

# Co-relation of Dietary and Circulatory Calcium on Bone Mineral Density in Female Population of Quetta

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## ABSTRACT

**Objective:** To determine the prevalence and severity of low bone density in the females of Quetta city and find out its co-relation with dietary factors and blood calcium level.

**Study Design:** Descriptive Corelation Study.

**Place and Duration of Study:** Two hundred women from all ethnic groups which included mixed population of Pathans, Balochi, and settlers (residing from last 30-65 years) of Quetta city were randomly enrolled in this study.

**Materials and Methods:** A sample of 200 adult healthy women, residents of Quetta city, aged 20-80 years were randomly selected to participate in the study. Blood calcium level was determined by Blood biochemical auto analyzer and bone mineral density of these subjects was measured by bonesonometer. A questionnaire was used to collect dietary, sociodemographic, age, dietary factors and other relevant detailed information affecting bone mineral density (BMD) status of women.

**Results:** Amongst all the subjects 66 (38%) were osteopenic, 17 (8.5%) and 117 (58.5 %) were normal. T-score was positively correlated with milk, ( $p < 0.01$ ) and negatively correlated to age ( $r = -0.61$ ,  $p < 0.01$ ), junk food and women bearing more than four children. BMD decreased with increasing age and low BMD was found to be more prevalent in women above 45.

**Key Words:** Dietary Calcium, Bone Mineral Density, T-score, Females.

## INTRODUCTION

Calcium plays an important role in building stronger, denser bones early in life and keeping bones strong and healthy later in life. Approximately ninety-nine percent of the body's calcium is stored in the bones and teeth. It plays an important role in many other cellular functions<sup>1</sup>. Calcium levels in mammals are tightly regulated with bone acting as the major mineral storage site. Calcium ions ( $\text{Ca}^{2+}$ ) are released from bone into the bloodstream under controlled conditions. When we do not eat enough calcium-rich food to meet the body's needs, the mineral is drawn from bones to maintain a relatively constant supply in the bloodstream. This, in turn, speeds up the loss of bone mass<sup>2</sup>. Throughout our life, old bone is constantly being broken down and removed, and new bone tissue is built to replace it. In a healthy adult, the bone cells osteoblasts synthesize the organic components of the bone matrix, which later mineralizes. Whereas, osteoclasts, break down or resorb bone<sup>3</sup>. Under normal homeostatic control, the amount of bone resorbed is roughly equal to the amount of new matrix formed. However, rate of bone-building changes as we age. Up to about age 35, new bone is added to the skeleton more rapidly than old bone is removed<sup>4</sup>. After that, bone is lost more quickly than it is built and, as a result, the skeleton becomes less dense. In Osteoporosis the normal architecture of bone is

disrupted and the matrix of bone is demineralized. The balance is disturbed and osteoclasts resorb bone faster than osteoblasts can replace it, reducing the bone strength<sup>5</sup>.

In this study the prevalence and severity of low bone density and its relationship with dietary factors as risk for Osteoporosis were accessed in females ranging from 20-80 years of age from Quetta.

## MATERIALS AND METHODS

Two hundred women from all ethnic groups which included mixed population of Pathans, Balochi, and settlers (residing from last 30-65 years) of Quetta city were randomly enrolled in this study. Participation in the study was entirely voluntary and there was no cost to the subject. All subjects were interviewed after obtaining verbal consent, by using a closed response questionnaire. Data collected from subjects included demographic information, self-reported age, language, household income, and consumption of dairy products/ other dietary habits.

Data from questionnaire was entered into Microsoft Access 2003 and then analyzed with SPSS 13.0 (SPSS Inc, USA). Anthropometric data included height, weight, subjects was also measured. to find out body mass index ( $\text{BMI} = \text{wt in kg/ht in m}^2$ ) of the subjects<sup>6</sup>.

Nutrition data was collected using a 24-hour dietary recall. Questionnaire was used to estimate dietary calcium. Dietary data was collected by a 24-hour

dietary recall method. During the interview, each subject was asked for number of servings per week on average they ate a given food item. Estimating a serving size for each food the amount of calcium per serving from tables of nutrient values was determined. These values were used to calculate calcium intake<sup>7</sup>.

Persons with fractures due to major trauma, patients with metabolic bone-related diseases or any treatment by bisphosphonate, calcium, and vitamin D3, were excluded from the study. Women taking Hormone Replacement Therapy (HRT) were also excluded from the study.

