



# A CROSS SECTIONAL STUDY TO ASSESS THE RELATIONSHIP BETWEEN ANTIOXIDANT STATUS AND SERUM LEVELS OF BONE MARKER WITH OXIDATIVE STRESS MARKER IN PRE AND POSTMENOPAUSAL WOMEN.

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## Abstract

**Introduction:** Menopause is defined as the time of women life when there is adaptation of physical, emotional, mental and hormonal changes associated with the cessation of menstrual period. About 60 million Indian women are above the age of 55 years and a majority would spend 1/3rd of their life on postmenopausal stage. **Aim & objective:-** To correlate the Serum level of Bone Markers (Vitamin D<sub>3</sub>, Calcium, Phosphorus, ALP), Oxidative Stress Marker (Malondialdehyde) along with the level of Total antioxidant status in Pre & Post-menopausal Women. **Material and methods:-** Total three hundred ten healthy women were included, out of which 155 were healthy pre-menopausal and 155 were postmenopausal women. Serum was analyzed for the measurement of Vitamin D [ElectroChemiluminescence binding assay, Serum Calcium [O-Cresolphthalein Complex method and Serum Phosphorus [Modified Gomorri's Method. **Result:-** Serum vitamin D, Calcium and Total antioxidant levels were significantly decreased in postmenopausal women (p-value<0.0001) as compared to premenopausal women; whereas the serum phosphorus level was found to be significantly increased in postmenopausal women (p-value<0.0001) **Conclusion:** The present study findings indicate decreased levels of serum vitamin D & serum calcium and increased the level of serum phosphorus in postmenopausal women. These changes can be used as a good marker for identifying bone related disorders in postmenopausal women. Intimation with supplementation at an early stage may further prevent bone disorder in the later stage of menopause.

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## Introduction

Menopause is defined as the time of women life when there is adaptation of physical, emotional, mental and hormonal changes associated with the cessation of menstrual period. Menopause mostly occurs between the age of 45 – 50 years; the average age is 47. About 60 million Indian women are above the age of 55

years and a majority would spend 1/3rd of their life on postmenopausal stage.<sup>1</sup> Calcium is the most important mineral in the human body, which accounts for about 99% in skeletal tissues, 0.9% in cells of soft tissues and 0.1% in bloodstream. The oxidative stress pathway activates the differentiation of pre-osteoclast into osteoclast and provokes bone resorption.

An increasing level of reactive oxygen species (ROS) results in rapid bone loss, especially in postmenopausal women.<sup>2</sup> Oxidative stress is the pathophysiological driver of the aging process and a cause of osteoporosis. Oxidative stress aggravates the osteoclastic activity in the bone remodeling process, without effect on the osteoblast. Moreover, bone resorption predominates bone formation, resulting in diminished bone density. The remodeling process is based on the balanced activities of bone resorption and formation. This process requires communication between different bone cells, namely, cells of the osteoblast lineage (osteoblasts and osteocytes) and bone-resorbing cells (osteoclasts), which are organized in specialized units called bone multicellular units (BMU). The bone remodeling controlled by various local and systemic factors in different pathways, such as Calcium, phosphorus, vitamin D is required for the maintenance of the skeletal system in women throughout life. During the menopausal phase, there is an increase in bone resorption rate of calcium that declines in bone mass due to fall in ovarian estrogen production.<sup>3</sup> The main role of calcium is to maintain bone health, especially in menopausal women. The calcium absorption is decreased due to the lack of vitamin D, resulting from the age-related declines in several functions.<sup>4</sup> Declining in the ovarian function during menopause is accompanied by a decrease in bone mass and altered in calcium and vitamin D metabolism along with phosphorus. Estrogen stimulates the osteoblast by decreasing the number and activity of osteoclast which in turn affect the bone remodeling.<sup>5</sup> The reduced calcium absorption has been attributed to reducing in circulating vitamin D levels and to gastrointestinal resistance to its action.<sup>6</sup> Accordingly, vitamin D is to help in maintaining the calcium balance of bone by promoting calcium absorption in the intestine by promoting bone resorption through an increase in osteoclast number.

Vitamin D also maintains the level phosphorus for bone formation and allows proper function of PTH hormone to maintain serum calcium levels. After the onset of menopause in women life, there is increased risk of osteoporosis, muscle weakness.

### **AIMS & OBJECTIVES**

#### **➤ The aims of the study is**

To correlate the Serum level of Bone Markers (Vitamin D<sub>3</sub>, Calcium, Phosphorus, ALP), Oxidative Stress Marker (Malondialdehyde) along with the level of Total antioxidant status in Pre & Post-menopausal Women

### **MATERIALS AND METHODS**

This was a cross-sectional type of study, carried out in the Department of Biochemistry, LN Medical College Bhopal, M.P. Total three hundred ten healthy women were included, out of which 155 were healthy pre-menopausal and 155 were postmenopausal women. Ethical committee and written informed consent were obtained from study subjects.

