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Original Article

Osteoprotegerin, Parathyroid Hormone and Vitamin D as Effective Factors on Serum-Urine Calcium Levels in Breast Cancer Patients

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Abstract

Breast cancer represents one of the most popular kinds of cancer worldwide. During the early stages of the disease, the level of Osteoprotegerin remained within normal limits, showing that the bone was not being damaged to get calcium due to an increase in parathyroid hormone. The current study aimed to assess a number of biochemical variables in a group of women with malignant breast cancer who had reached menopause (less than 45 years old). One hundred thirty women were randomly divided into three groups as follows. The first group (G1) is made up of women who have never had breast cancer or any other disease, and their number (40) corresponds to the same age range (below menopause) as the control group. The second group (G2) comprises women diagnosed with breast cancer at an early stage whose numbers were relatively low (45). The third group (G3) included women of the same age who received one or two doses of chemotherapy and whose total number was (45) over the same period. The variables studied include Vitamin D, Parathyroid Hormone, Osteoprotegerin, blood calcium, and urine calcium, all of that are thought to play a role in the progress of the disease. Vitamin D levels were extremely low in the second group (G2), while they were slightly higher in the third group (G3) but remained extremely low. The first group (G1) maintained parameters within acceptable limits. There was a significant difference between the two breast cancer groups (9.38 1.43) and (4.98 1.67) when compared to the control group (20.04 2.80). (G1). The two breast cancer groups (G2) and (G3) had higher parathyroid hormone levels than the control group (G1), and there was a significant difference between the two breast cancer groups (136.52 58.56) (G3) and (G2) (167.79 35.21) compared to the control group (68.52 20.44) (G1). There was no significant difference in Osteoprotegerin levels between the two breast cancer groups (313.38 109.02) (G3) and (315.0 123.98) (G2) compared to the control group (G1) (324.11 104.73). The three groups' blood calcium levels were all within normal ranges, and there was no statistically significant difference between them (9.21 0.45), (9.23 0.38), and (9.23 0.38) (G3) (9.28 0.43). (G1), but urine-calcium levels were lower in both groups of breast cancer patients compared to the control group, and there was a significant difference between the two breast cancer groups (63.96 15.66) (G3) and (68.42 14.05) (G2) compared to the control group (213.77 63.94) (G1). In breast cancer patients, vitamin D deficiency and high parathyroid hormone levels were discovered, suggesting that vitamin D may play a role in cancer prevention. Osteoprotegerin levels were within normal ranges early in the illness, although this may alter as the patient matures and the disease advances.

Keywords: Breast cancer, Vitamin D, Osteoprotegerin, Parathyroid hormone

1. Introduction

During the early stages of the disease, the level of Osteoprotegerin remained within normal limits, showing that the bone was not being damaged to get calcium due to an increase in parathyroid hormone, but the results may alter as the patient grows older and the

condition advances. The breast is above the pectoral fascia anterior to the major pectoral muscle, precisely between the 2nd and 6th ribs. All breasts are composed of 15-20 lobes, while the lobe contains (20-40) lobules (1). Conjunctive and glandular tissue are connected through airways in nursing moms. In the nipple, milk is delivered by these ducts. The darker-pigmented region surrounding the nipple is known as the areola. The breast contains adipose and connective tissue, which surrounds and protects the ducts and gives shape to the breast. Breast growth is heavily influenced by female sexual hormones, particularly estrogens (mostly responsible for duct proliferation) and progesterone, primarily responsible for enhancing lobular cell differentiation (2). The same embryonic tissues give rise to the breasts in both boys and girls. During puberty, estrogens and growth hormones work together to induce breast development in female humans and other animals. Other women's breasts only grow during pregnancy. The breast's size and shape are determined by the network of ducts that converge on the nipple, which is covered by subcutaneous fat. Milk is generated and stored in lobules, or clusters of alveoli, at the ends of the ducts in response to hormonal cues (3). The essential function of the breast is to produce, store and release milk. Milk is produced in the lobules of women's breasts after giving birth when activated. During pregnancy, estrogens, progesterone, and prolactin combine to complete the breast's growth, particularly lobuloalveolar maturation, in preparation for lactation and nursing. Although the breast mainly consists of the mammary gland (glandular mammary) and subcutaneous connective tissue, it also contains lymphatic tissue-immune system tissue, which is responsible for removing cellular fluids and waste from the body (4). A Tumor is swelling in abnormal growth of tissue forming. It is either benign or malignant (5). Benign tumors are common types of breast tumors ordinary do not lead to cancers, but they play a critical role in increasing the risk of breast cancer growth in the future (6). Also, benign tumors do not affect invasion tissues as well as do not spread all over the body (7). Unlike benign tumors, malignant tumors grow fast, becoming massive clusters of cells that can transmit cancer to other body parts via the lymphatic system or bloodstream.

