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Research Article

ROLE OF FLUORIDE ON ENZYMES OF BONE TURNOVER IN POSTMENOPAUSAL OSTEOPOROTIC WOMEN

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Abstract: Osteoporosis is a major health problem of old age. This silently progressing metabolic bone disease is widely prevalent in India. It has been observed that females, after menopause, accelerated process of osteoporosis occur and it is observed in India that about 50% suffer from osteoporosis. Fluorosis is a serious public health problem in many parts of the world where drinking water contains more than 1 ppm of fluoride. Higher intake of fluoride will result in dental and skeletal fluorosis and affect collagen synthesis and bone mineralization. Approximately 99% of the body burden of fluoride is associated with calcified tissues. Fluoride concentration in bone is not uniform and higher fluoride levels in the body are associated with calcified tissues. The study was conducted on 100 subjects with their consent which consisted of control group of 50 non-pregnant women in their reproductive age group. Study group consisted of 50 women who had attained menopause either naturally or surgically, all residing in the endemic fluorotic area of Nalgonda district, Andhra Pradesh, India. 5 ml of venous blood was collected from both the groups. Serum fluoride levels (0.68 ± 0.39; $p<0.005$) was statically significant when compared to the controls (0.45 ± 0.28). Total alkaline phosphatase (132 ± 37.60; $p< 0.001$) and bone specific alkaline phosphatase (97.41 ± 38.74; $p< 0.001$) were statistically significant indicating that there is more osteoblastic activity in patients of osteoporosis. Bone turnover markers, serum total ALP and BSALP have a role in assessment of fluorosis in postmenopausal women.

Key words: Fluorosis, post menopausal women, alkaline phosphatase,bone specific alkaline phosphatase, osteoporosis

INTRODUCTION

Osteoporosis is a chronic, progressive disease characterized by low bone mass, microarchitectural deterioration, and decr

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mechanisms by which resident bone cells appropriately control bone mass and architecture in response to load bearing.[3] Primary osteoporosis is often used to define the condition in postmenopausal women in whom no specific pathogenetic mechanism other than estrogen deficiency is present.[4]

Osteoporosis is a major health and economic problem. This silently progressing metabolic bone disease is widely prevalent in India, the prevalence of osteoporosis increases with age for all sites. As regards the burden of osteoporosis in the Indian scenario, 50% women have osteoporosis and in actual numbers it accounts for 30 million women.[10] According to National Health And Nutrition Examination Survey (NHANES III), an estimated 14 million American women over age of 50 years are affected by low bone density at the hip.[2]

Bone metabolism is a dynamic and continuous process to maintain a balance between the resorption of old and injured bone initiated by osteoclasts and the formation of new bone under the control of osteoblasts.

In general, the processes of bone formation and resorption are "coupled", so that there is no net change in the bone mass. Through childhood to early adulthood, formation exceeds resorption so that bone density increases and then plateaus until the age of 30 – 40 yeas. After that, resorption exceeds formation and bone density decreases through the rest of life, which in turn may lead to osteoporosis.[2]

In this study, alkaline phosphatase and bone specific alkaline phosphatase are studied as the indicators of bone turnover and serum fluoride is estimated to know the state of fluorosis.

MATERIALS AND METHODS

This study was carried out in the Department of Biochemistry in association with Department of Medicine, Department of Orthopaedics and Department of Obstetrics and Gynaecology,Kamineni Institute of Medical

Sciences, Narketpally, Nalgonda district, Andhra Pradesh.

The study was conducted on 100 subjects with their consent which consisted of control group of 50 non-pregnant women in their reproductive age group. Case group consisted of 50 women who had attained menopause either surgically or naturally, all residing in the endemic fluorotic area of Nalgonda district.

Inclusion Criteria

Women who had attained menopause either surgically or naturally were included in the patient group and women in their reproductive age group, who were non-pregnant were included in the control group. All the subjects were from surrounding villages of Nalgonda district.

Exclusion Criteria

Women with liver disorders, alcoholism hyper / hypothyroidism and women on medication with vitamin D, calcium and hormone replacement therapy were excluded from the study.

Collection of Blood Sample For Analysis

5 ml of random venous blood was drawn from the out patients, into a sterile disposable syringe which was transferred into centrifuge tubes. The sample was centrifuged at 3000 rotations per minute for 10 minutes and the serum was collected from the centrifuge tubes.

Statistical Analysis

The statistical analysis was performed using SPSS software 11 version. The descriptive results are expressed as mean and standard deviation. Significance of difference between the patient and control groups observed is assessed by using the student t test. The p values are expressed along with mean values and standard deviation. The p values less than 0.05 were considered statistically significant.

The following biochemical parameters were estimated in both the groups.

