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Sri Rekha P
Senior Resident, Department Of Biochemistry, Kurnool Medical College, Kurnool, Andhra Pradesh

U Venkateswarlu
Medical Officer Echs Hospital, Kurnool, Andhra Pradesh

Sarada U
Assistant Professor, Department of Biochemistry, Kurnool Medical College, Kurnool, Andhra Pradesh

Correspondence:
Sri Rekha P
Senior Resident, Department Of Biochemistry, Kurnool Medical College, Kurnool, Andhra Pradesh

Comparitive study of Biochemical bone turnover markers in pre & post-menopausal women

Sri Rekha P, U Venkateswarlu, Sarada U

Abstract
Menopause is associated with adverse changes in bone turnover. When bone resorption exceeds formation, bone loss and osteoporosis will occur. This is explained by Biochemical marker of bone turnover profile. Though Bone Mineral Density (BMD) measurements are the gold standard in calculating bone mass, the changes are usually late and the damage is irreversible. Combined use of biochemical markers of bone turnover helps in early identification of women who are at increased risk of fractures.
Objective: To compare Serum Total Alkaline Phosphates (ALP), Total Calcium, Urinary OHP in postmenopausal cases and pre-menopausal controls.
Methods: The study included 25 healthy postmenopausal women as cases, and 25 healthy Premenopausal women. Estimation of ALP was done by kinetic method Recommended by International Federation of Clinical Chemistry (1FCC), Serum total Calcium by modification of Ortho- Cresolphthalein Complexone (OCPC) method, Urinary OHP by modified Neumann and Logan method.
Results: Bone formation markers, serum total ALP was significantly increased ($p<0.001$) and total calcium was significantly decreased ($p<0.001$) in postmenopausal women compared to premenopausal women. Bone resorption marker, urinary OHP was significantly increased ($p<0.001$) in postmenopausal women
Conclusion: This study concluded that, Serum total ALP, total calcium and Urinary OHP combined together provided a fairly useful index of bone resorption in postmenopausal women and these common biochemical parameters can be used to categorize postmenopausal women into rapid and slow bone losers. Preventive measures like calcium supplementation or Hormone replacement therapy can be initiated early in those who are rapid bone losers and prevent the osteoporotic fractures.

Keywords: Postmenopausal women; osteoporosis; Total Alkaline Phosphatase; Total calcium; Urinary hydroxy proline

1. Introduction
Bone is remodeled constantly throughout life. Peak bone mass is around the age of 30 years after which rate of bone resorption is greater than the rate of bone formation (0.3% to 0.5% bone loss per year). Approximately 3% of cortical bone is replaced each year and 25% of trabecular bone is resorbed and replaced each year [1].
Osteoporosis is defined as bone mineral density less than 2.5 SD below the mean peak value in young adults of the same race and sex (t score of -2.5). Women of all ethnic groups show an additional accelerated phase of bone loss, which occurs for about 10 years after the cessation of ovarian function. Total bone loss in osteoporosis may exceed 30 to 40%. In women there is a 15 to 18% risk of hip fracture after the age of 50 years, versus 6% in men. Menopause is associated with adverse changes in bone turnover. When bone resorption exceeds bone formation, bone loss and osteoporosis results. Also changes in Bone Mineral Density (BMD) being late and relatively irreversible, it is important to have a means of identifying high risk individuals and to monitor their treatment before fracture occurs [2].
Bone biochemical markers have been suggested to reflect the menopausal high bone turnover. These markers could be useful in separating women into fast and slow bone losers.
Biochemical markers of bone turnover are:

- Markers of bone formation, reflecting the osteoblastic activity are Alkaline Phosphatase: it is measured in the serum or plasma.
- Markers of bone resorption, reflecting osteoclastic activity, are mainly the degradation products of type 1 collagen such as Hydroxy proline, hydroxypyridinium cross links and Tartrate resistant acid Phosphatase
- Markers of mineralization are Serum Calcium.