Calcium's role in carcinogenesis controls cell proliferation, differentiation, and apoptosis (8). Cell proliferation is reduced, and mammary cell differentiation occurs when calcium concentrations are increased in studies (9). Calcium supplementation has been demonstrated to inhibit 7,12-dimethylbenz anthracene-induced mammary gland epithelial hyperproliferation and mammary tumor genesis (10). Osteoprotegerin (OPG) was initially discovered to be a bone turnover inhibitor. The interaction of the nuclear factor kappa-B receptor activator and bone homeostasis maintains bone homeostasis (RANK), OPG, and RANKL, a soluble activation ligand. Osteoclasts are formed when bone marrow precursor (monocyte/macrophage) cells are prevented from developing into osteoclasts due to the binding of OPG to RANKL, which acts as a decoy receptor (11). Parathyroid hormone (PTH) plays a crucial role in health. Therefore, high levels of PTH, such as hyperparathyroidism, or low levels, like hypoparathyroidism, cause health problems (12). $1,25(OH)_2$ vitamin D, phosphate, and calcium are physiological variables influencing PTH mRNA levels in the parathyroid glands (13). The calcium-sensing receptor (CaSR), a GPCR triggered by ionized calcium and other cat ions, allows parathyroid cells to detect and respond to small changes in calcium circulation. Changing levels of extracellular calcium have a negative effect on PTH secretion (14). The kidney and skeleton are involved in PTH's actions, and PTH governs the kidney's treatment of calcium, sodium, phosphate, and hydrogen ions (15). Vitamin D (25(OH)D to 1,25(OH)2D, which improves bone cell activity and enhances calcium absorption from the stomach, is controlled by this enzyme (16).

The current study aimed to assess a number of biochemical variables in a group of women with malignant breast cancer who had reached menopause.

2. Materials and Methods

2.1. Patients and Control

Samples were taken from women between the ages of 25 and 45 before menopause at the Iraqi Medical City, Oncology Teaching Hospital, Department of Breast Cancer Early Diagnosis. It took five months to obtain the blood samples. Breast cancer control, new diagnoses, and treatment are all included in these studies. There were three groups in this project. One group comprised 40 Iraqi women's blood samples; the other two groups each contained 45. The first group is the control group (G1). The newly diagnosed people are in the second group (G2). Women who received the first and second doses of chemotherapy are in the third group (G3). Before menopause, the ladies were between the ages of 25 and 45.

2.2. Specimen Collection

Blood samples were obtained from healthy and breast cancer patients by drawing 5 ml of blood from each participant. For 10 minutes, the samples were spun at 3000 rpm in a centrifuge. The serum is then separated into portions and stored at -20 C until Sera Vitamin D, Osteoprotegerin (OPG), Parathyroid (PTH), and Calcium are analyzed. Then 5 mL of urine samples were obtained to determine the calcium level in the urine. The samples were centrifuged for 15 minutes at 3000 x g before being stored at -70°C until analysis.

2.3. Determination of Sera Osteoprotegerin (OPG), Parathyroid Hormone (PTH), and Vit D

Sera ofOPG, PTH, and Vit D were identified by using the enzyme-linked immunosorbent assay (ELISA), used human PTH and human Vit D, AccuBind Elisa kit, USA to detect serum PTH and serum Vit D, while serum OPG was detected by using human OPG ELISA Kit, My Biosource, USA.

2.4. Determination of Serum-Urine Ca^{+2}

Serum-urine calcium concentrations were detected using a spectrophotometric method (AGAPE, Switzerland) kit for serum and urine samples.

2.5. Statistical Analysis

This study's data was examined with the statistical program SPSS-24 (Statistical Packages for Social Sciences-version 24) for Windows 10. The mean + standard deviation displays the parameters (SD). The one-way ANOVA and T-test are used to assess differences between the groups. Correlation analysis relied on Pearson's coefficient of correlation. *P* values less than 0.05 are considered significant (S), *P* values less than 0.01 are referred to as high significant (HS), and *P* values larger than 0.05 are referred to as non-significant (NS).