Venous blood samples were collected from women aged 20-80 years residing in Quetta city. The area of needle prick or skin was cleaned with 70% alcohol swab and allowed to dry before being punctured. Blood samples were collected through venapuncture using disposable syringes. About 2 ml blood was withdrawn from all the subjects.

Blood calcium analysis was done by Biochemical analyzer (Techno 786.) using calcium testing kit provided by Merck.

Bone mineral density of the same subjects was determined by Bone Sonometer (Hologic Inc. USA) to assess the strength of the predictive relationship of heel ultrasound, the T-score. All heel ultrasound measurements were made using the left heel and the results were interpreted according to the WHO classification of T-scores<sup>8</sup>.

-1 to +1 = Normal Bone density

-1 to -2.5 = Osteopenia

-2.5 onwards = Osteoporosis

SPSS 13 was used for data entry and analysis. Groups were compared by using Student's t test in parametric values which is significant at \*=significant ( $P < 0.05$ ) and \*\*=highly significant ( $P < 0.01$ ).

Any correlations between parameters were evaluated by using Spearman's Correlation Test. Correlation Statistics is significant at the 0.01 and 0.05 level (2-tailed). Results are given as percentages and mean  $\pm$  SD.

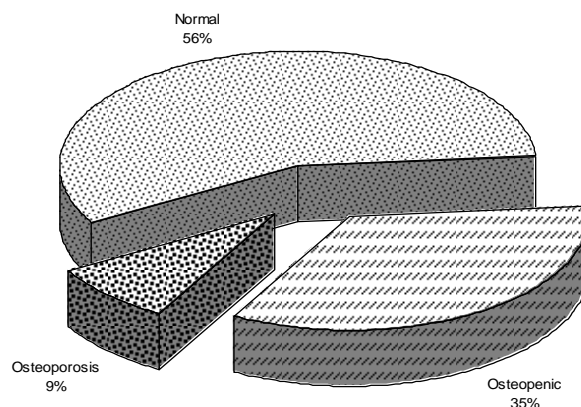
## RESULTS

Results of blood calcium analysis and bone mineral density data collected randomly from 200 women residing in Quetta city, who belonged to different ethnic groups which included mixed population of Pathans, Balochi, and settlers. Data collected from subjects including self-reported age, language, household income, and consumption of dairy products and other dietary habits.

The data were computed for means and standard deviations of means, student t-test and other statistical analysis to find the significant values. Pearson correlations were calculated among food groups and nutrients were correlated with T-score (BMD).

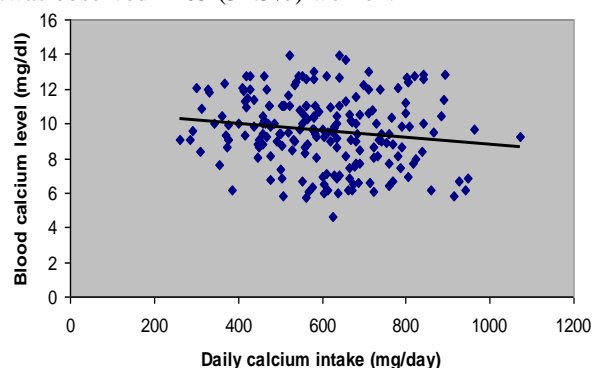
According to the BMD status of all the subjects studied

(Fig 1) 114 (57%) were normal, 69 (34.5%) women were osteopenic (i.e. T-score ranges from -1 to -2.5) and 17 (8.50%) had osteoporosis (T-score less than -2.5). Moreover, osteopenia and osteoporosis was found to be more prevalent in women bearing more than four children.



