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Evaluation of vitamin D3, calcium and parathyroid hormone among women with osteoporosis in Wasit Province, Iraq

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Abstract--Background: Osteoporosis (OP) is one of the most common metabolic diseases, characterized by low bone mass and abnormal bone microarchitecture leading to the development of fractures and subsequent disability. Disruption of bone remodeling by osteoporosis results in an imbalance between bone formation by osteoblasts and resorption by osteoclasts. **Aims:** This study was aimed to investigate relationship between vitamin D3, calcium and PTH, with estimating their effects on osteoporosis in women and the benefits in diagnosis. **Materials and method:** In this study, 50 women who were referred to DEXA unit in Al-Karama Teaching Hospital to diagnosed with Osteoporosis. They were compared with 40 healthy individuals who did not have any clinical symptoms. Blood samples were used after separating them by a centrifuge for the purpose of examining levels of vital indicators were detected by Enzyme linked immunosorbent assay (ELISA). **Results:** The results of the current study showed a decrease in the level of Vitamin D3 and calcium, increasing in the levels of Parathyroid hormone, with significant differences between the study groups at $P \leq 0.0001$ compared to the control group. **Conclusion:** Through the study it is clear that osteoporosis is an age-related disease and vitamin D, calcium and PTH play a role in their effect on the bones leading to fragility, and therefore can be used to detect and evaluate the disease.

Keywords--Osteoporosis, Vitamin D3, Calcium, Parathyroid hormone.

Introduction

Osteoporosis is a metabolic bone disease of the skeleton caused by bone metabolism disorders, and is the most common bone disease in humans (1). It is characterized by a decrease in bone mass and increased bone fragility (2, 3). The mechanism of osteoporosis is mainly the excessive bone resorption caused by the increased osteoclast, and the decreased bone formation caused by the decreased osteoblast (4). Although it is seen in all age groups, gender, and races, it is more common in Caucasians (white race), older people, and women. Studies have shown that bone loss starts from the age of 30-40 years in both men and women (5). Sex is a well-established non-modifiable factor for osteoporosis. The risk of osteoporosis is greater in women than in men and is a major health issue because the occurrence of certain types of fractures (hip, vertebra, upper humerus, pelvis, upper leg, several simultaneous and spine due to accelerated bone turnover secondary to estrogen deficiency (6, 7). With an aging population and longer life span, osteoporosis is increasingly becoming a global epidemic. The most commonly used technique for measuring bone mineral density is dual energy X-ray absorptiometry (DEXA) (8). Also various parameters are now available that allow a specific and sensitive assessment of the rate of bone formation and bone resorption of the skeleton, such as Vitamin D3 that is considered an important nutrient in the maintenance of bone health. Its decrease because of insufficient sources of endogenous and exogenous, may eventually result in bone mass reduction, especially in elderly women (9).

Calcium is one of the major mineral components of the skeletal system, found primarily in bone (99%), blood and ECF. Adequate calcium intake is essential for normal growth and development of the skeleton and teeth and for adequate bone mineralization (10). PTH plays an essential role in the maintenance of calcium homeostasis in mammals (11). The main function of the hormone is to maintain calcium levels within the normal range through its action on the bone, kidneys, and intestine. It also decreases serum phosphorus through inhibiting renal reabsorption (12).

Materials and Methods

Study samples

The study samples included 90 samples of females from different age groups ranging from (25-65) years (osteoporotic women), as they were divided into two groups, a group of Post-menopausal osteoporosis patients from (45-65) years, and a group of Pre-menopausal osteoporosis patients from (25-45) years and compared with the control group: 40 samples of apparently healthy people were taken into account, in terms of age for the group of patients.

Sample collection

Samples were collected from Al-Karama Teaching Hospital and private laboratories in Wasit Governorate for the period from November 2021 to January 2022 after taking information from patients and healthy people and filling out the form for each person. After the area of blood drawn was sterilized with cotton and diluted alcohol 70%, 5 ml of venous blood is withdrawn for patients and healthy people, then placed in glass tubes container on gelatin in order to

separate the clotted part of the serum from the blood. Leave for 1-2 hours at room temperature for complete coagulation and gloat formation. Then the blood samples are separated using a centrifuge at 3000 rpm for (10 minutes)The serum was then transferred to test tubes (Eppendorf tubes) by micropipette and stored at (-20°C) until biochemical tests were performed using the enzyme-linked immunosorbent assay (ELISA).