These markers reflect alterations in bone remodeling much earlier than they are apparent Radio graphically. They have untapped potential in the evaluation of patient at risk for accelerated bone loss especially in Postmenopausal women. The occurrence of osteoporosis in women is very common in India.

It is widely accepted that "any single measurement of single biochemical marker of bone turnover has limited value in individual person", so they are often studied in combinations [3].

Hence, in the present study, we intended to measure serum Alkaline Phosphatase, Total calcium and Urinary Hydroxyproline in Postmenopausal women and evaluate the efficacy of using these biomarkers to assess the bone turnover in Postmenopausal women, and the possible risk of developing osteoporosis and fractures.

2. Materials and Methods

A study of Biochemical Bone turnover markers was conducted in healthy Postmenopausal women (cases) and healthy Premenopausal women (controls) from Government General Hospital, affiliated to Kurnool Medical College, Kurnool. The study was approved by Ethical and Research Committee of Government General Hospital to use human subjects in the research study. The study was conducted from April 2012 to February 2013.

Subjects: A total number of 50 subjects were selected for the present study based on the inclusion and exclusion criteria.

Cases: Out of 50 subjects, 25 healthy post-menopausal women were considered as cases.

Controls: Out of 50 subjects, 25 healthy pre-menopausal women were considered as Controls.

Inclusion Criteria : For cases healthy women who had attained menopause for 3 to 5 years and not started taking hormone replacement therapy, non-obese and without use of calcium supplementation or any other medications known to affect bone metabolism.

For controls healthy women of reproductive age group 30 – 40 years with regular menstruation who were non pregnant and not taking oral contraceptive pills.

Exclusion Criteria: Women of known osteoporotic features, Diabetes mellitus, Renal insufficiency, Cirrhosis or those on hormonal replacement therapy and on the steroid treatment for more than 6 months, were all excluded.

Collection of Samples: About 5 ml of venous blood from all subjects was collected a aseptically from antecubital vein. Serum was separated immediately by centrifugation and kept at 4 degrees C until analysis was carried out. A random urine sample was collected at the same time in a clean plastic container.

Parameters Measured: Serum Alkaline Phosphatase, Serum Calcium, Urinary Hydroxyproline.

Serum Alkaline Phosphatase was measured by Kinetic method recommended by the international Federation of Clinical Chemistry. Using appropriate reagents and appropriate assay parameters [4, 5]. Serum Calcium was measured by Orthocresolphthalein complexone method [5, 6]. Urinary Hydroxyproline was estimated by Modified Neumann and Logan method [7, 8, 9].

Statistical Analysis: Statistical analysis of biochemical parameters done by unpaired' test

3. Observations & Results

Table 1: Age Wise Distribution of Premenopausal Controls and Postmenopausal Cases:

Age	Postmenopausal cases	Premenopausal controls
20-30	0	14
31-40	0	9
41-50	7	2
51-60	16	0
61-70	1	0
>70	1	o
Mean ± SD	54.84±7.1	31.96±7.1

Table 2: Comparison of Bmi, Levels of Serum Albumin, Total Alkaline Phosphatase Calcium and Urinaryhydroxyproltne/Creatinine Ratio between Premenopausal Controls and Postmenopausal Cases