3. Results and Discussion

3.1. Vitamin D in the Groups under Study

The results of 25 hydroxy vitamin D appeared, as shown in figure 1 and table 1. In all of the groups investigated, there was a highly significant difference. G1 with G2 (20.042.80), (4.981.67), *P*-value (0.001), G1 with G3 (20.042.80), (9.381.43), *P*-value (0.001), and G2 with G3 (4.981.67), (9.381.43), *P*-value (0.001), and G2 with G3 (4.981.67), (9.381.43), *P*-value (0.001). Contrary to the findings of the chemotherapy group and healthy women, women with malignant tumors had significantly lower vitamin D levels than women without malignant tumors.

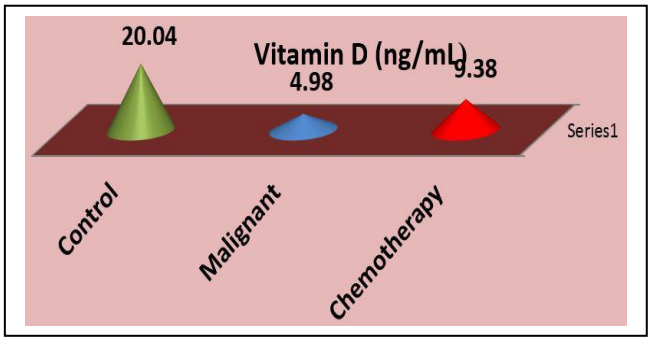

Figure 1. Vitamin D levels in three under study groups

Table 1. Comparison of Vitamin D levels between control and patient groups

Parameters Vitamin D (ng/mL)	Groups			*P*-value		
	Con. (G1) N=40	Mal. (G2) N=45	Chem. (G3) N=45	G1 & G2	G1 & G3	G2 & G3
Mean±SD	20.04±2.80	4.98±1.67	9.38±1.43	0.001	0.001	0.001

P=0.05 is significant, *P*=0.001 is highly significant, and *P*>0.05 is non-significant

When vitamin D levels were measured among women with breast cancer, the findings revealed a dramatic decline compared to the control group, although the results of the first doses of chemotherapy were somewhat higher than those of women newly diagnosed with breast cancer. This slight increase is because the women who took the first doses of chemotherapy were given high doses of vitamin D to compensate for their severe deficiency, which is part of the treatment mechanism in the first weeks of the disease. Breast cancer is linked to low vitamin D levels in several ways. This defect promotes cancer cell growth and delays programmed cell death. Another effect of this deficiency is by affecting calcium and parathyroid hormone levels.

According to local studies of vitamin D levels for women, the drop in vitamin D levels is acceptable in a society suffering from a shortage of vitamin D levels for women, and the reduction in vitamin D levels is separated into numerous portions (17). This significant vitamin D shortage in the blood will affect the condition. Vitamin D has been linked to breast cancer in various epidemiological studies. A recent study shows that at least 90% of women with breast cancer are vitamin D deficient and that high breast cancer incidence is linked to low vitamin D levels (18). Many types of cancer, including breast, colorectal, gastric, hematological, head and neck cancer, kidney, ovarian, pancreatic, liver, prostate, and skin cancer, have been related to low vitamin D levels in the blood. Vitamin D suppresses cell growth and promotes apoptosis. UVB exposure and/or [25(OH)D] concentrations have been linked to a lower risk of cancer in over 15 forms of cancer (19). This study focuses on premenopausal breast cancer women. Vitamin D deficiency has been linked to several diseases. Vitamin D's effect on estrogen receptors is important. Estrogen is crucial in breast cancer by inducing uncontrolled cell division, leading to tumor growth (20). Vitamin D has an estrogenic effect on the ability of chest cells to form tumors by increasing the Estrogen Receptor (ER). Vitamin D can help these women avoid breast cancer by reducing estrogen receptor expression and hormone production and signaling (21). Around 70% of breast cancer patients have estrogen receptor expression and activity. To decrease its activation (selective estrogen receptor modulators), production (aromatase inhibitors), and tamoxifen (Tam), a selective estrogen receptor modulator that competes with Estradiol (E2) by binding to Estrogen Receptor, is given (22).