**Figure No.1: Distribution of female subjects according to bone mineral density.**

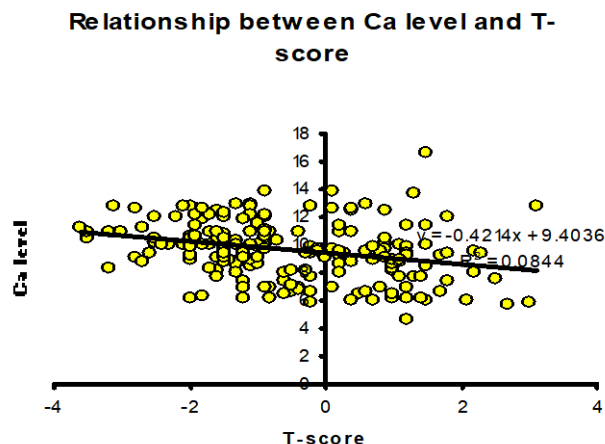
Blood calcium level was determined by Blood biochemical analyzer using kit method (Merck). Results of correlation of blood calcium level and daily calcium intake is shown in Fig 1. The mean calcium level and standard deviation of subjects was  $9.62 \pm 2.06$  mg/dl, with a maximum value of 12.5 mg/dl and a minimum level of 4.66 mg/dl. The normal range of blood calcium level is 9.00 -11.00 mg/dl. Low calcium level of about 4.50 - 8.50 mg/dl and below was present in 54 women (27%), while 81 (40.5%) women had a normal Ca level of about 8.51-10.50 mg/dl and a little above the normal range (10.51 mg/dl to 12.5 mg/dl) was observed in 65 (32.5%) women.



**Figure No.2: Blood calcium level (mg/dl) and daily calcium intake of all subjects (N=200).**

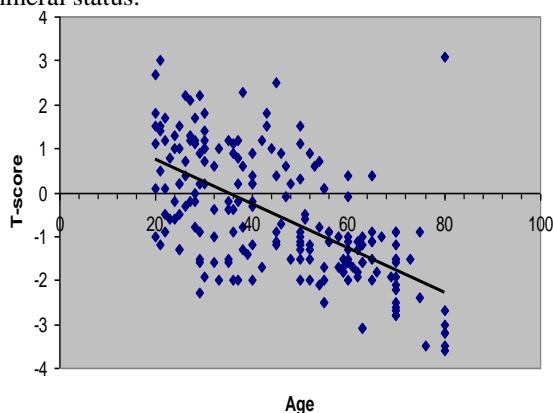
Respective T-scores were co-related with blood calcium levels of all the studied subjects were analyzed graphically and by Chi square test. However, no relationship in blood calcium level and T-score was established as shown in Fig 3 and Table 1. Relationship between Blood Ca level and BMD status

is analysed in Table 1. However, maximum number (114) subjects had normal BMD status.



**Figure No. 3: Calcium level and T-score of the studied subjects.**

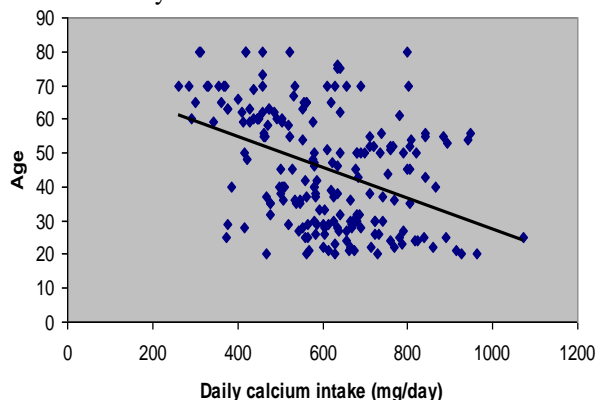
Although a large number of the subjects having normal blood calcium level lied in the range of normal bone mineral density (T-score more than -1), but low level (5-8 mg %) and higher levels of blood calcium (12-16 mgs %) were also observed in the subjects having normal bone mineral density (Fig 2). Further analysis by Chi square test (Table 2) also did not established any relationship between blood calcium level and bone mineral status.



**Figure No. 4: Age (years) and T-score of all subjects (N=200)**

The overall mean age and standard deviation of the subjects was  $45.6 \pm 17.2$  years. Youngest subject was 20 years old and maximum age was of 80 years. BMD and blood calcium level showed a negative correlation between T-score and age i.e.  $r = -0.61$ ,  $p < 0.01$  (Fig 4). BMD decrease with increasing age and low BMD was found to be more prevalent in women above 45<sup>9</sup>. Moreover, peak bone mass was observed in young adult women of 20-30 years of age.