Statistical Analysis

All obtained data were documented using the Microsoft Office Excel (2016), and analyzed statistically by the GraphPad Prism (2012) software. One-Way Analysis of Variance (ANOVA) and t-test were applied to detect significant differences between values of the measured marked as well as among different age groups. Significant differences were represented as $P < 0.05$ (*), $P < 0.01$ (**), $P < 0.001$ (***), and $P < 0.0001$ (****).

Results

The results of the current study showed, as shown in Table (1) the Mean and Standard Deviation (SD) of the level (Vit.D3, calcium, PTH) in the study groups. The level of Vit.D3 in the patients group was (21.49 ± 0.779) and in the control group (34.26 ± 0.997) . The difference was significant ($p < 0.0001$) between study groups. The level of Ca in the patients group was (8.778 ± 0.09443) and (9.378 ± 0.07725) in the control group with a significant difference ($p < 0.0001$), for the levels of PTH (63.04 ± 1.139) in the patients group and (46.63 ± 1.151) in the control group with a statistically significant difference ($p < 0.0001$).

Table (1): Levels of Vit. D3, Calcium, PTH in the study groups

Parameters	OP Patient (N=50)	Controls (N=40)
	Mean \pm SD	Mean \pm SD
Vit.D3 (ng/ml)	21.49 \pm 0.779 B	34.26 \pm 0.997 A
Ca (mg/dl)	8.778 \pm 0.09443 B	9.378 \pm 0.07725 A
PTH (pg/ml)	63.04 \pm 1.139 A	46.63 \pm 1.151 B
Variation in horizontal large letters refers to significant differences at $P < 0.05$.		

Concerning age, Vitamin D3 and Calcium significant lowest value ($P < 0.0001$) was observed in P2 (postmenopausal osteoporotic women) when compared to P1 (premenopausal osteoporotic women) as well as the values of C2 (postmenopausal healthy women) and C1 (premenopausal healthy women). In addition, the value of P1 was decreased significantly in comparison with those of C1 and C2.

As for PTH significant differences ($P < 0.0001$) were reported in values of study groups, the findings of both patient group; P1 and P2 were revealed significant increases ($P < 0.0001$) when compared to those of control group, C1 and C2 .Also, the study showed that the value of C2 was higher than value of C1, and the value of P2 was higher than this of P1.(Table 2; Figures 1, 2 and 3).

Table (2): Association between age and levels of Vit. D3, calcium, and PTH

Parameters	P1 (N=25)	P2 (N=25)	C1 (N=20)	C2 (N=20)
Vit.D3 (ng/ml)	24.18 ± 0.992 C	18.8 ± 0.942 D	35.41 ± 1.412 A	33.105 ± 1.397 B
Ca (mg/dl)	8.904 ± 0.121 C	8.652 ± 0.143 D	9.45 ± 0.131 A	9.305 ± 0.083 B
PTH (pg/ml)	59.492 ± 1.296 B	66.596 ± 1.603 A	44.01 ± 1.251 D	49.245 ± 1.296 C

Variation in horizontal large letters refers to significant differences at P < 0.05 .

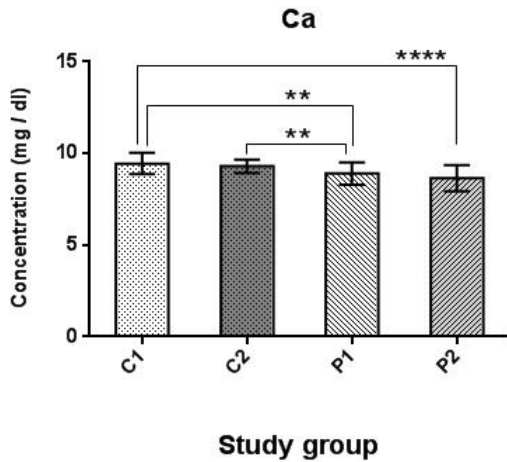


Figure (1): Association between the age and the levels Calcium in control and patient groups

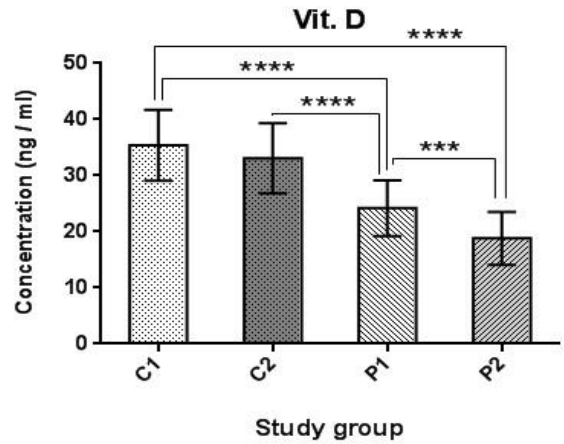


Figure (2): Association between the age and the levels of vitamin D3 in control and patient groups