3.2. Parathyroid Hormone (PTH) in the Groups under Study

The parathyroid hormone results are shown in figure 2 and table 2, according to the Mean±SD of G1, G2, and G3 showed the difference between G1 and G2 (68.52 20.44), (167.79 35.21), *P*-value (0.001) and G1 and G3 (68.52 20.44), (136.52 58.56), *P*-value (0.001) is extremely significant. However, there was no statistically significant difference between G2 and G3 (167.79 35.21), (136.52 58.56), and P-value (0.779). The statistical value observed increased parathyroid hormone in patient groups compared with the control group.

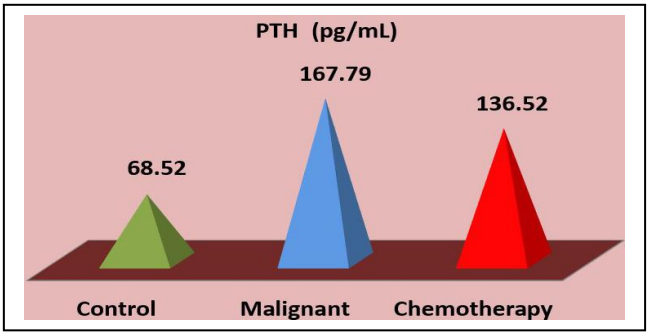

Figure 2. PTH levels in three under study groups

Table 2. Comparison of parathyroid hormone PTH levels between control and patient groups

ParametersPTH (pg/mL)	Groups			*P*-value		
	Con. (G1) N=40	Mal. (G2) N=45	Chem. (G3) N=45	G1 & G2	G1 & G3	G2 & G3
Mean±SD	68.52±20.44	167.79±35.21	136.52±58.56	0.001	0.001	0.779

P=0.05 is significant, *P*=0.001 is highly significant, and *P*>0.05 is non-significant

According to the findings, women with breast cancer had higher levels of parathyroid hormone, which is linked to poor calcium and vitamin D levels, one of the causes of breast cancer because high levels of parathyroid hormone raise cancer risk. A clinical study connected elevated PTH levels to an increased risk of breast cancer (23). Previous research has suggested that high levels of PTH are carcinogenic, tumor-stimulating, and linked to an increased risk of breast cancer (24). This study claims that PTH is carcinogenic and increases the risk of breast cancer. Previous research has linked elevated PTH levels to an increased risk of breast cancer (23). As seen in this study, the increased risk of breast cancer in women with hyperparathyroidism is further supported by a meta-analysis (25), which contradicts current findings demonstrating no substantial link between hypoparathyroidism and breast cancer risk. PTH is secreted by the parathyroid glands, which control calcium and phosphorus levels in the body. Another major impact of PTH on the kidney is that it promotes the conversion of 25-hydroxyvitamin D into 1,25-dihydroxy vitamin D, which is beneficial for the kidney (calcitriol). As can be seen, PTH indirectly stimulates an increase in 1-hydroxylase (CYP27B1) activity, which is the result of both up-regulation of the 1-hydroxylase gene and stimulation of the 1-hydroxylase enzyme, the active hormone that increases calcium absorption from the intestine is calcitriol, which is released into the circulation to create vitamin D (26).

A previous study looked at Vitamin D measurement levels and factors that affect stimulation in patients with breast tumors. On the other hand, measurement levels of PTH as another criterion on the accuracy of vitamin D measurements were also looked at, where the levels of this hormone rise when vitamin D levels are lower than those from measurements, and this is because one type of hyperparathyroidism, the expansion of this hormone, happens (27). Therefore women who have breast cancer showed high levels of calcium in the blood are caused by the effect of the hormone mentioned will have roles in the incidence of breast cancer, as well as diseases that accompany high levels of calcium in the blood, kidney stones and other diseases (28).

3.3. Osteoprotegerin (OPG) in the Groups under Study

The results of Osteoprotegerin (OPG) in figure 3 and table 3 showed a non-significant difference in all groups under study G1 with G2 (324.11 104.73), (313.38 109.02), *P*-value (0.941), G1 with G3 (324.11 104.73), (313.38 109.02), *P*-value (0.919), and G2 with G3 (315.0123.98), (313.38 109.02), *P*-value (0.998), respectively.