Questionnaire data was used to estimate dietary calcium. Estimating a serving size for each food the amount of calcium per serving from tables of nutrient values was determined. These values were used to calculate calcium intake. From normal dietary routine of our subjects, daily calcium intake was estimated through calcium content (mg/ 100g) of different foods<sup>10</sup>. Mean calcium intake was 603.42 mg/100g of daily diet. Lowest calcium intake was 262mg/ day while highest calcium intake was 1071mg/100g of daily diet which is lower than recommended daily calcium intake for adults is 1000- 1300 mg/ day, Relating Fig 4 with Fig 5 it could be observed that a high dietary calcium intake is required for a healthy effect on bone mineral density<sup>11</sup>.



**Figure No. 5: Age (years) and daily calcium intake (mg/day) (N=200).**

**Table No. 1: Relationship between Blood Ca level and BMD status of the studied subjects.**

BMD status		Blood Ca_level (mg/100ml)			Total
		$\leq 8.50$	8.51 – 10.50	10.51+	
Normal	Count	44	43	27	114
	% within BMD status	38.6%	37.7%	23.7%	100.0%
Osteopenic	Count	9	32	28	69
	% within BMD status	13.0%	46.4%	40.6%	100.0%
Osteoporosis	Count	1	6	10	17
	% within BMD status	5.9%	35.3%	58.8%	100.0%
Total	Count	54	81	65	200
	% within BMD status	27.0%	40.5%	32.5%	100.0%

Chi square = 22.1201\*\*

BMD = Bone mineral density

Junk food consumption (Table 2) was calculated by estimating the serving size as done for assessment of dietary calcium. T- test was used to test the significance

of the results, alpha level was set at 0.05.

NS= non-significant ( $P>0.05$ ); \*= Significant ( $P<0.05$ );

\*\*= Highly significant ( $P<0.01$ )

n = Number of observations

**Table No. 2: T-Score of studied subject with respect to junk food (t-test).**

	Consumers/Non consumers	n	Percent	T.Score+Std. Deviation	P value
Cold Drinks	Drinking 1-2 bottles/wk	46	23.0	1.30+0.19	>0.05*
	Not Drinking	154	77.0	1.45+0.12	
Fast Food	Eating	15	7.5	-1.96+0.25	<0.01**
	Not consuming	185	92.5	1.42+0.10	
Chocolate	5-20 chocolates/wk	33	16.5	-1.24+0.22	<0.01**
	Not consuming	167	83.5	1.41+0.11	
Ice-cream	3-4 servings/wk	56	28	1.16+0.15	<0.01**
	Not consuming	144	72	1.42+0.12	

About 46 (23%) mostly young women consumed soft drinks, ice-cream and other junk food, while 154 (77%) mostly belonging to low income groups consumed simple diet and did not use junk food and soft drinks.

A small number of the subjects in our study consumed cola/carbonated drinks the average per week consumption was too low (1-2 glass/week). Thus, its correlation with blood calcium level, and T-score was not significant ( $P>0.05$ ). However, subjects consuming other junk food and chocolates were found to be osteopenic having T-score values of -1.96 and -1.24 respectively.

Subjects having ice cream in their diet (28%) had a mean T-score of  $1.16\pm0.15$  indicating normal BMD as compared to women not having ice cream, (two-tailed  $p<0.01$ ) providing evidence that there is significant difference between two groups.

## DISCUSSION

The study suggests that most of the subjects had 114 (57%) normal BMD, 69 (34.5%) women had a low BMD and were osteopenic (i.e. T-score ranges from -1 to -2.5) and only 17(8.50%) had severely low BMD and were suffering with osteoporosis (T-score less than -2.5). Moreover, osteopenia and osteoporosis were more prevalent in elderly women and amongst women bearing more than four children. T-score and age were negatively correlated ( $r = -0.36$ ,  $p<0.01$ ) reflecting bone mineral density decreasing (or bone loss) with increasing age. Young women of 20-30 years of age had Peak bone mass with a T-score of -1 to 3<sup>12</sup>. Age is a potentially confounding variable due to the decline that occurs in bone density with aging. Moreover, our study showed that women are only involved with home chores and have very little physical activities, which is further reduced after 60 years of age resulting in a low BMD. However, the significant positive effect of physical activities is observed on BMD as shown by Ferda et al.<sup>13</sup>. Gallacher<sup>14</sup> observed that in old age especially after 50 years of age bone becomes

incredibly fragile. Delaney<sup>15</sup> also reported that the lack of physical activity or weight bearing exercises is an important risk factor for osteoporosis.

