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Calcium Intake and Bone Mineral Variables among Adolescent Schoolgirls in Rural and Urban Areas of Sri Lanka

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ABSTRACT. *Adolescence is a critical period for bone health because the accumulation of bone mass peaks during the teenage years. Failure to attain optimal peak bone mass can increase an individual's risk of osteoporosis and fractures. The aims of the present study were to determine calcium intake of adolescence girls in rural and urban areas and to determine the relationship between dietary intakes of calcium and Bone Mineral Density (BMD) / Bone Mineral Content (BMC). A sample of 652 school girls aged 11 – 16 years residing in Colombo Municipal Council and 529 school girls from Pannala Education Division were selected. Three-day diet diaries were used to measure the dietary energy, calcium intake and skeletomuscular nutrienst. Three-day physical activity records were used to determine physical activity level. BMD and BMC were determined using Dual energy X–ray Absorptiometer scanning the middle finger of the hand. Mean calcium intake of urban and rural adolescents were 488.4 (SD 180.3) mg/day and 364.9 (SD 160.8) mg/day, respectively. The calcium intake of adolescents was significantly lower (P<0.05) than the Recommended Daily Allowances (RDA) (1000 mg). Ninety eight percent of adolescents girls aged 11-16 years did not achieve the RDA for calcium. High calcium intake group had significantly higher BMD and BMC (P<0.0001). The calcium intake showed a significant positive correlation with BMD (r = 0.14, P = <0.01) and BMC (r = 0.17, P = <0.01). The proportion of adolescent girls not achieving the RDA for iron, zinc, magnesium, vitamin A and vitamin C were 89.6, 93.9, 93.9, 96.2 and 54.8% respectively. In conclusion, low calcium intake together with other nutritional inadequacies affects BMD and BMC in adolescence.*

INTRODUCTION

Adolescence is a critical period for lifetime bone health because the accumulation of bone mass peaks during the teenage years. Failure to attain optimal peak bone mass can increase an individual's risk of osteoporosis and fractures (Mora and Gilsanz, 2003). Osteoporosis is of particular concern for women because approximately one third of women aged more than 65 yeas suffer from osteoporosis. Over a life time, women may lose up to a total of 35% of cortical bone mass and 50% of trabecular bone, whereas men may lose only two third of

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these amounts (Riggs and Melton, 1986). Peak bone mass and subsequent bone losses are important determinants of osteoporosis later in life (Hansen *et al.,* 1991).

Several nutrients contribute to bone health. However, calcium is particularly important during rapid growth that occurs in adolescence. Protein, magnesium, zinc, cupper, iron, phosphorus and vitamin D, A, C and K are the other nutrients required for normal bone metabolism. Approximately 80-90% of Bone Mineral Content (BMC) is composed of calcium and phosphorus (Ilich and Kerstetter, 2000). One of the major determinants of bone strength is Bone Mineral Density (BMD). It is determined by either fixed factors (genetic disposition, gender and health status) or modifiable factors (nutrition, physical activity, smoking and medication) (Hustmyer *et al.*, 1994). Forty five percent of the adult skeleton is built and enlarged during adolescence (Christiansen *et al*., 1979). Evaluation of child's bone mass is the best way to evaluate his or her risk of developing osteoporosis (Nordin, 2000).

Functional calcium absorption is highest during early adolescence. The average daily accumulation of calcium in adolescent girls is 2.87 mmol/d and increases to 9.23 mmol/d during the peak growth spurt. Researches have suggested that adolescence could be an important time to optimize bone density by consuming adequate dietary calcium (Matkovic, 1991). Obtaining adequate calcium intakes from childhood through early adulthood is central to osteoporosis prevention efforts aimed at influencing peak bone mass in females (Heaney and Matkovic, 1995). World Health Organization (WHO) recommends 1300 mg/day calcium intake for the adolescent girls (FAO/WHO, 2001) but according to the Sri Lankan recommendation, calcium requirement for the adolescents is 500 – 600 mg/day (Ministry of Health Care and Nutrition of Sri Lanka, 2000). The recommended levels for adolescents in Sri Lanka have been recently revised to 1000 mg/d.

Dietary calcium intake has been related to the achievement of peak bone mass in adolescence (Ott, 1990). The relationship between bone mass and spontaneous calcium intake is controversial due to various ages studied, but several studies indicated that dietary calcium intake is a predictor of bone mineral density in adolescents (Chan, 1992; Kardinaal *et al*., 1999; Carter *et al*., 2001).