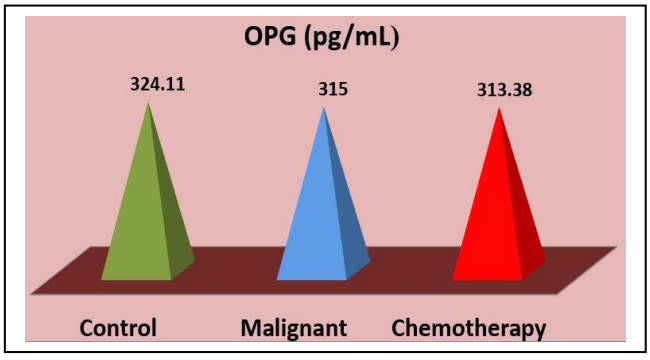

Figure 3. Osteoprotegerin (OPG) levels in three under study groups

Table 3. Comparison of Osteoprotegerin (OPG) levels between control and patient groups

Parameters OPG (pg/mL)	Groups			*P*-value		
	Con. (G1) N=40	Mal. (G2) N=45	Chem. (G3) N=45	G1 & G2	G1 & G3	G2 & G3
Mean±SD	324.11±104.73	315.0±123.98	313.38±109.02	0.941	0.919	0.998

P=0.05 is significant, *P*=0.001 is highly significant, and *P*>0.05 is non-significant

The OPG hormone levels of women with breast cancer were within normal ranges, and there was no significant difference between the two groups. This shows that the bones were not destroyed early in the illness. Despite the high levels of the parathyroid hormone, it did not affect bone breakdown, although osteoporosis is one of the methods used to restore calcium levels within the normal limits in the bloodstream. Normal estrogen levels regulate the reactions of OPG production and thus regulate the processes of bone destruction and building. These results represent the early stages of breast cancer, but in the advanced stages of the disease, women with breast cancer were given treatment for osteoporosis in advanced stages of treatment, and this may be evidence of bone destruction in the advanced stages of the disease.

Previous research demonstrated that tumor cells express the OPG protein, prompting an examination of the protein's role in tumor biology. OPG influences breast tumor activity, according to a growing body of evidence. Prior research examined OPG as a potential RANKL inhibitor in the bone microenvironment. Recent research has focused on OPG expression and interactions in RANKL-independent primary breast cancers. OPG may work with TRAIL, another TNF superfamily member, to decrease apoptotic induction in the primary tumor. Concern over OPG's tumor-promoting role in conjunction with BRCA1 gene abnormalities has increased. Functional studies have been summed up in previous research on OPG and breast cancer (29). OPG plays a role in tumor cell survival and cancer-induced bone damage. Because OPG is involved in bone turnover, most studies have focused on its role in tumors where bone disease is a common symptom, such as breast and prostate cancers, as well as the hematological malignancy of multiple myeloma. A previous study suggests OPG is involved in all three cancer types. A previous study has revealed that OPG protects breast and prostate cancer cell lines against TRAIL-induced apoptosis (30). Similar results have been demonstrated (31). According to this research, OPG may directly impact the survival of specific malignancies. Although this was only applicable to weakly differentiated hormone-independent cell lines in breast and prostate cancer cells, it is still unclear whether or not this is significant (30).

3.4. Calcium in the Groups under Study

3.4.1. Serum-Calcium

The results of serum calcium, as shown in figure 4 and table 4, shows a non-significant difference in all groups under study. G1 with G2 (9.28±0.43), (9.23±0.38), *P*-value (0.866) and G1 with G3 (9.28±0.43), (9.21±0.45), *P*-value (0.780) and also G2 with G3 (9.23±0.38), (9.21±0.45), *P*-value (0.986) respectively.

3.4.2. Urine-Calcium

The results of urine-calcium, as shown in figure 5 and table 5, according to the Mean±SD of control, Chemotherapy, and Malignant groups showed highly significant difference between G1 with G2 (213.77±63.94), (68.42±14.05), *P*-value (0.001) and G1 with G3 (213.77±63.94), (63.96±15.66), *P*-value (0.001) respectively but showed non-significant difference between G2 with G3 (68.42±14.05), (63.96±15.66), *P*-value (0.897).