#### **Inclusion criteria:-**

The women were apparently healthy. Control Group: - Women with the reproductive age group 30 – 45 years, with a normal menstrual cycle. Study Group: - Post-menopausal women 46 – 60 years, with one year of amenorrhea and were not receiving any hormonal replacement therapy.

#### **Exclusion criteria:-**

The women with some sort of menstrual disorders e.g. Irregular menses, menorrhagia, with any bone fracture in previous 1 year, on hormonal replacement therapy, oral contraceptives, smoker, alcoholic, under any estrogen therapy or any supportive treatment for menopausal symptoms for at least 6 months prior to study, supplementation with nutritional antioxidants, diabetes, hypertension, malabsorption and any bone diseases were excluded from this study.

**Biochemical Parameters:** Venous blood (5ml) was drawn from all participants and serum was separated. Serum was analyzed for the measurement of Vitamin D [ElectroChemiluminascence binding assay, Serum Calcium [O-Cresolphthalein Complex method and Serum Phosphorus

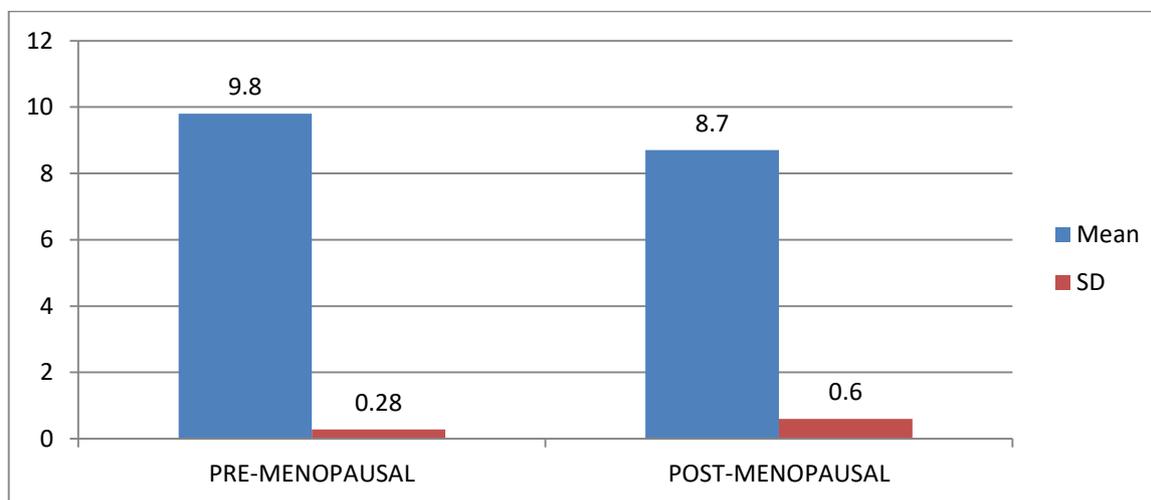
[Modified Gomorri's Method Statistical Analysis: All data obtained from the estimations were expressed as the mean± standard deviation (SD) and student t-test was used for compare mean± SD between the group.

## RESULT

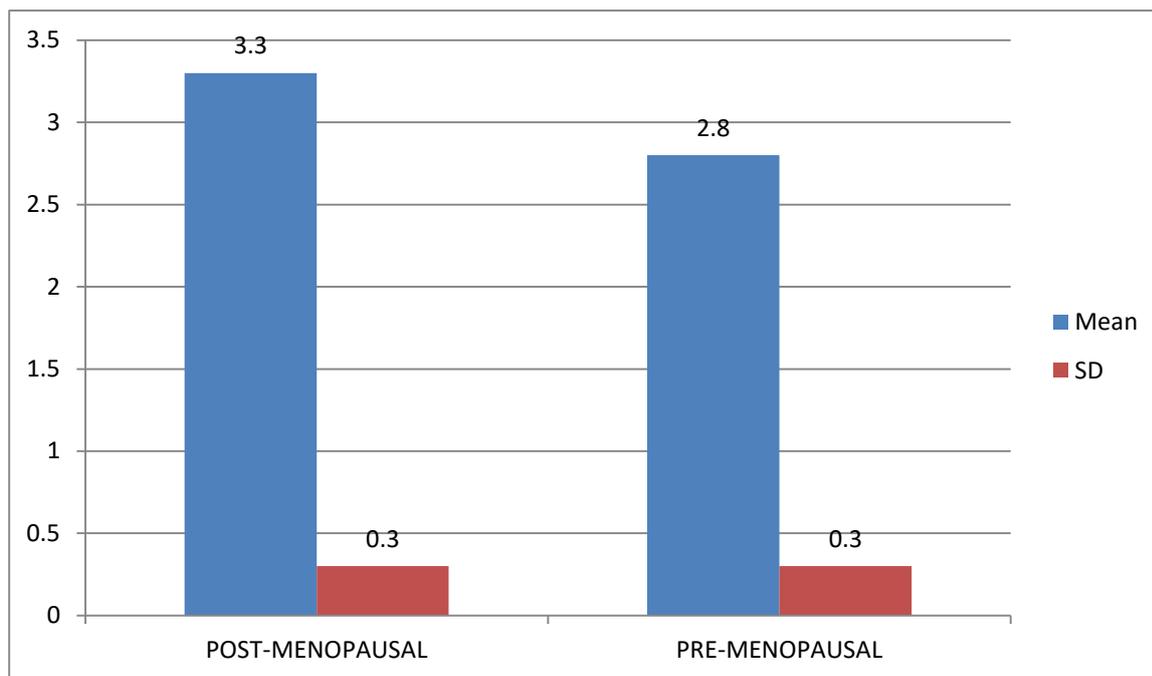
**Table : - Biochemical Markers in Premenopausal & Postmenopausal women.**