There have been no studies conducted to determine the relationship between calcium intake and BMD in adolescents in Sri Lanka, where dietary calcium intake is probably lower than developed countries. The aims of the present study were to assess calcium and other skeletomuscular nutrient intake of rural and urban adolescent schoolgirls and to determine the relationship of BMD/BMC with dietary factors and physical activity level (PAL). BMD and BMC were determined using dual energy x-ray absorptiometry (DEXA) since it offers rapid, accurate and precise assessment of bone components with low radiation exposure (Mazess *et al.,* 1990).

MATERIALS AND METHODS

Subjects

This study was conducted as a cross sectional study. One thousand two hundred and fifty-five adolescent schoolgirls aged 11–16 years and attending either national or provincial schools situated in Colombo Municipal Council (CMC) and Pannala Education Division were invited to participate in this study. Public and national schools were randomly selected from a list of schools in each educational division to obtain a cross section of socioeconomic backgrounds. Schoolgirls who had chronic systemic diseases, who used drugs affecting bone minerals or engaged in sport activities > 7 h per week were excluded from the study. The final sample had 652 urban and 529 rural subjects from Colombo and Pannala schools, respectively. Informed consent was obtained from the parents of the subjects and the study was approved by Ethical Review Committee of Sri Lanka Medical Association.

Dietary assessment

Three-day diet diary (two weekdays and one weekend day) was used to assess calcium and other nutrient intake of the subjects. Of the 1181 subjects recruited, 240 rural subjects and 212 urban subjects returned the diet diaries (response rates: rural 43.3% and urban 30.8%). One hundred and seventy four and 86 completed food records from urban and rural sectors (final response rates: urban 25.3% and rural 15.2%), respectively were analyzed for nutrient intakes using FoodBase 2000 version 2 (Brain Chemistry and Human Nutrition Institute, UK) computerized food composition tables. Energy, protein, calcium, iron, zinc, magnesium, vitamin A and vitamin C intakes were determined using FoodBase 2000.

Physical activity

Three-day physical activity record (two weekdays and one weekend day) was used to assess PAL. Total Energy Expenditure (TEE) was calculated using Basal Metabolic Rate (BMR) (Schofield, 1985) and activity factors (WHO, 1985) of all activities reported. One hundred and seventy four and 178 completed physical activity records from urban and rural sectors respectively, were analyzed to assess PAL.

Measurements of BMD and BMC

BMD and BMC in the mid-finger of non dominant hand of all subjects were measured using accu DEXA (Schick, 7100, USA) which was validated against central DEXA previously (Lekamwasam *et al*., 2007). Coefficient of variance (CV) of measurements for the same subject was <2%. Six hundred and fifty two and 529 BMD/BMC measurements from urban and rural sector respectively were used for the final analysis.

Anthropometric measurements

Weight and height were measured using an electronic digital scale (Salter, UK) and stadiometer (Invicta, 0955, UK), respectively. BMI (Body Mass Index) for age was calculated using Epi-info version 3.4 (Center for Disease Prevention and Control, USA). Six hundred and fifty two and 529 subjects were measured for anthropometry from urban and rural sectors.

Statistical analysis

Adolescent school girls were divided into two groups based on their calcium intake, physical activity level (using median value) and BMI (using underweight cutoff). Student's t test was used to identify significant differences between comparison groups for means. One-sample t-test was used to determine significant differences between dietary intakes and Recommended Dietary Allowances (RDAs; Medical Research Institute, 2000). Prevalence rates in rural and urban sectors were compared using Chi-square test. Pearson's correlation co-efficient was used to determine association between dietary, anthropometric and physical activity variables and bone mineral variables in a sub-sample who completed diet diary and activity record (n=259). Statistical analysis was performed using SPSS version 13.0.

RESULTS

Mean dietary calcium intake of the study population was 426.6 (SD 170.5) mg/day. Mean calcium intakes of rural and urban adolescents were 364.9 (SD 160.8) mg/day and 488.4 (SD 180.3) mg/day, respectively (Table 1). Urban adolescents had significantly higher calcium intake than rural counterparts (P<0.01). Only 2% of adolescents in urban areas and 1% of adolescents in rural areas had met the RDA for calcium (Table 1). The proportion of urban adolescent schoolgirls not achieving the RDA for iron, zinc, magnesium, vitamin A and vitamin C were 89.5%, 91.5%, 93.0%, 100% and 59.3% respectively. Percentages of rural adolescents not meting the RDA for above nutrients were 89.7%, 96%, 94.8%, 92.5% and 64.9% respectively. Percentage of adolescents not meting the RDA for energy was approximately similar in rural and urban areas.