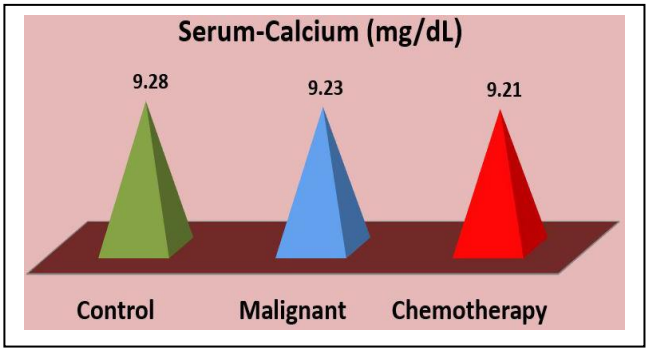

Figure 4. Serum-calcium levels in three under study groups

Table 4. Comparison of serum-calcium levels between control and patient groups

Parameters	Groups			*P*-value		
Calcium (mg/dL)	Con. (G1) N=40	Mal. (G2) N=45	Chem. (G3) N=45	G1 & G2	G1 & G3	G2 & G3
Mean±SD	9.28±0.43	9.23±0.38	9.21±0.45	0.866	0.780	0.986

P=0.05 is significant, *P*=0.001 is highly significant, and *P*>0.05 is non-significant

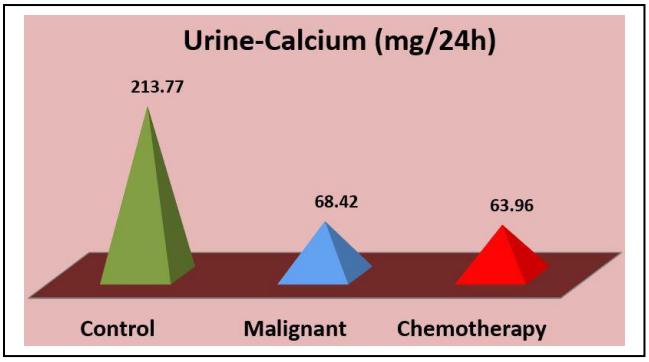

Figure 5. Urine-calcium levels in three under study groups

Table 5. Comparison of Urine-calcium levels between control and patient groups

Parameters	Groups			*P*-value		
Calcium (mg/24h)	Con. (G1) N=40	Mal. (G2) N=45	Chem. (G3) N=45	G1 & G2	G1 & G3	G2 & G3
Mean±SD	213.77±63.94	68.42±14.05	63.96±15.66	0.001	0.001	0.897

P=0.05 is significant, *P*=0.001 is highly significant, and *P*>0.05 is non-significant

Our research found that the calcium levels in the blood of both groups of breast cancer patients were average compared to the control group, indicating that the bones were not destroyed early in the disease. While the results of calcium levels in the urine showed a decrease in calcium levels for both groups of women with breast cancer compared to the control group; this indicates that the parathyroid hormone worked to reabsorb calcium from the kidneys and increase calcium absorption in the digestive tract to restore calcium levels within the normal limits in the bloodstream.

Calcium and its regulating hormones, such as parathyroid hormone (PTH) and vitamin D, have been linked to an increased risk of breast cancer in women (32). Vitamin D and calcium are two important dietary factors associated physiologically. Their anti-carcinogenic activities have been demonstrated in tests

using normal and malignant breast cells. Calcium's role in carcinogenesis stems from its ability to regulate cell proliferation, differentiation, and death (8). In experimental studies, raising the calcium concentration causes cell proliferation to slow down and the differentiation of mammary cells to occur in mammary cells (9). There is evidence that calcium's anticancer properties are mediated via vitamin D. Calcium, for example promotes apoptosis in vitamin D-induced breast cancer cells (33). Calcium inhibits fat-induced breast cell proliferation by conserving intracellular calcium concentrations (10). Breast cancer risk can be increased by a number of variables, including vitamin D and calcium, which are metabolically integrated and tightly connected dietary components (34).

When serum-calcium levels fall, the parathyroid glands respond by releasing enough PTH, causing the increase of hormone secretion mentioned to return blood calcium to an average level (35). High levels of parathyroid hormone stimulate the excretion of calcium (Ca2+) from bones into the bloodstream to fill the lack of blood calcium levels (hypercalcemia), which exposes the bone to get soft and osteoporosis (16). On the other hand, low levels of calcium are caused by low levels of vitamin D, which lead to high levels of PTH and raise calcium concentrations linked to benign tumors and subsequent breast cancer risk (36).

