and socio-economic classes are required to further address this issue to generate more representative results.

CONCLUSION

Osteoporosis represents a major public health issue as the frequency of osteoporotic fracture is rising dramatically in both developed and undeveloped countries. The long term insufficiency of both calcium intake and vitamin D status in these subjects might be associated with increased risk of hyperparathyroidism and could increase fracture burden due to osteoporosis many-fold. Due to the similar exclusive indoor lifestyle, occupation, social background, living accommodation shared by more than 4 million female garment workers of Bangladesh, this situation could be a major public health concern. Therefore, both health professional and health policy makers should pay attention on this "silent epidemic" and take active measures to educate young Bangladeshi women about the importance of adequate calcium intake, vitamin D status and bone health. Extensive dairy farming, shrimp, fish and poultry farming and home gardening could be encouraged to increase the easy and low-cost availability of calcium-rich food items to ensure an adequate supply of calcium in the daily diet.

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CONTRIBUTORS

MZI and CL-A designed the study, secured funding. MZI was the principal investigator of the study, managed data collection and laboratory analysis of serum samples, involved in data entry, statistical analysis and manuscript write-up. MK participated in statistical analysis. MK and CL-A checked the manuscript thoroughly and approved the final version. CL-A and MZI are guarantors of the study. None of the contributing authors had any financial or personal conflicts of interest.

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