Table 1. Mean (SD) daily nutrient intake of adolescents in rural and urban areas

Nutrients	RDA	Total group (n=259)	Rural (n=86)	Urban (n=174)	Rural vs Urban
Energy (kcal)	2100	1589 (432)	1569 (404)	1609 (459)	NS
Calcium (mg)	1000	426.6 (170.5)	364.9 (160.8)	488.4 (180.3)	P<0.01
Iron (mg)	18	12.3 (6.4)	12.9 (7.1)	11.7 (5.8)	NS
Zinc (mg)	7.8	5.6 (1.9)	6.6 (1.4)	4.7 (2.4)	NS
Magnesium (mg)	230	132.4 (58.5)	124.9 (69.8)	139.9 (47.2)	NS
Vitamin A (µg)	600	282.14 (147.5)	198.5 (114.4)	365.8 (180.6)	NS
Vitamin C (mg)	40	53.3 (64.1)	56.2 (77.7)	50.4 (50.6)	P<0.01
Protein (g)	67	46.5 (17.1)	49.3 (20.3)	43.8 (14.0)	NS

* P <0.01, t test (urban vs rural)
NS – not significant

Table 2 shows the percentage of schoolgirls consuming less than RDA for energy and selected nutrients. In urban sector, significantly greater percentage of schoolgirls consumed vitamin C less than RDA compared with those in urban sector.

Table 2. Percentage of rural and urban adolescent schoolgirls consuming less than RDA for energy and selected nutrients

Nutrient	%<RDA	%< RDA	P*
Protein	96.5	88.8	0.426
Energy	89.5	85.1	0.312
Calcium	98.9	97.8	0.317
Iron	89.5	89.7	0.988
Zinc	91.9	96.0	0.105
Magnesium	93.0	94.8	0.565
Vitamin A	100.0	92.5	0.155
Vitamin C	59.3	64.9	0.001

* Chi-square test (rural vs urban)

Table 3 shows the BMC, BMD, anthropometric data and PAL of rural and urban adolescent schoolgirls. Rural adolescent schoolgirls had significantly lower BMD and BMC values compared with urban adolescent schoolgirls (P < 0.01).

Table 3. Mean (SD) BMD, BMC, anthropometric data and PAL among rural and urban adolescent girls

Variable	Rural	Urban	Rural vs Urban
Age (years)	14.1 (1.4)	13.8 (1.7)	NS
Height (m)	150.0 (7.4)	52.4 (6.3)	NS
Weight (kg)	40.7 (9.6)	44.2 (10.3)	P<0.01
BMI (kgm^{-2})	17.9 (3.4)	18.8 (3.7)	NS
PAL	1.4 (0.1)	1.4 (0.1)	NS
BMD(g/cm^2)	0.375 (0.07)	0.403 (0.06)	P<0.01
BMC (g/cm)	0.999 (0.33)	1.116 (0.30)	P<0.01

Table 4. Effect of calcium intake, PAL and BMI on BMD and BMC

	Mean (SD) BMD	Mean (SD) BMC
Calcium intake (n=259)		
Low ≤427 mg/day	0.373 (0.07)	0.973 (0.30)
High >427 mg/day	0.412 (0.07)	1.166 (0.31)
P	<0.0001	<0.0001
PAL (n=352)		
Low ≤ 1.37	0.391 (0.07)	1.053 (0.32)
High >1.37	0.393 (0.07)	1.086 (0.32)
P	0.904	0.407
BMI (n=352)		
≤18.5 kgm^{-2}	0.372 (0.07)	0.981 (0.29)
> 18.5 kgm^{-2}	0.418 (0.07)	1.183 (0.32)
P	<0.0001	<0.0001

Adolescent schoolgirls who had lower calcium intake and lower BMI had significantly lower BMD and BMC compared with those who had higher calcium intake and higher BMI.

BMD and BMC were not significantly different in two physical activity subgroups (Table 4).

There was a significant positive correlation between daily calcium intake and BMD (r = 0.15, P < 0.01) and BMC (r = 0.17 < 0.01) in combined sample (n= 259). These correlations are illustrated in Figures 1 and 2.