The normal mammary gland relies heavily on calcium metabolism. Calcium is transported from the maternal blood to the milk via a high concentration gradient during lactation. This is accomplished through the mammary gland's production of PTH-related peptides (PTHrP). As a result, during lactation, the breast acts as an extra parathyroid gland (32). Calcium, PTH, and 1,25OH2D have receptors in the mammary gland. Proper breast physiology is dependent on calcium and the hormones that control it. Cell signaling, growth, and death are all aided by intracellular calcium, a critical messenger (37). Normal and malignant breast cells express CaSR, which has been related to skeletal metastasis in breast cancer patients (38). Extracellular calcium levels may be increased in experimental conditions to promote cell differentiation, reduce cell proliferation, cause apoptosis, and down-modulate invasion, all of which have anti-tumor effects (39). Although calcium levels in benign breast tissue have been linked to an increased risk of breast cancer, another study found that increasing extracellular calcium levels induced the growth inhibitor 1,25OHD to be released from breast cancer cells in another investigation (40).

Some evidence found that high extracellular calcium levels have a similar effect to estrogen's "estrogen-like" effect in vitro, and the effect of estrogen and its role in the incidence of breast cancer was mentioned, which effect through a high concentration of calcium within the cell content through reactions between calcium-sensing receptor, calcium receptor and ER. So the effect of calcium on other receptors is indicated by the ER, which can explain the effect of the presence of calcium, like to estrogen effect, and the last will play a major role in causing breast cancer, So the effect of calcium ions through receptors found in both healthy chest cells and cancer cells which calcium ions have a similar effect of estrogen (41). There may be a decrease in calcium levels in the food intake, which can influence the metabolism of vitamin D and thus high levels of parathyroid hormone; therefore, Vit.D, PTH, and Ca levels are related (35).

Earlier studies linked increased calcium and vitamin D consumption to a lower incidence of premenopausal breast cancer. Premenopausal women's protection may be more apparent in cases of more aggressive breast cancers. The relationships between calcium and vitamin D intake and postmenopausal breast cancer were unaffected by tumor features. The preliminary study found that increased calcium and vitamin D intakes were related to a lower risk of breast cancer in premenopausal women but not after menopause. Premenopausal and postmenopausal women who ingested calcium and vitamin D had a lower risk of breast cancer 10 (42). Breast cancer risk is highly linked to mammographic breast density. Women with breast density in more than 75% of their breasts have a

four to five times higher risk of breast cancer than those with little or no breast density (43). Mammographic breast density has been indicated as a suitable intermediate objective for predicting breast cancer risk. Vitamin D and calcium supplementation reduced breast density in premenopausal and postmenopausal women (44). A recent study indicated that premenopausal women's breast density was related to dietary calcium and vitamin D consumption, but not supplementary calcium and vitamin D. No significant links were found between calcium and vitamin D consumption and postmenopausal women's breast density (45).

3.5. Correlation Coefficient of Parameters under Study

The correlation coefficient value showed the Pearson correlation coefficient (r) and P-value of vitamin D versus Age, BMI, PTH, OPG, and Serum-urine Calcium, as shown in figures 6, 7, 8, 9, 10, and 11 and table 6.