1. Serum alkaline phosphatase
2. Heat inactivation analysis of alkaline phosphatase
3. Serum fluoride

RESULTS

The present study was undertaken in the Department of Biochemistry, Kamineni Institute of Medical Sciences, Narketpally. The patients group included the postmenopausal women aged between 50 to 70 years. The control group women were in their reproductive age group, non-pregnant, healthy individuals aged between 20 to 40 years. The mean age for the patient and control group is shown in the table no 1.

Table No.1:Age distribution among patients and controls:

	Mean ±SD Controls(n=50)	Mean ± SD Patients (n=50)
Age (years)	31.8 ± 3.58	58.3 ± 4.13

Table no.2: Serum Alkaline phosphatise (ALP) and Bone specific alkaline phosphatase (BSALP):

	Mean ± SD Controls(n=50)	Mean ± SD Patients (n=50)	p value
Serum total alkaline phosphatase (U/L)	87.5 ± 11.05	132 ± 37.6	< 0.001
Bone specific alkaline phosphatase (U/L)	47.51 ± 14.98	97.41 ± 38.74	< 0.001

Serum total alkaline phosphatase and bone specific alkaline phosphatase were estimated as the indicators of bone turnover in both patient and control groups. The mean values of total alkaline phosphatase and bone specific alkaline phosphatase are shown in table 3, which show a significant difference in both the groups. The mean values for both the parameters were two fold higher when compared to that of control group. The p value of total alkaline phosphatase ($p < 0.001$) and bone specific alkaline phosphatase ($p < 0.001$) are statistically significant and indicate that there is more osteoblastic activity in patients of osteoporosis.

Table no.3: **Serum Fluoride (F):**

	Mean ± SD Controls(n=50)	Mean ± SD Patients (n=50)	p value
Serum fluoride (ppm)	0.45 ± 0.28	0.68 ± 0.39	<0.005

Water fluoride from the surrounding villages of Nalgonda district (n=10) was estimated using Ion selective electrode, which showed the mean of 4.35 ± 2.28 ppm. The subjects included in the study were the local residents of Nalgonda district who were exposed to the fluoride consumption for >10 years.

Serum fluoride levels were measured to know the fluorotic state in both case and control groups. The mean values for both the groups are shown in table 5, which shows the increased levels in the patient group. Statistical analysis shows ($p < 0.005$) significant difference in patient and control group. This indicates that although both patients and controls are residing in the area of endemic fluorosis, patients had higher levels of fluoride. This could be due to difference in age.

Figure.**1** Radiograph pelvis; anteroposterior view. The x-ray shows normal bone density pattern. Both the hip joints are normal. Both the sacroiliac joints are normal.

Figure 2. Radiograph pelvis; anteroposterior view. Trabeculuae are prominent and cortex is thinned indicating the osteoporotic changes in the bone.

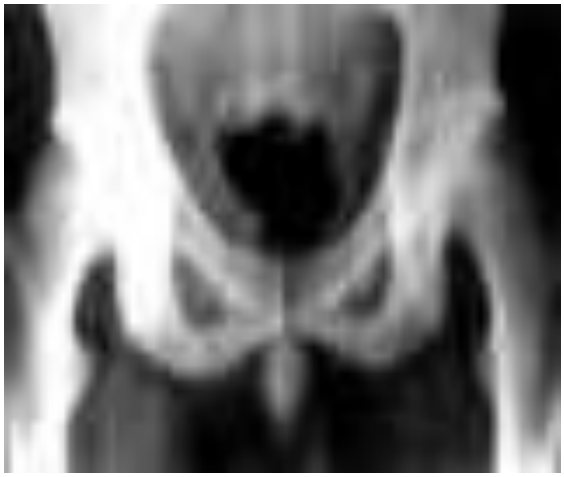

Figure 3. Radiograph pelvis; anteroposterior view. Bone density is increased with the cortico-medullary differentiation being lost. Ossification of ligaments seen which is due to fluorosis.

DISCUSSION

Serum total alkaline phosphatase and bone specific alkaline phosphatase which were estimated as the indicators of the bone turnover showed a statistically significant difference in the mean values when compared between patient and control groups, with two fold higher levels of alkaline phosphates and bone specific alkaline phosphatase in patient group. Though there is increase bone turn over in postmenopausal women, the alkaline phosphatase levels can either be increased or normal. A study by K. S. Leung et al [6] and Lennart Krook et al [7] have shown that in patients with fluorosis there is significant increase in the total ALP and BSALP. Patrick Garnero et al [8] has shown that there is highly significant increase in the total ALP and BSALP levels in the patients exposed high intake of fluoride.

