The Influence of Some Vitamins and Biochemical Parameters on Iraqi Females' Patients with Malignant Breast Cancer

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Abstract
Background: Cancer is one of the most common and deadly diseases in humans, and breast cancer (BC) is the second type of cancer among women, and it is the second cause of death, while lung cancer is the first, in agreement with the World Health Organization (WHO). Objective: To find the effect some vitamins and their relationship on the development of malignant breast cancer for Iraqi women. Methods: For purpose of starting the study, we decided to take (80) women as a sample of this study and they were split into two categories, the 1st category include (40) females as patients related breast cancer who taking vitamins pills as treatment, and the second group include (40) women as healthy subjects. Both groups had tests such as vitamins D, E and A, malondialdehyd (MDA), liver function Aspartate Aminotransferase (AST/ GOT) and Alanine transaminase (ALT/ GPT). Body mass index (BMI) and estradiol hormone (E2) concentration were measured as well. The patients were given vitamin D as (D3) pills (5000/ IU), vitamin E pills (400/IU), and vitamin A pills (10000/IU) one pill per day for a month. All vitamins as well as MDA were measured after taking medication for a month. Results: After laboratory measurements, the results were statistically analyzed. There was a strongly noticeable where ($P<0.001$) variation in vitamin D when comparing patient group with control. Vit E, A, MDA, BMI. When comparing the estradiol hormone of the healthy group with the group of patients, there was a statistically significant relationship ($P\leq0.05$). On the other hand, we did not notice any significant relationship with regard to liver enzymes for the same groups ($P>0.05$). Vitamins D, E and A as well as MDA concentrations were elevated after taking medication for a month. Conclusions: Maintaining the normal level of vitamins in addition to maintaining a normal body weight plays an important role in reducing the development of breast cancer.

Keywords: Breast Cancer (BC), Vitamin D (Vit D), Body Mass Index (BMI).

1. Introduction

The steady and rapid is developing of cells that leads to the creation of groups
Whose cell division cannot be controlled and which may spread more widely in the rest of the body is as a malignant tumor [1].
Vit D is a fat-soluble vitamin and has two types D2 and D3. The first step in the formation of vitamin D inside the human body begins from the compound 7-dehydrocholesterol, which is converted into pre-vitamin D3 by radiation reaching the human skin, where the wavelength of these rays is estimated (290-315) nanometers.
A decrease in the level of vitamin D in patients with breast cancer makes chemotherapy or radiotherapy less efficient than if the levels of vitamin within normal values and as recent research suggests [2, 3].
One of the most important vitamins that is considered a very effective antioxidant in fighting diseases, especially breast cancer, is vitamin E, which is a fat-soluble vitamin. Vitamin E insufficiency in people causes fringe neuropathy which is hereditary turmoil caused by a transformation in the quality of frataxin and mitochondrial restricting protein [4].
One of the important vitamins to protect the skin and support important parts of the digestive system, lungs, as well as the eye, and plays an important role in the development of the human body, especially for infants. All these benefits are for vitamin A [5].
Malondialdehyde is an organic compound with a low molecular weight di aldehyde or propane have two oxo groups with the formula CH2 (CHO) 2. Arachidonic acid induces the platelets to produce large amounts of MDA. MDA can generate from reaction between free radical such as singlet oxygen, superoxide anions, and hydrogen peroxide and hydroxyl radical, and polyunsaturated fatty acids through process result of the lipid peroxidation [6].
Alanine amino transferase (ALT) is consider cytoplasmic enzyme it's found in hepatocytes. Skeletal muscle, renal, and red blood cells, however smaller amounts from ALT was released into the circulation when hepatocytes damage or increase cell membrane permeability so it's a sensitive marker for hepatocellular injury [7].
Aspartate amino transferase (AST) is found in cytoplasm and mitochondria of liver and other cells like heart and muscles. So that increase AST levels didn't give specific marker for liver damage because ALT levels increase too reference above. By the way, in breast cancer AST and ALT test must be measured to check liver function because high levels in these enzyme means cancer has spread to the liver [8].
Being overweight is one of the most important health problems facing patients in general and cancer

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patients in particular. It is a health problem that has become global and can be greatly reduced or eliminated by following a healthy diet. Weight gain from (25_29 Kg/m²) is excessive obesity. Breast cancer is the first among the wide spread cancers and is the cause of cancer-related deaths worldwide [9].

Estradiol hormones (ES) are female gender hormones redound in development and growth of female genital and female gender characteristics, E2 are created in the ovary, placenta, testes and adrenal cortex, Naturalist ES are steroid hormones which have C18-carbon skeleton, and have three basic biologically active forms; Estrone (E1), ,17-β Estradiol (E2) and Estriol (E3). E2 is the elevated biological activity. E2 or 17-β Estradiol has, 2 hydroxyl groups, generated by the ovaries before menopause [10].

2. Materials and Methods

The study was conducted during 2020, (40) Iraqi female's patients diagnosed with malignant breast cancer were selected from Kadhimiya Teaching Hospital and the Tumor Teaching Hospital, an accurate medical history was obtained from the patients. As a control group, (40) normal females became involved in this work.

In this work, the sample was classified into two types: patients and control group, both of them in the age before menopause, with an average age ranging between 35-46 years.

Blood samples (5 ml) were collected in plain tubes, the blood was centrifuged at a rate of 3000 revolutions per minute for ten minutes. The sera were liquefied and frozen until the examination was performed.