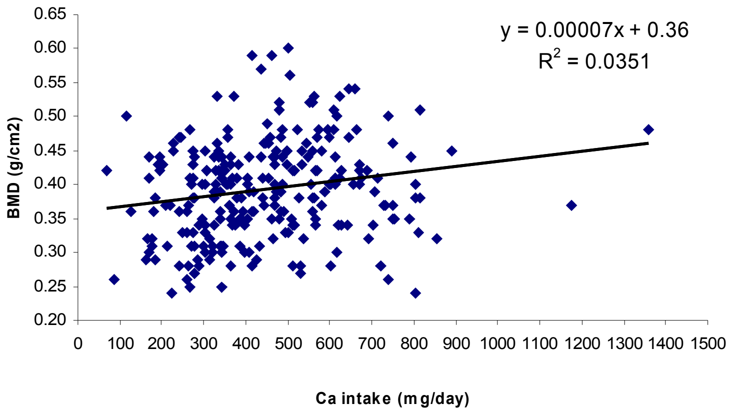

Figure 1. Relationship between calcium intake and BMD among adolescent girls (n=259)

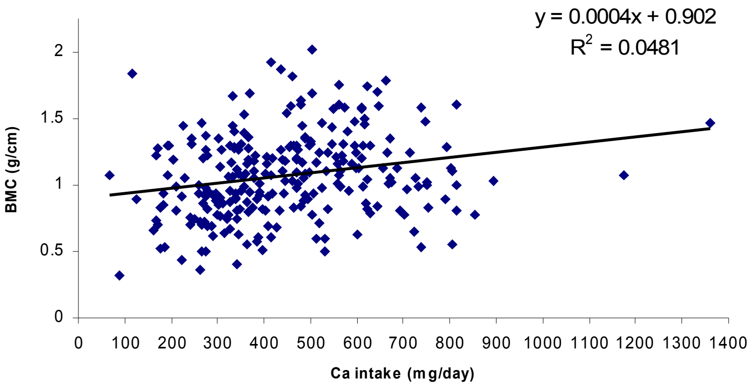

Figure 2. Relationship between calcium intake and BMC among adolescent girls (n=259)

Table 5. Pearson's correlation coefficients of calcium intake and other variables between BMD/BMC of adolescents school girls (n=259)

Variables	Correlation Coefficients (r)	
	BMD	BMC
Unadjusted		
Calcium intake	0.14*	0.17*
Energy intake	0.17*	0.19*
TEE	0.30*	0.30*
PAL	0.03	0.01
Age	0.58*	0.33*
Weight	0.49*	0.52*
Height	0.57*	0.60*
BMI	0.31*	0.31*
Calcium intake adjusted for;		
Energy intake	0.10	0.13*
TEE	0.20*	0.24*
Age	0.24*	0.27*
Weight	0.14*	0.17*
Height	0.06	0.09
BMI	0.19*	0.23*
† Age and BMI	0.15*	0.18*
All of above factors	0.11	0.14*

* P< 0.01

† Combination of other two factors not showed significant relationship with BMD and BMC.

Energy intake, TEE, age, weight, height and BMI also showed significant positive correlations with BMD and BMC (Table 5). Further, calcium intake had a significant correlation with BMD when adjusted for total energy expenditure, age, weight or BMI. Statistical significance of the correlation between calcium intake and BMD disappeared when adjusted for energy intake or height. Calcium intake had a significant correlation with BMC when adjusted for all single factors except for height. Age and BMI adjusted calcium intake has a significant correlation with both BMD and BMC. When adjusted for all factors, calcium intake showed a relationship with only BMC.

DISCUSSIONS

This study was carried out to determine dietary calcium intake and other skeletomuscular nutrient intake, BMD and BMC of rural and urban adolescent schoolgirls and to determine the relationship between nutrient intake and the bone variables. In this study we determined BMD and BMC of 1181 of adolescent school girls and adequacy of calcium and other skeletomuscular nutrient intake of 259 adolescent school girls. Mean calcium intake was not satisfactory (426.6 mg/day) compared with RDA (1000 mg/day). Calcium intake was low among the rural adolescent girls compared with urban adolescent girls. About 99% in rural and 98% in urban adolescents did not meet the RDA for calcium. Similar finding was observed in a recent study done in Sri Lanka which indicated that calcium intake varies between 361- 386 mg/day among adolescents of age between 13 -18 years (MRI, 2004). According to a study conducted in the estate sector in Sri Lanka, the calcium intake was 326-386 mg/day (Jananthan & Chandrasekara, 2005). In developing nations overall calcium intake was found to be relatively lower (244 mg/d) than that of developed world (850 mg/d) (Nordin, 2000). Low calcium intake reported in adolescent schoolgirls is a disadvantage for bone mass accretion during the critical growth period.

We observed lower total energy and protein intake than the RDA in both rural and urban sectors. This reflects overall insufficient or low quality food intake by adolescent schoolgirls. This could lead to poor micronutrient intake, which is supported by the low intake of iron, zinc, magnesium and vitamin A by the study population. Calcium, magnesium, zinc and vitamin A consumption was lower among rural adolescents girls compared with urban adolescent girls. Lower energy intake by rural adolescent girls compared with urban adolescent girls explains the lower micronutrient intake by former.