S. No	Parameters	Premenopausal women	Postmenopausal women
1.	Vitamin D (ng/ml)	19.7±2.5	11.5±1.8
2.	Calcium (mg/dl)	9.8±0.28	8.7±0.6
3.	Phosphorus(mg/dl)	3.3±0.39	2.8±0.3
4.	MDA (µmol/ml)	1.55±0.46	2.84±0.69
5.	Antioxidant Status (µmol/L)	15.12±2.42	10.53±2.54

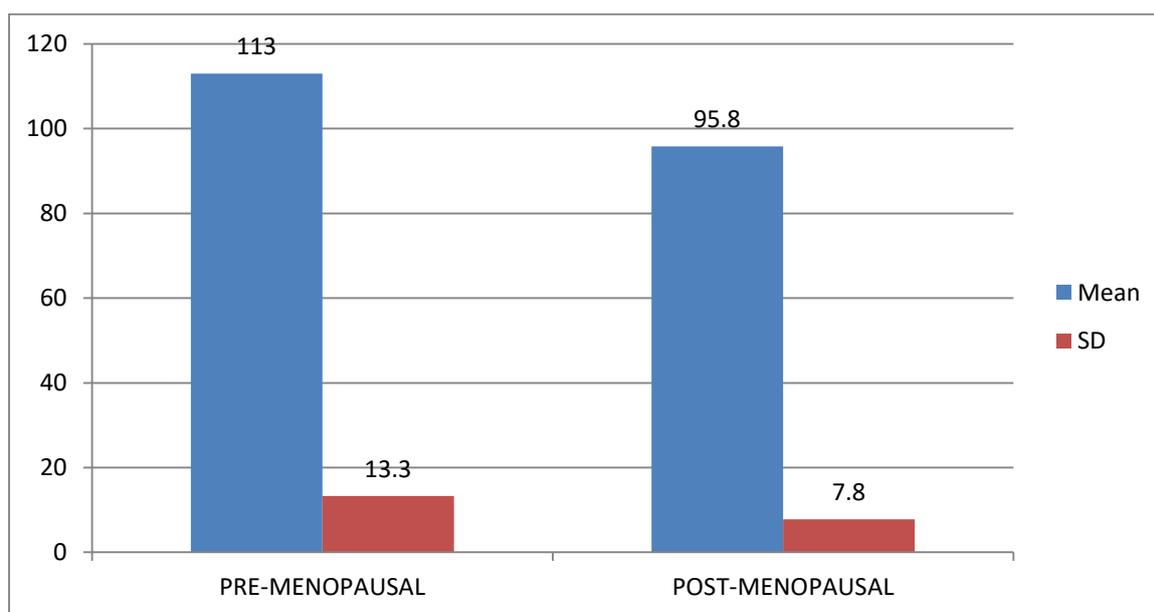
All the value are represent Mean & SD between the Pre and Post Menopausal women.



