

The Association of Serum Osteocalcin with the Bone Mineral Density in Post Menopausal Women

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ABSTRACT

Background: The markers of bone remodelling, such as serum osteocalcin, may be used to assess osteoporosis and to predict the fracture risk in elderly persons, especially in women. The bone mineral density which reflects the bone mass and strength, also predicts osteoporotic related hip fractures. So, this work highlights the association between the bone turnover and the bone mass and strength.

Aim: To assess the association between the biochemical markers of bone remodeling and osteocalcin with the bone mineral density in non osteoporotic and osteoporotic women among post menopausal subjects.

Materials and Methods: Sixty postmenopausal women whose ages ranged from 55-65 years included in this study, were further divided into group 1 (thirty non osteoporotic subjects) and group 2 (thirty osteoporotic subjects). For all the subjects, serum osteocalcin was measured by ELISA. BMD was measured by the Dual Energy X- Ray Absorptiometry (DXA) scan.

The women with osteoporosis were diagnosed, based on the T- score of the bone mineral density, by the DXA scan. The Student's "t" test was performed between the variables of both the groups and a correlation test was also performed between osteocalcin and BMD by using SPSS.

Results: A negative correlation was found between the osteocalcin level and the bone mineral density in post menopausal women. The mean values of both serum osteocalcin and BMD between the osteoporotic and the non osteoporotic subjects were statistically significant.

Conclusion: An increased bone turnover coincides with the trabecular deterioration in osteoporotic women of the post menopausal age group. A combination of biochemical markers and BMD may be a better predictor of the fracture risk than when it was assessed by either alone. The biochemical markers of the bone turnover cannot be a substitute for the serial BMD measurement, but they may be useful when they are considered in conjunction with the BMD measurement.

Key Words: BMD, Osteocalcin, Osteoporosis

INTRODUCTION

According to the WHO, osteoporosis is a disease which is characterized by a low bone mass and a microarchitectural deterioration of the bone tissue, which lead to an enhanced bone fragility and a consequent increase in the fracture sites. There are two forms of osteoporosis, based on whether the disease is primary or secondary to other identifiable medical conditions or treatment. Primary osteoporosis can be classified into two types, based on the uncoupling defects which are seen in the remodeling unit. Type 1 (postmenopausal) osteoporosis is caused by an acceleration in the bone turnover as a result of hormonal deprivation. Although the entire remodeling unit is activated by oestrogen deprivation, the bone resorption exceeds the bone formation because of the time constraints on the osteoblastic activity, ultimately resulting in bone loss. In type 2 or senile osteoporosis, the osteoblastic activity for forming new bone is impaired, even though the resorption is either normal or enhanced, resulting in a chronic imbalance in the bone remodeling, leading to persistent defects in the bone mass, ultimately resulting in an increased susceptibility for bone fractures. Worldwide, the life time risk for women to have an osteoporotic fracture is 30-40% [1].

With the increasing longevity of the Indian population, it is now being realized that, as in the west, osteoporotic fractures are a

major cause of morbidity and mortality in the elderly population [2]. The biochemical markers of bone metabolism are tools of great importance in understanding the pathophysiologic basis for bone metabolic diseases. The determination of the protein fragments which are produced by osteoblasts like osteocalcin or enzymes which are secreted during osteogenesis, such as alkaline phosphatase, are commonly used to assess the osteoblastic activity. More recently, the bone turnover markers have been studied for their ability in predicting bone loss. Serum osteocalcin is a valid marker of the bone turnover when the resorption and formation are coupled and it is a specific marker of the bone formation when the formation and resorption are uncoupled.

On the other hand, the osteoporosis related fracture risk is also assessed by the Bone Mineral Density (BMD), which is a measure of the bone mass and a predictor of fracture, since the bone mass affects the bone strength or the ability to withstand trauma. It has been well established that 90% of the variance in the bone strength is related to the BMD. The risk of fracture is known to be higher in women with low BMD, with the risk doubling for a reduction of one standard deviation in the BMD [3] Osteoporosis may be predicted from the bone turn over markers and BMD, because a low BMD is associated with a high turn over [4]. It can also be predicted independently by BMD, since an increased

bone turnover negatively affects the bone microarchitecture and the fragility [5]. In this study, the status of BMD and serum osteocalcin between osteoporotic and non osteoporotic subjects among post menopausal women was investigated.

