



## ASSESSMENT OF ANY DIFFERENCE IN BIOCHEMICAL PARAMETERS OF BONE MATRIX DEGRADATION AND BONE TURN OVER BETWEEN PRE, POST AND ELDERLY POST MENOPAUSAL WOMEN.

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### ABSTRACT

**INTRODUCTION:** Bone matrix component are released in the circulation throughout our life. After menopause bone resorption becomes greater than bone formation due to reduced estrogen secretion. The biochemical parameters for matrix degradation products as urinary hydroxyproline, serum calcium, enzymatic and bone turn over marker as serum alkaline phosphatase can be assessed for early diagnosis of osteoporosis. The present study was done to assess any difference in biochemical parameters of bone matrix degradation and bone turn over between pre, post and late post menopausal women.

**MATERIAL & METHODS:** The present study has been conducted in Biochemistry Department of Calcutta National Medical College Kolkata among 80 premenopausal, 80 post menopausal and 20 elderly menopausal women attending in the thyroid OPD of the department. From the study population fasting blood and urine was collected and estimated for serum ionized calcium, serum alkaline phosphatase and urinary hydroxyproline after an informed consent with the standard laboratory procedure.

**RESULTS:** It was found that mean Serum calcium, serum alkaline phosphatase was significantly decreased and urinary hydroxyl-proline was significantly increased in post menopausal & elderly post menopausal women than the pre-menopausal group. It was also found that UHP was positively correlated in all the groups and serum ionized calcium was correlated only in pre-menopausal women.

**SUMMARY & CONCLUSION:** Estimation of serum ionized calcium, alkaline phosphatase, urinary hydroxyproline can be used an easy tool for early diagnosis and early intervention in osteoporosis in countries like India where estimation of bone mineral density by bone scan is costly for most of the sufferers.

**KEYWORDS :** Menopause, Urinary hydroxyproline, Osteoporosis

### INTRODUCTION:-

Bone is dynamic tissue which is being remodeled throughout the whole life. It is composed primarily of the inorganic minerals (calcium and phosphate) and type I collagen constitutes the organic matrix. Bone formation is an orderly process in which inorganic mineral is deposited in relation to organic matrix.

Bone matrix components are released into the circulation, either by the osteoblasts or by the osteoclasts. During bone resorption, first calcium and phosphorus are released into the extracellular fluid and organic matrix is then resorbed<sup>1</sup>.

Peak bone mass is around the age of 30 years after which rate of bone resorption is greater than the rate of bone formation. Osteoporosis is defined as a disease that cause a reduction in the mass of bone per unit volume under the influence of reduced estrogen secretion by exhausted ovarian follicles and is one of the dreaded afflictions of aging<sup>2,3,4</sup>. This predisposes to increased risk of fractures, and morbidity in menopausal women<sup>5</sup>.

During increased osteoclastic activity urinary excretion of collagen degradation products like hydroxyl lysine, hydroxyl proline and inorganic minerals e.g ionized calcium in plasma get increased<sup>6</sup> along with the enzymes of osteoblasts e.g alkaline phosphatase.

The prevalence of post menopausal osteoporosis in India is 20%<sup>7</sup>.

The presently available methods for measurement of bone density like DEXA scan are very expensive, changes being late and relatively irreversible<sup>8</sup>, bone biopsy is an invasive procedure. These markers reflect alterations in bone remodeling earlier than radio graphically.

So the biochemical parameters e.g bone organic matrix degradation products as urinary hydroxyproline, serum calcium, and enzymatic bone turn over marker as serum alkaline phosphatase can provide cost effective and accurate tolls to diagnose and follow up of osteoporosis.

### OBJECTIVES:-

To assess serum ionized calcium in premenopausal and postmenopausal women.

To measure the serum alkaline phosphatase in premenopausal and postmenopausal women

To measure the urinary hydroxyproline in premenopausal and postmenopausal women

To assess any significant alteration and correlation of these parameters between these two groups.

### METHODOLOGY:-

The present study was a descriptive, cross-sectional one done among the women attending the thyroid OPD of Biochemistry Department of Calcutta National Medical College. From the study population 80 premenopausal, 80 post menopausal and 20 elderly menopausal were selected as study population. Women with pregnancy, endocrine disorders, liver disorders, renal disorders, congestive cardiac failure and women on oral contraceptive pills and statins were excluded from the study. Informed consent was obtained from the study population and permission was obtained from Institutional Ethics Committee. From the study population fasting blood and urine was collected and estimated for serum ionized calcium, serum alkaline phosphatase and urinary hydroxyproline after an informed consent with the standard laboratory procedure. The instruments used for the study was Semi auto analyzer- Transasia ERBA CHEM 5 PLUS. The instruments and reagents were validated by concerned authority for quality control and the results were subjected to co-efficient of variation (cv).

Serum ionized calcium (mmol/lit) was analysed by 9180 Electrolyte Analyser. Principle based on Ion Selective Electrode (Roche Diagnostics GmbH Mannheim Germany), Serum Alkaline phosphatase (IU) was estimated by Pnpp kinetic method (kit-ERBA)<sup>9</sup> and Urinary hydroxyproline (mg/24 hrs urine) was assessed by colorimetric (Modified Neuman and Logan Method)<sup>10</sup>.

The result was analysed with statistical software SPSS version 20 by using appropriate parametric and non parametric tests.

### RESULTS:-

The present study has been conducted in Biochemistry Department of Calcutta National Medical College Kolkata among 80 premenopausal, 80 post menopausal and 20 elderly menopausal (above 60 years)

women attending in the thyroid OPD of the department. The study assessed mean ionized

calcium, mean alkaline phosphatase and mean UHP in these three groups. It was revealed that in premenopausal age group mean ionized calcium, mean alkaline phosphatase and mean UHP were 2.36 mmol/lit, 116.40 IU and 16.29 mg/24 hrs urine respectively; in post menopausal age group these were 2.06 mmol/lit, 82.08 IU and 27.29 mg/24 hrs urine and in elderly post menopausal these were 1.88 mmol/lit, 86.1 IU and 37.81 mg/24 hrs urine respectively (Table 1).