Fluoride induced cell injury in both osteoblasts and osteocytes initiates a repair response and results in increased serum alkaline phosphatase production in both of these cell population. The repair response in osteoblasts results in increased proliferation, matrix production and serum alkaline phosphatase production. When the repair process in osteoblasts fails, the osteoblast undergoes either apoptosis or necrosis, and is replaced by proliferation of osteoprogenitor cells. These new osteoblasts will then be injured and cycle of increased repair response in osteoblasts would contribute to increased serum alkaline phosphatase.[9] The increase in plasma alkaline phosphatase may reflect a fluoride induced regeneration of new bone.

Bone specific alkaline phosphatase is a sensitive marker of increased bone turnover. It is found in the plasma membrane of osteoblasts which is released into the circulation by an unknown mechanism. Fluoride increases the levels of bone specific alkaline phosphatase by causing injury to osteoblasts initiating a repair response. [47].

Arjun L Khandare et al[10] and M Yildiz et al[11] showed that there is significant increase in the serum fluoride level in the patients with high fluoride intake. Our study shows siginificant increase in the serum fluoride in the patient group.

The serum fluoride levels of both case and control groups are low compared to that of the water fluoride levels estimated. This is because, the clearance of fluoride from plasma by the skeleton proceeds at a high rate that exceeds even for the calcium. Approximately 50% of the fluoride absorbed each day becomes associated with calcified tissues within 24 hours, while virtually all of the remainder is excreted in the urine. [9]

Our study showed increased serum total alkaline phosphatase and bone specific alkaline phosphatase enzymes indicating the increased bone turnover.

CONCLUSION

- Biochemical markers are non-invasive, inexpensive and can be repeated often.
- Biochemical markers have a role in the diagnosis of postmenopausal osteoporosis in endemic fluorotic area.
- Bone mineral status measured using serum total calcium and phosphorus have less significant role in fluorotic patients.
- Bone turnover markers, serum total ALP and BSALP have a role in assessment of fluorosis in postmenopausal women.

REFERENCES

1. Karen F. Bart L. Clarke. Diagnosis, screening, prevention and treatment of osteoporosis. *Mayo Clin Proc*. **2006**; 81 (5): 662 – 672.
2. Lance Lanyon, Tim Skerry. Postmenopausal osteoporosis as a failure of bones adaptation to functional loading: a hypothesis. *Journal of Bone and Mineral Research*, **2001**; 16 (11): 1937 – 1947.
3. S.Sharma, V.R.Tandon, A.Mahajan. Postmenopausal osteoporosis. *International Journal of Gynecology and Obstetrics India*, **2006**; 9 (4): 156 – 168.
4. Sarvardekar LS, Shah RS. Bone density in normal Indian women: Assessment by USG and DEXA. *Obs and Gynae Today* **2004**; 9 (11): 256 – 264.
5. Indumati V. Vidya S. Patil, Rama Jailkhani. Hospital based preliminary study on osteoporosis in postmenopausal women. *Indian Journal of Clinical Biochemistry*, **2007**; 22 (2): 96 – 100.
6. K.S.Leung, K.P.Fung, A. H.L.Sher, C.K.Li, K.M.Lee. Plasma bone-specific alkaline phosphatase as an indicator of osteoblastic activity. *J Bone Joint Surg 1993*; 75: 288-292.
7. Lennart krook, Ronald R Minor. Fluoride and alkaline phosphatase. *Fluoride* **1998**; 13(3): 77 – 182.
8. Patrick Garnero, Pierre D. Delmas. Assessment of the serum levels of bone alkaline phosphatase with a new immunoradiometric assay in patients with metabolic bone disease. *Journal of Clinical Endocrinology and Metabolism*. **1993**; 77 (4): 1046 – 1053.
9. HM Nawawi, TN Yazid, NMN Ismail, AR Mohammad, S.Ima Nirwana, BAK Khalid. Serum bone specific alkaline phosphatase and urinary deoxypyridinoline in postmenopausal osteoporosis. *Malaysian J Pathol*, **2001**; 23(4): 79 - 88.
10. Arjun L.Kandare, Rao GS, N Balakrishna. Dual x-ray abosrptiometry (DXA) study of endemic skeletal fluorosis in a village of Nalgonda district, Andhra Pradesh. *Fluoride*, **2007**; 40(3): 190 – 197.
11. M.Yildiz, M.Akdogan, N.Tamer, B.Oral. Bone mineral density of the spine and femur in early postmenopausal Turkish women with endemic skeletal fluorosis. *Calcif Tissue Int*, **2003**; 72: 689 – 693.
12. G. M. Whitford. Intake and metabolism of fluoride. *Adv Dent Res* **1994**; 8:5 – 14.