MATERIALS AND METHODS

Study Design

The present study was conducted at Sree Balaji Medical College and Hospital and Bharat Scan Limited, which are situated at Chennai, India. The institutional ethical committee approved the study and informed consents were obtained from all the participants. The study group included sixty post menopausal women. Based on the clinical features and the radiological evidence from the DXA scan, they were divided into two groups, non osteoporotic (Group 1 N= 30) and osteoporotic (Group 2 n= 30), based on the T-score. The patients who were taking hormone replacement therapy (HRT) and aniconvulsants and the patients with chronic debilitating illnesses like, renal diseases, liver diseases and Diabetes mellitus were excluded from this study. The bone mineral density in the hip region was measured by the DXA scan. Serum osteocalcin was estimated by ELISA (Osteocalcin ELISA Kit manufactured by Bio Source, Europe, SA).

Bmd Measurements

The BMD measurements (g/cm^2) for the upper end of the left femur were obtained by dual energy X- Ray Absorptiometry (DXA) with the use of a lunar DPX GE medical system. The BMD measurements were performed at Bharath Scan Centre. The World Health Organization recently published a document in which it attempted to clarify the definition of BMD and to assist clinicians in their interpretation of the bone-densitometry results. According to that report, a normal value for the bone-mineral content was within one standard deviation of the mean value for young adults of the same age and sex (i.e., the T score was more than -1). Osteopaenia was considered to be present when the value for the bone-mineral content was more than one standard deviation but not more than 2.5 standard deviations below the mean for young adults (i.e., the T score was less than -1 and more than -2.5). Osteoporosis was considered to be present when the value was more than 2.5 standard deviations below the mean bone content for young adults (i.e., the T score was less than -2.5). Severe osteoporosis was considered to be present when the value for the bone mineral content was more than 2.5 standard deviations below the mean for young adults. As a general rule, one SD approximates to 10% of the total BMD. Thus, a T-score of -1 implies that the BMD is about 10% less than the mean of a young, healthy, same sex population.

Osteocalcin Measurements

Blood samples were collected at a specific time from all the participants, under aseptic conditions. The plasma samples were stored at -20°C . Osteocalcin was estimated by ELISA by using monoclonal antibodies (MAbs) which were directed against the distinct epitopes of human osteocalcin. The amount of substrate turnover was determined colourimetrically by measuring the absorbance which was proportional to the human osteocalcin concentration. The detection range of the kit was 0.5-300ng/ml, while the coefficient of variation was 2.28%.

STATISTICAL ANALYSIS

The data were entered into the SPSS format and the continuous variables were summarized by using the mean and the standard

deviation. The Student's test was used to compare the mean difference in the serum osteocalcin and BMD between the non osteoporotic and osteoporotic subjects among post menopausal women. A correlation test was also performed between the serum osteocalcin levels and BMD.

RESULTS

In this study, there were 60 post menopausal women for whom the major characteristics have been shown in [Table/Fig-1]. The mean levels of osteocalcin and BMD show a significant difference between the postmenopausal osteoporotic and non osteoporotic women, as shown in [Table/Fig-1]. The results of the Pearson's correlation test showed a significant inverse correlation between the serum osteocalcin concentration and the total femoral BMD ($r = -0.595, p = 0.019$) and a positive correlation with age ($r = 0.303, p = 0.002$). BMI was positively correlated with both BMD ($r = 0.339, p = 0.000$) and serum osteocalcin ($r = 0.182, p = 0.061$) as shown in [Table/Fig-2].

| Numbers | Group I | Group II | P Value |
|----------------------|-------------------|-------------------|---------|
| Age | 50.44 \pm 12.1 | 53.6 \pm 12.5 | Sig |
| BMI | 26.8 \pm 4.39 | 26.7 \pm 4.3 | NS |
| BMD | 0.908 \pm 0.093 | 0.704 \pm 0.096 | Sig |
| Osteo Calcin (ng/ml) | 11.26 \pm 3.07 | 16.16 \pm 4.5 | Sig |