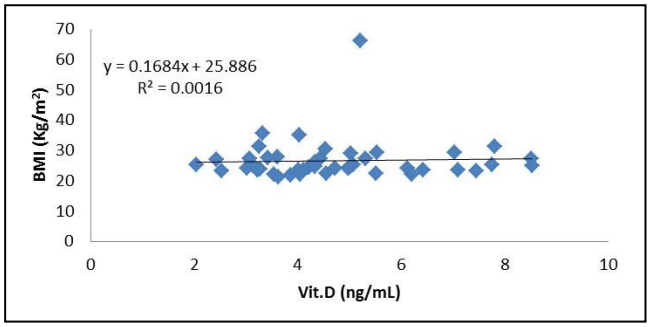

Figure 6. Vit. D vs. BMI

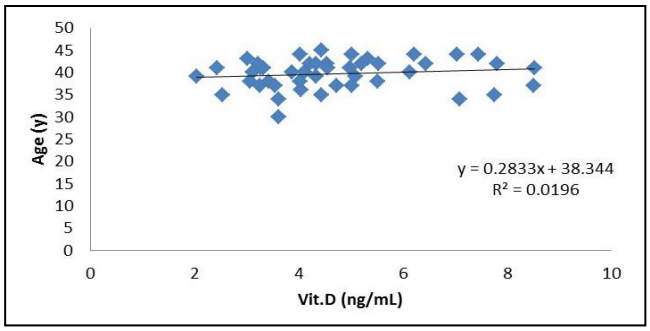

Figure 7. Vit. D vs. Age

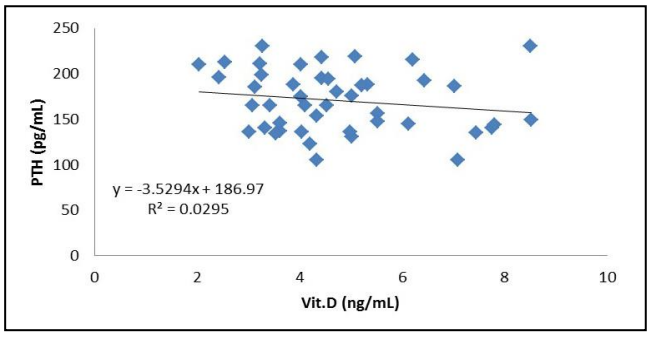

Figure 8. Vit. D vs. PTH

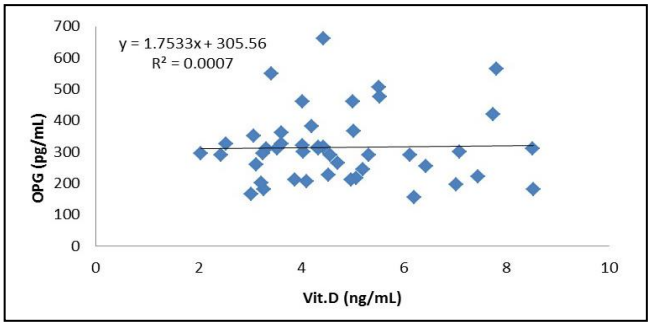

Figure 9. Vit. D vs. OPG

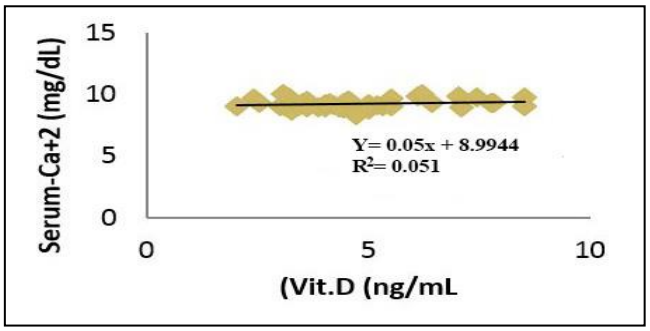

Figure 10. Vit. D vs. Serum-Calcium

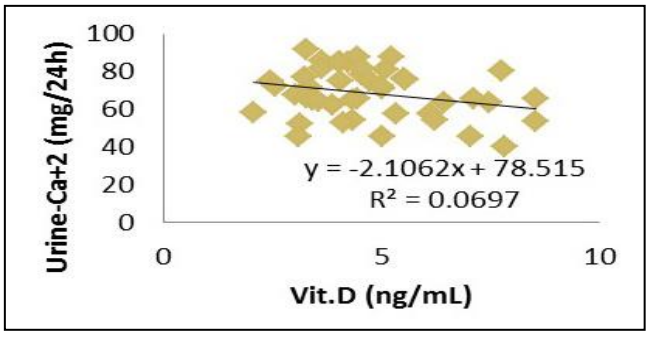

Figure 11. Vit. D vs. calcium in urine

Table 6. The correlations of vitamin D and parameters under study in early diagnosis patients

Parameter	r	*P*-value
Age (Y)	0.140	0.359
BMI (kg/m^2)	0.040	0.794
PTH (pg/mL)	-0.172	0.259
OPG (pg/mL)	0.026	0.867
Serum-Ca^{+2} (mg/dL)	0.260	0.085
Urine-Ca^{+2} (mg/24h)	-0.264	0.080

Vitamin D insufficiency and high parathyroid hormone level were found in breast cancer patients, indicating that vitamin D may play a role in cancer prevention. During the early stages of the disease, the level of Osteoprotegerin remained within normal limits, showing that the bone was not being damaged to get calcium due to an increase in parathyroid hormone, but the results may alter as the patient grows older and the condition advances.

Authors' Contribution

Study concept and design: E. M. E.
Acquisition of data: E. M. E.
Analysis and interpretation of data: A. H. J.
Drafting of the manuscript: A. H. J.
Critical revision of the manuscript for important intellectual content: A. H. J.
Statistical analysis: A. H. J.
Administrative, technical, and material support: A. H. J.

Ethics

The study protocol was approved by the medical ethics board of the AL-Zahraa University for Women, Kerbala, Iraq.

Conflict of Interest

The authors declare that they have no conflict of interest.

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