In the present study, adolescent girls had mean BMD and BMC of 0.389 g/cm^2 and 1.110g/cm respectively. Rural adolescent girls had lower BMD and BMC values than that of urban adolescent girls. In this study we assumed BMD and BMC of mid finger reflect total body BMD and BMC as the portable DEXA used in this study had been validated against central DEXA which measures total body BMD. Also, BMD values of mid finger were similar to BMD values of forearm among Northern Iris adolescent girls (0.367 g/cm^3) (Boot *et al*., 1997).

The present study suggests that calcium intake *per se* play a crucial role in BMD since both subgroups of girls with different calcium intake exhibited different BMD values. We also found a significant independent relationship between dietary calcium intake and bone parameters among adolescent girls. It can also be suggested that lower bone density in rural adolescents might be attributed to low calcium and other micronutrient intake. Some studies indicated that there may not be geographical and environmental effect on BMD. But some other studies indicated that BMD is largely influenced by environmental and geographical factors (Johnell *et al.,* 1992). Some studies also showed that persons who consumed greater quantities of calcium early in life have greater bone mass later (Stear *et al*., 2003). A study conducted in China, stated that calcium supplementation increases the BMC of children with habitual low calcium intake (Boot *et al.,* 1997). Johnston *et al*., (1992) showed that calcium supplementation (1000 mg calcium/day) enhanced the rate of increase in BMD in prepubertal children. However, to conclude the reason for low BMD and BMC in rural adolescent girls, their food consumption patterns and dietary calcium sources should be evaluated.

Insufficient nutrient intakes identified in the study population were iron, zinc, magnesium and vitamin A. Although iron has no known effect on bone health, others have a role to play (FAO/WHO, 2001). In the body, 60% of magnesium is set in bone tissue. Zinc has a role in bone metabolism and development. Therefore, calcium is not the only dietary issue when considering optimal bone mass achievement. However, it should also be noted that mineral ingestion from water, which may contribute considerably to calcium intake, was not considered in the present study.
Present study indicated that weight had significant positive relationships with BMD and BMC. Those who were underweight showed lower BMC and BMD compared with those who were not underweight. The effect of weight on BMD is due to load on weight-baring

bone comparable to the influence of physical activity. Children who are underweight and inactive are at risk of developing low BMD (Boot *et al*., 1996).

A randomized controlled study conducted in UK found that increase in physical activity could increases the BMD among 16-18 years old adolescent girls (Stear *et al.,* 2003). Mechanical loading during physical activities promote the increase in skeletal mass, especially during growth in the first two decades of the life (Turner, 1999). Physical activities during critical growing years make important contributions to accrual of bone mass, independently of calcium intake. Therefore, low calcium intake may be compensated by regular physical activities in the accrual of peak bone mass. Level of adequacy of calcium in the diet may vary according to amount of physical activities during the period of growth (Anderson, 2000). Past study indicated that threshold intake of calcium during childhood and adolescents optimized the effect of physical activity on bone (Vatanparast *et al*., 2005). However, in our study, PAL had no significant correlation with BMD or BMC. Moreover, physical activity *per se* does not play a major role in BMD or BMC since both subgroups with different PAL showed similar BMD and BMC.

Although DEXA is a safe, readily available, and easily performed method for children of all ages, its application to identify individuals out of the normal range is still controversial (Prentice *et al.,* 1994). DEXA measures bone mineral content within the projected area. In the present study areal BMD was measured. Several results showed relation between areal BMD and bone strength and fracture risk, which justifies the use of areal BMD (Mora *et al*., 1995). However, a "fracture threshold" has not yet been characterized for children and adolescents using current technologies (Gonnelli and Cepollaro, 2002).

According to the results of the present study, it is difficult to provide reference values for BMD and BMC measured by DEXA in rural and urban adolescent girls in Sri Lanka because the population did not achieve optimal calcium intakes. Even low calcium intake appears to influence BMD and BMC in adolescent girls.

CONCLUSIONS

In conclusion, both urban and rural adolescent schoolgirls may represent a group at risk for developing less than optimal BMD and BMC, due to their nutritional intakes. Lack of adequate calcium intake accompanied by inadequate energy, protein, magnesium and vitamin A may have adverse effects in formation of healthy bones during adolescent period. Failure to achieve high BMD and BMC may result in the earlier onset of osteoporosis. These findings thus suggest appropriate dietary interventions and education, which can reduce the risk of future osteoporosis risk of adolescent girls.

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