In our study the lowest calcium intake was 262mg/ day, but the average intake was 603.42 mg/100g of daily diet. The dietary calcium intake of Indian women was only about 300mg /day, which is almost 700 mg less than the RDA in the West<sup>16</sup>. T-score of women having junk food was slightly reduced as observed earlier. However, a normal T-score (better BMD) as compared to women not having ice cream 144 (72%). Van der Hee et al<sup>17</sup> demonstrated that absorption of calcium from ice cream is no different than from low-fat milk. Ice cream as a potential bone health food, says a new study from Unilever. Typical ingredients and frozen format of ice cream do not negatively influence calcium absorption, wrote the researchers. In our study women also consumed junk food, but the consumption of such food is very low, thus, its relation with T-score was not significant i.e.,  $P>0.05$  in this study group.

## REFERENCES

1. Guyton, C., John E. Hall. Text Book of Medical Physiology. Para Thyroid Hormone, Calcitonin, Calcium and Phosphate Metabolism, Vitamin D, Bone, and Teeth. 15th ed 2011.
2. Velimir M, Prem KG, Nancy EB, John DL, Bin Li, Jasminka ZI, et al. Calcium supplementation and bone mineral density in females from childhood to young adulthood: a randomized controlled trial. Am J of Clin Nutr 2005; 81:(1):175-188.
3. Riggs BL, Melton LJ, editors. Osteoporosis: Etiology, Diagnosis, and Management. 2<sup>nd</sup> ed. Lippincott-Raven: Philadelphia; 1995.
4. Laura KB. Calcium and Peak Bone Mass: How Much Is Needed and When? BoneKey-Osteovision 2005; 2: 11-14.
5. Favus MJ, Christakos S, Goldring SR, et al, editors. Primer of the Metabolic Bone Diseases and Disorders of Mineral Metabolism, 3<sup>rd</sup> ed. Lippincott-Raven: Philadelphia; 1996.

6. Njeh CF, Boivin CM, Langton CM. The Role of Ultrasound in the Assessment of Osteoporosis: A Review. *Osteoporos Int* 1997; 7:7-22.
7. Nobmann, E. Nutrient Values of Alaska Native Foods, available from the Heath Sciences Information Service, Consortium Library, University of Alaska Anchorage, 3211 Providence Drive, Anchorage, AK 99508; 1993.
8. Faulkner KG, Von SE, Miller P. Discordance in Patient Classification Using t-scores. *J of Clin Densitomet* 1999; 2(3):343-50.
9. Gallagher JC, Goldgar D, Moy A. Total bone calcium in women: Effect of age and menopause status. *J Bone Min Res* 1987;2: 491-496.
10. Kerstetter JE, O'Brien KO, Insogna KL. Dietary protein, calcium metabolism, and skeletal homeostasis revisited. *Am J Clin Nutr* 2003;78: 584-592.
11. Michaëlsson K, Bergström R, Holmberg L, Mallmin H, Wolk A, Ljunghall S. A high dietary calcium intake is needed for a positive effect on bone density in Swedish postmenopausal women. *Osteoporos Int* 1997;7:155-161.
12. Anderson JJB, Tylavsky FA, Haliona L, Metz JA. Determinants of peak bone mass in young adult women: a review. *Osteoporosis Int* 1993;1 (Suppl): S 32-36.
13. Ferda Özdemir, Derya Demirbağ Kabayell, Mevlüt Türe. Do Dietary Calcium Intake and Hormone Replacement Therapy Affect Bone Mineral Density in Women? *Trakya Univ Tip Fak Derg* 2008; 25: 105-109. Med J Trakya University.
14. Gallagher JC, Riggs BL, Deluca HF. Effect of estrogen on calcium absorption and serum vitamin D metabolites in postmenopausal osteoporosis. *J Clin Endocrinol Metab* 1980; 51:1359-1364.
15. Delaney MF. Strategies for the prevention and treatment of osteoporosis during early postmenopause. *Am J Obst Gynecol* 2006;194: 12-23.
16. Sujata VV, Veena HE., Anuradha VK, Shashi AC, Deepa P., Uma Divate. Bone Status of Women Over 40 Years of Age from Two Socioeconomic Strata. *Endocr Pract* 2008;14:665-671.
17. Regine M, Van der Hee, Silvia M, Marieke S, Guus SMD, Anton GR, et al. Calcium Absorption from Fortified Ice Cream Formulations Compared with Calcium Absorption from Milk 2009;109(5): 830-835.

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