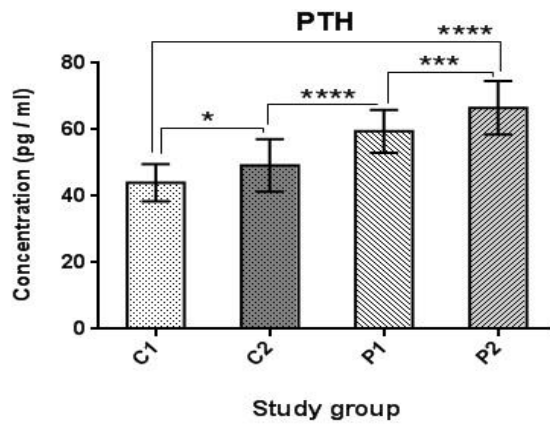


Figure (3): Association between the age and the levels of PTH in control and patient groups

Discussion

The data of this study showed significant difference in the level of serum Vitamin D3 between osteoporosis group and control group. Our study indicates very high incidence of hypovitaminosis D among postmenopausal women with osteoporosis and that's consistent with Nakamura et al. (13) as well as Albrahim and Binobead (14). This decrease in serum vitamin D3 levels may be due to less women's outdoor activity also with reduced exposure to sunlight and our long- resign habits will prevent vitamin D3 in the skin (15). Therefore, Prevention of vitamin D3 deficiency by appropriate diet, activity, sunlight exposure appears to be the primary prerequisite in reducing the incidence of osteoporotic fractures among women. Vitamin D3 is required for optimal calcium absorption and thus is also important for bone health. Indeed, in patients who are vitamin D3 deficient, no more than 15 % of dietary calcium is absorbed, whereas in persons who are not vitamin D3 deficient 30 – 80% of dietary calcium is absorbed (16).

In the current study serum Calcium showed significantly difference between osteoporosis groups and control groups. The results of this study are consistent with his findings Pardhe et al. (17) who mentioned that the calcium serum was significantly decreased in osteoporotic women ,and explained that may be due to the decrease of estrogen which causes bone resorption in postmenopausal women, preventing the absorption and utilization of bone calcium thus,causing the risk of osteoporosis. Our results also agreed with those of those who mentioned low calcium in people with osteoporosis (18).

On the other hand, researcher Rana found no significance difference in calcium level in osteoporotic women and in control group (19). Where Calcium levels are tightly regulated and homeostasis is maintained in serum regardless of their store in bone (hydroxyapatite), hence these parameters did not change significantly in patients with osteoporosis and controls .This may be due to the fact that the low level of vitamin D3 in the body negatively affects the absorption of calcium in the intestines, leading to lower levels in plasma and a rise in the level of thyroid hormone that compensates for this absorption by withdrawing calcium from the bones leading to its fragility and these results are consistent with the results of previous studies indicating that calcium deficiency leads to a decrease in bone mineralization (20).

Serum PTH in our study was significantly difference between osteoporotic groups compared to control group. our results are consistent with results of Ponnappakkam et al. (21) and Bhattarai et al. (22) who found a rise in thyroid hormone levels in patients with osteoporosis. Khedr (23) noted that PTH can produce metabolic or anabolic effects on bone metabolism depending on the body's need.

While Martins et al. (24) noted that there was a relationship between PTH and vitamin D3 deficiency, the presence of sufficient amounts of vitamin D3 maintains the level of calcium in the blood, thus preventing the high level of PTH which stimulates the exit of calcium from the bones and leads to reabsorption and bone remodeling and then bone loss as a result, the risk of fractures increased.

Conclusion

We concluded from this study a Positive strong correlation appears clearly in this study between age and osteoporosis and this indicate that it is an age- associated diseases. a positive correlation was found between both serum vitamin D3 levels and Calcium, and anegative correlation between PTH and both serum vitamin D3 levels and Calcium. Vitamin D, calcium and PTH play a role in their effect on the bones leading to fragility, and therefore can be used to detect and evaluate the disease.

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