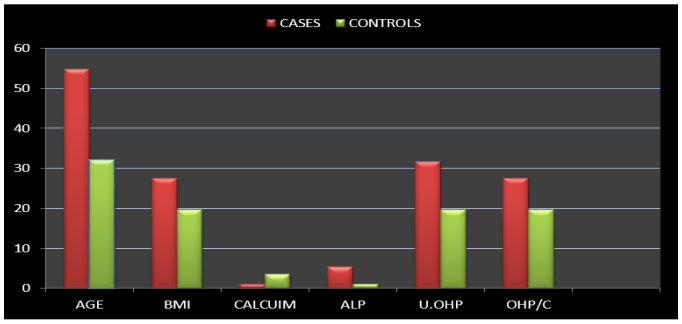
Group	No.	BMI (Kg/mt^2)	Alb (g/dl)	ALP (ukat/L)	CA (mmol/L)	OHP/Cre ratio
Post-menopausal cases	25	27.23±2.O5	4.34±1.12	5.41±3.8	1.03±0.03	27.51±6.63
Pre-menopausal controls	25	23.87±2.30	4.48±1.27	1.09±0.4	3.50±0.60	19.64± 2.94
't'* value(unpaired t test)		5.44	0.41	6.86	2.50	5.42
'P' value		<0.001	0.68	<0.001	<0.0001	<0.0001
Statistical significance		HS	NS	HS	HS	HS

The mean serum calcium in post-menopausal women was 1.03mmol/ L and 3.50mmol/L in pre-menopausal women which is statistically significant. Mean ALP was 5.4 in postmenopausal women and 1.09 in pre-menopausal women which is statistically significant. Mean urinary hydroxy proline was 31.64 in postmenopausal women and 19.61 in premenopausal women which is statistically significant. The statistical analysis by unpaired t-test shows that BMI and levels of serum total ALP, and urinary hydroxy proline creatinine ratio is increased in postmenopausal women

Table 3: Relationship of Various Biochemical Parameters of Bone Turnover in Postmenopausal Cases with Age and BMI

PARAMETERS	Relation with age		Relation with BMI	
	r-value*	p-level	r-value *	p-level
Albumin (g/dl)	-0.41	0.03	-0.2	0.24
ALP(ukat/L)	0.23	0.25	0.5	0.003
Serum total calcium (mmol/L)	0.1.	0.61	0.17	0.4
Hydroxyproline(mg/dl)	-0.06	0.75	0.12	0.5
Creatinine (gm/day)	0.45	0.02 S	0.41	0.04 S
OH P/Cr	-0.44	0.02 S	-0.47	0.01 S

Statistical analysis using pearson's correlation co-efficient shows that the correlation of age with serum albumin, total ALP, total calcium Hydroxyproline (mg/dl),is not significant with p-values of **0.03**, **0.25, 0.61, 0.75** respectively and with urinary hydroxy proline/ creatinine ratio is significant. Correlation of BMI with serum albumin, total ALP, total calcium and Hydroxy proline (mg/dl), urinary hydroxy proline /creatinine ratio is not significant with p-values of **0.24, 0.003, 0.4, 0.5, 0.04, 0.01.**respectively



Graph 1: Comparision of Mean of age BMI and bone turn over markers with premenopausal and postmenopausal women

4. Discussion

Osteoporosis leads to considerable morbidity and mortality in post-menopausal women. The mean age at menopause was observed to be 47.56 years [10].

The prevalence of osteoporosis increases with age, and by WHO definition up to 70% of women over the age of 85 years have osteoporosis [11].

The main aim of study was to evaluate biochemical bone turn over markers in post-menopausal women and pre-menopausal women.

Serum Alkaline Phosphatase activity is the most commonly used marker of bone formation. A moderate increase in ALP indicates a mineralization defect in the elderly patients [12]. At the menopause, the rate of bone demineralization increases precipitously [13]. It is suggested that in most clinical situations, measurement of serum ALP provides sufficient diagnostic information at a much cheaper rate when compared to BMD test and also BMD machine is not available at peripheral centers [14].

Urinary hydroxyproline is increased in states of physiologically high turnover, such as somatic growth and during menopause [15]. This simple, direct urinary assay of hydroxyproline to measure resorption has clinical applications as part of screening programmes to assess the risk of osteoporotic fractures [16].

Serum Calcium estimation will show early onset of osteoporosis, thus the osteoporotic fractures can be prevented by giving prophylactic Calcium therapy.