[Table/Fig-1]: General characteristics are shown as mean and standard deviation and Test of Significance

| | | Osteocalcin | Bmd | Bmi | Age |
|-------------|---------------------|-------------|--------|-------|--------|
| Osteocalcin | Pearson Correlation | 1 | 0.595 | 0.182 | 0.303 |
| | Sig. (2-tailed) | | 0.019 | 0.061 | 0.002 |
| | N | 60 | 60 | 60 | 60 |
| BMD | Pearson Correlation | 0.595 | 1 | 0.339 | -0.267 |
| | Sig. (2-tailed) | 0.019 | | 0.000 | 0.005 |
| | N | 60 | 60 | 60 | 60 |
| BMI | Pearson Correlation | 0.182 | 0.339 | 1 | 0.109 |
| | Sig. (2-tailed) | 0.061 | 0.000 | | 0.264 |
| | N | 60 | 60 | | 60 |
| Age | Pearson Correlation | 0.303 | -0.267 | 0.109 | 1 |
| | Sig. (2-tailed) | 0.002 | 0.005 | 0.264 | |
| | N | 60 | 60 | 60 | |

[Table/Fig-2]: Bivariate correlation between the variables

DISCUSSION

The imbalance between bone resorption and bone formation due to loss of the ovarian function, which reduces the skeletal mass, is the hall mark of menopause. Hence, loss of the ovarian function is the most important factor in the development of osteoporosis in post menopausal women. The bone mineral density was significantly decreased in post menopausal osteoporotic than in non osteoporotic subjects, which was concomitant with the findings of the study of Johannes et al., 2007 and Neeta kumari et al., [3,6]. The mean level of osteocalcin was also significantly raised in post menopausal women who were osteoporotic than in non osteoporotic individuals.

In almost all cases of osteoporosis, the bone formation remains at least partially coupled to the bone resorption, even though

the resorption rate can far exceed the formation. Osteocalcin is synthesized during the bone formation [7] and it exhibits a compact, calcium dependent, alpha helical conformation, in which the Gamma Carboxyglutamic Acid (GLA) residues bind and promote absorption to hydroxyapatite in the bone matrix. In this way, bone mineralization take place. The deficiencies of calcium and phosphorus in osteoporotic women lower the formation of the hydroxyapatite crystals, which make the free osteocalcin to circulate in the blood. This may explain the increased concentrations of osteocalcin in the sera of osteoporotic post menopausal women [8].

The increased levels of osteocalcin in the post menopausal group are attributed to an oestrogen deficiency, a finding which correlates to those of similar studies [9], which indicate a clear correlation between the oestrogen deficiency and the raised osteocalcin levels. The decreased oestrogen concentrations at the menopausal ages lead to a lower intestinal absorption of calcium, resulting in low serum calcium concentrations and an increased osteoclastic resorption of the bone. Both increase the bone turnover, thereby contributing as risk factors for the development of osteoporosis [10]. This study should be exclusively conducted on a large number of post menopausal women, to obtain a cut off value for osteocalcin, before suggesting a hormone replacement therapy for osteoporosis. A high bone turnover can disrupt the trabecular architecture and its deterioration is a contributory factor to the bone fragility, which increases the incidence of trabecular perforation and buckling, thus reducing the bone strength in osteoporosis, ultimately resulting in decreased levels of bone mineral density. BMD is the best quantifiable predictor of osteoporotic fractures. But the efficacy of the treatment cannot be judged immediately, since the structural recovery of the bone takes a little longer. Serum osteocalcin being a dynamic marker, the efficacy of the treatment can be assessed by repeating the estimation of osteocalcin and by comparing it with its original value. Thus, the assessment of osteoporotic risk fractures can be done effectively by a combination of BMD, which provides a static feature of the skeleton and the biochemical marker, osteocalcin, which provide a dynamic measure of the bone remodeling unit, as was evidenced from the study of Vanitha et al., [11].

CONCLUSIONS

In this study, a negative correlation was observed between the serum osteocalcin levels and the BMD measurement. The mean levels of both serum osteocalcin and BMD between nonosteoporotic and osteoporotic post menopausal women were statistically significant. The assessment of the osteoporotic fracture risk can be predicted better by a combination of the biochemical markers of the bone turnover, serum osteocalcin, a dynamic marker and the BMD measurement, which provide a static picture of the skeleton than the assessment by either alone.

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