**Table 1: Distribution of means of UHP, Alkaline phosphatase, Ionized calcium in the premenopausal, post menopausal and elderly post menopausal women**

Age group	Mean± S.D(Ionized calcium) (mmol/lit)	Mean± S.D (Alkaline phosphatase) (IU)	Mean± S.D(UHP) (mg/24hrs urine)
Premenopausal	2.36±0.328	116.40±24.93	16.29±0.813
Post menopausal	2.06±0.335	82.08±11.84	27.29±3.438
Elderly post menopausal	1.88±0.201	86.11±11.96	37.81±4.02

The means of UHP, Alkaline phosphatase, Ionized calcium were compared among the premenopausal, post menopausal and elderly post menopausal women. In which it was found that Serum calcium, serum alkaline phosphatase was significantly decreased and urinary hydroxyl-proline was significantly increased in post menopausal & elderly post menopausal when compared to their respective premenopausal group as tested by annova test (Table 2).

**Table 2: Comparison of means of UHP, Alkaline phosphatase, Ionized calcium in the premenopausal, post menopausal and elderly post menopausal women**

ANOVA		Sum of Squares	df	Mean Square	F	Sig.
Ionised CA	Between Groups	5.344	2	2.672	25.718	.000
	Within Groups	18.388	177	.104		
	Total	23.731	179			
UHP	Between Groups	8899.106	2	4449.553	438.076	.000
	Within Groups	1797.795	177	10.157		
	Total	10696.900	179			
ALP	Between Groups	49986.244	2	24993.122	70.079	.000
	Within Groups	63125.398	177	356.641		
	Total	113111.642	179			

It was found in the study that UHP was positively correlated in all the age groups whereas alkaline phosphates was not correlated with age. Serum ionized calcium was correlated in pre-menopausal age group but not in other two groups (Table 3).

**Table 3: Correlation of UHP, Alkaline phosphatase, Ionized calcium in the premenopausal, post menopausal and elderly post menopausal women**

Age group	PARAMETERS (Correlation coefficient)		
	Ionized calcium	Alkaline phosphatase	UHP
Premenopausal	0.355(p=0.01)	0.041(p=0.715)	0.418(p<0.001)
Post menopausal	0.132(p=0.244)	0.091(p=0.422)	0.443 (p<0.001)
Elderly post menopausal	0.005(p=0.984)	0.070(p=0.770)	0.730(p<0.001)

#### DISCUSSION:-

In our study, we have selected 180 females out of which 80 are premenopause and 80 of post-menopause (age between 45-65years) & elderly post-menopause (age>65 years) groups.

Serum calcium, serum alkaline phosphatase is significantly decreased and urinary hydroxyl-proline is significantly increased in post menopausal & elderly post menopausal when compared to their respective pre-menopausal group. Urinary hydroxyl-proline is positively correlated with increasing age in all 3 groups. Alkaline phosphates is not correlated with age and serum ionized calcium is correlated in pre-menopausal age group but not in other two groups.

UHP is the increased only when the collagen is broken down but serum alkaline phosphatase is effected by many other factors along with osteoclastic activity. Physiologically serum ionized calcium is maintained normal even at the cost of bone loss under the effect of parathormone.

Our results are in correlation with the studies of Indumathi.v<sup>11</sup>, ashuma sachadeva, shashi seth<sup>2</sup> and Ren XH, XD<sup>12</sup>.

The prevalence of osteoporosis increases with age for all sites, and by WHO definition up to 70 % of women over the age of 89 years have osteoporosis<sup>13</sup>. A high mortality is well recognized in osteoporotic patients with hip fracture compared with the general population<sup>14</sup>. The fracture burden has increased in India upto 36 million by 2013<sup>15</sup>. Bone mass decreases with aging, and it is now well established that a low bone mass is the major determinant of all osteoporotic fractures. The dramatic increase in the bone turnover rate with an imbalance between bone formation and bone resorption in the first year after the cessation of ovarian function is responsible for the accelerated rate of postmenopausal bone loss. High bone turnover rate seems to play an increasing role as a determinant of bone mass with increasing postmenopausal age<sup>15</sup>. Typically, the urinary markers of bone resorption include hydroxyproline, which serve as rapid predictors of changes in collagen metabolism<sup>5</sup>. With increasing age the collagen breakdown increases which results in increased urinary hydroxyproline. Several factors might be responsible for excess bone resorption such as low estrogen level, calcium and vitamin D deficiency and age related reduced calcium absorption. We find a positive correlation of serum calcium with age in pre menopausal age group.

Many studies show that the reduction in bone loss during anti-resorptive treatment is demonstrated by decreased excretion of hydroxyproline post antiresorptive therapy<sup>18</sup>.

So biochemical markers like calcium, urinary hydroxyproline are not only predictor of bone loss but they help in better understanding of the pathological process and the follow up with treatment.

#### CONCLUSION:

Osteoporosis leads to considerable morbidity and mortality in post menopausal women. The mean age at menopause was observed to be 47.56 years<sup>16</sup>. The prevalence of osteoporosis and increases with age, and by WHO definition up to 70% of women over the age of 85 years have osteoporosis<sup>17</sup>. Countries like India where estimation of bone mineral density by bone scan is costly for most of the sufferers. Estimation of simple parameters like serum ionized calcium, alkaline phosphatase, urinary hydroxyproline can be used an easy tool for early diagnosis and early intervention.

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