We conclude that biochemical markers of bone turnover are valuable tools of detecting osteoporosis as they reflect the activity level of the entire skeleton as compared to BMD test which selects only specific small areas like Calcaneum or Vertebra for Diagnosis.

5. Conclusion

In Post-menopausal women with osteoporosis, biochemical markers of bone formation and bone resorption increase, because of more osteoclastic activity.

In the present study postmenopausal women serum calcium is decreased, serum alkaline phosphatase is increased and urinary hydroxyproline is elevated when compared to Pre-menopausal women.

In the present study the biochemical marker of bone formation is decreased and biochemical markers of bone resorption are increased in Post-menopausal women.

The present study also indicates that urinary hydroxyproline excretion increased with increasing age.

We thus conclude that biochemical markers of bone turnover are valuable tools in detecting osteoporosis as they reflect activity level of entire skeleton.

6. References

1. Sachdeva A, Seth S, Khosla AH, Sachdeva S. Study of some common biochemical bone turnover markers in postmenopausal women. Indian J ClinBiochem 2005; 20(1):131-134.
2. Heikkinen AM, Parviainen M, Niskanen L, Komulainen M, Tuppurainen MT, Kroger H *et al.* Biochemical bone markers and bone mineral density during postmenopausal hormone replacement therapy with and without vitamin D3: A prospective, controlled, randomized study. J ClinEndocrinolMetabol 1997; 82(8):2476-2482.
3. Sachdeva A, Seth S, Khosla AH, Sachdeva S. Study of some common biochemical bone turnover markers in postmenopausal women. Indian J ClinBiochem 2005; 20(1):131-134.
4. Wilkinson JH, Winsten S. ClinChem 1969; 15:487.
5. Baird DD, Tylavsky FA, Anderson JJB. Do vegetarians have earlier menopause? Am J Epidemiol. Proceedings of the Society of Epidemiol Research. 1988; 907:J4
6. McMahon B, Worcester J. Age at menopause U.S. 1960-62. Vital Hens Stat 1966; 9(1):J5
7. Melton LI, Khosla S, Atkinson EJ, O'fallon WM, Riggs BL. Relationship of bone turnover to bone density and fractures. J Bone Mineral Res 1997; 12(7):1083-1091.
8. Garnero P, Sornay-Rendu E, Chapuy MC, Delmas PD. Increased bone turnover in late postmenopausal women is a major determinant of osteoprosis. J Bone Mineral Res 1996; 11:337-347.
9. luaska KK, Lenora J, Gerdhem P, Akesson K, Vaananen HK, Obrant KJ. Serial assessment of serum bone metabolism markers identified women with the highest rate of bone loss and osteoporosis risk. J ClinEndocrinolMetabol 2007; 93:2622-2632.
10. Hauselmann HJ, Rizzoli R. A comprehensive review of treatment for postmenopausal osteoporosis. OsteoporosInt 2003; 14:2-12.
11. Agrawal S, Jain A, Mahajan D, Raghunandan C. Correlation of bone mineral density with biochemical markers in post-menopausal women. Indian J ClinBiochem 2009; 24(3):262-265.
12. Manolagas SC. Birth and death of bone cells: Basic regulatory mechanisms and implications for the pathogenesis and treatment of osteoprosis. Endocrine Reviews 2000; 21(2):115-137.
13. Withold W. More on total and bone-specific alkaline phosphatase. ClinChem 1997; 43:1670a-1671a.
14. Auth C, Seibel KJ, Robins SP, John P, Bilezikian. Markers of bone metabolism. In: Beckers Text Book of Endocrinology Chapter 56. 3rd edn, 2008, 548-557.
15. Boyle WJ, Simonet WS, Lacey DL. Osteoclast differentiation and activation. Nature 2003; 423:337-342.
16. Agrawal S, Jain A, Mahajan D, Raghunandan C. Correlation of bone mineral density with biochemical markers in post-menopausal women. Indian J ClinBiochem 2009; 24(3):262-265.