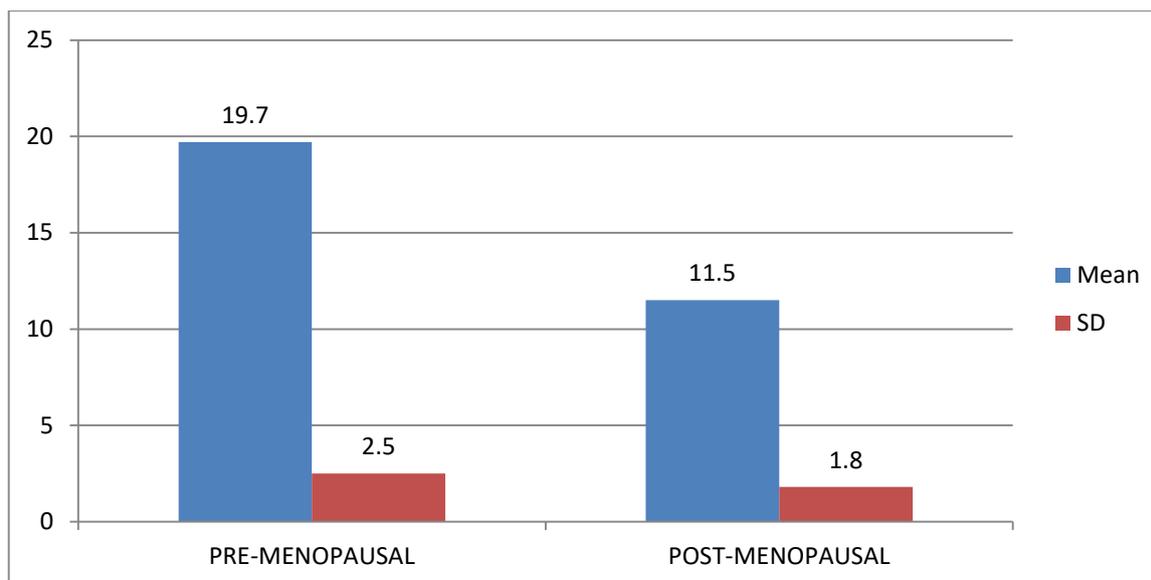
**Fig:-1 Mean and SD of Calcium in Pre and Post Menopausal Women.**



**Fig:-1 Mean and SD of Phosphorus in Pre and Post Menopausal Women.**



**Fig:-1 Mean and SD of Alkaline Phosphates in Pre and Post Menopausal Women.**



**Fig:-1 Mean and SD of Vitamin-D in Pre and Post Menopausal Women.**

## DISCUSSION

Bone is a dynamic tissue that is being constantly remodeled throughout life in a balanced way between formation and resorption is disturbed leading to osteoporosis during menopausal period.<sup>7</sup> Osteoporosis is one of the most common morbid conditions in postmenopausal life that occur due to the impact of hormonal changes, markedly reduces the quality of life.<sup>8</sup>

The present study shows that the Menopausal women were characterized by altered bone markers, Oxidative stress marker and Antioxidant status during the Postmenopausal age.

Several studies have shown that low serum vitamin D and calcium levels and high PTH levels are most important risk factors in osteoporosis.<sup>9</sup> Vitamin D deficiency (defined here as  $\leq 20\text{ng/mL}$ ) is almost less than half of the study population i.e., among postmenopausal women (47.74%). In our study we also found out that more than half of the postmenopausal women (52.25%) had serum vitamin D levels lower than  $10\text{ng/mL}$  Vitamin D deficiency (defined here as  $\leq 20\text{ng/mL}$ ) is almost less than half of the study population i.e., among

postmenopausal women (47.74%). In our study we also found out that more than half of the postmenopausal women (52.25%) had serum vitamin D levels lower than  $10\text{ng/mL}$ . In our study we found that the Calcium level of Premenopausal women  $9.8\pm 0.28$  found to be higher as compared with Postmenopausal women calcium level  $8.7\pm 0.6$  with p-value is  $\leq 0.0001$ . Decline in ovarian function during menopause is accompanied by decrease in bone mass and altered calcium metabolism. Decreased estrogen level affects the bone remodeling by stimulating osteoclast, decreasing number and activity of osteoclast and synthesizing cytokine affecting bone resorption. In postmenopausal women, the mechanism by which ovarian hormone modulate negative calcium balance is not fully understood however estrogen deficiency may induce calcium loss due to decreased intestinal calcium absorption and decreased renal calcium conservation, with a possible rise in gut calcium excretion.<sup>10</sup> In the present study, the MDA levels were found to be higher in postmenopausal women ( $2.84\pm 0.69$ ) as compared to premenopausal women ( $1.55\pm 0.46$ ). This study results are consistent with the

findings of a previous study comparing MDA levels between fertile and postmenopausal age groups.<sup>11</sup> Furthermore, recent two study done on lipid peroxidation and lipid metabolism in postmenopausal women and the results of the study suggest that the aging and lack of estrogen may be responsible for the increase in MDA levels.<sup>12</sup> In the present study, mean antioxidant status in postmenopausal women ( $10.53 \pm 2.54$ ) was significantly lower than that of the premenopausal women ( $15.12 \pm 2.42$ ). Lubov Kolesnikova<sup>13</sup>, Natalya Semenova<sup>14</sup> et.al was found same result antioxidant lowered in postmenopausal women when compared with premenopausal women and suggested that this could be due to estrogen deficiency.

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