We determine vitamin D levels by using a Microplate Enzyme Immunoassay, Colorimetric main reagents that required for a solid phase sequential enzyme immunoassay include inert antibody, enzyme antigen accompany and native antigen [11].

High performance liquid chromatography is one of the most powerful tools for the estimation of vitamins. Vitamins (A and E) were separated and quantitatively determined according to simultaneous determination of vitamin A and vitamin E in serum or plasma by liquid chromatography [12, 13].

Liver function Aspartate Aminotransferase (AST/ GOT) and Alanine transaminase (ALT/ GPT) enzyme measured by UV-assay according to IFCC (International Federation of Clinical Chemistry and laboratory Medicine) without pyridoxal phosphate activation.

The absorbance concentration was calculated for MDA in serum by Buege and Aust procedures, at λ max 535 nm [14].

E2 was assessment by Elisa kit depend on the principle of delayed competitive binding assay between E2 in the test sample and conjugated E2 enzyme for a constant amount of anti-estradiol monoclonal antibody epitopes (biotin reagent) [15, 16].

Body mass index (BMI) was calculated by equation as below

BMI = weight (kg)/ height (m2)

By the same techniques that were mentioned previously, the concentrations of vitamins D, E, A and MDA were measured in serum of patients group after taking the treatment for a month.

Statistical Analysis

Statistical analysis was carried out using the student t-test, where the mean was calculated for with standard deviation of two groups patients and healthy subject, P-value of <0.001 and <0.05 express as a highly significant and significant respectively, while p>0.05 express as a non-significant.

3. Results

In this study, (40) Iraqi women with malignant breast cancer were taken, the results we obtained were compared with a (40) healthy women as a control group, with an average age of (35-46) years for both groups. The mean±SD of Vitamin D for patients with breast cancer (3.95 ± 2.88) (ng/ml), while control group was (22.49 ± 6.25) (ng/ml). The mean±SD of vitamins E and A for patients (491.2 ± 185.5) (µg\dl) and (49.5 ±15.0) (µg\dl) compared to the healthy group were (522.5 ± 234.3) (µg\dl) and (59.1 ±19.2) (µg\dl) respectively. The mean±SD of MDA was (6. 1 ±0.21) (mg/dl) for patients compared to healthy control (4.8 ±0.31) (mg/dl) as shown in Table (1).

Table 1: Serum levels of vit D, E, A and MDA in serums of patients with malignant breast cancer and control group.

Parameters	Control (Mean±SD)	Patients (Mean±SD)	P-value
Vit. D (ng/ml)	22.49 ± 6.25	3.95 ± 2.88	P<0.001
Vitamin E (µg\dl)	522.5 ± 234.3	491.2 ±185.5	P≤0.05
Vitamin A (µg\dl)	59.1 ±19.2	49.5 ±15.0	P≤0.05
MDA(mg/dl)	4.8 ±0.31	6. 1 ±0.21	P≤0.05

In Table (2), the mean±SD of liver enzymes GOT and GPT was within the normal level for the group of patients, and we did not notice a significant difference, as the results were for the patients (19.54 ±6.17) (U/L) and (15.25 ±9.81) (U/L) while the results for control group (19.4 ± 7.23) (U/L) and (19.61 ± 10.55) (U/L) respectively. As for the body mass index, there was a significant difference in its value for patients (26.54±3.85) (kg/m2) compared with healthy subjects (29.52 ± 3.36) (kg/m2). The mean±SD of estradiol for patients with malignant breast cancer(54.5 ±15. 44) (pg./ml) while its value for healthy control(65.71± 20.11)(pg./ml).

Table 2: Serum levels of GOT, GPT, BMI and E2 in serums of patients with malignant breast cancer and control group.

Parameters	Control (Mean±SD)	Patients (Mean±SD)	P-value
GOT(U/L)	19.4 ± 7.23	19.54 ±6.17	P>0.05
GPT (U/L)	19.61 ± 10.55	15.25 ±9.81	P>0.05
BMI (kg/m2)	29.52 ± 3.36	26.54±3.85	P≤0.05
E2(pg./ml)	65.71± 20.11	54.5 ±15. 44	P≤0.05

After the patients were taken the medication, some parameters were measured to see the response of these patients to the treatment. We noticed an increase in the mean±SD of vitamin D, E and A after taking the medication, where their levels became(35.49 ± 8. 65) (ng/ml),(612.3 ± 152.1) (µg\dl) and (69.3 ±8.2) (µg\dl)while they were before taking medication (3.95 ± 2.88) (ng/ml),(491.2 ±185.5) (µg\dl) and (49.5 ±15.0) (µg\dl)respectively, and this led to an improvement in the patients' health compared to their health condition before taking this medication, where the mean±SD of MDA after taking the treatment decreased to(3.1 ±0.11)(mg/dl) while its level was (6. 1 ±0.21)(mg/dl) before the treatment. as shown in table (3).

Table 3: Serum levels of Vit D, E, A and MDA in serums of patients with malignant breast cancer and control group before and after taking medication.

Parameters	Before (Mean±SD)	After (Mean±SD)	P-value
Vit. D (ng/ml)	3.95±2.88	35.49 ± 8. 65	P<0.001
Vitamin E (µg\dl)	491.2±185.5	612.3 ± 152.1	P≤0.05
VitaminA (µg\dl)	49.5±15.0	69.3 ±8.2	P≤0.05
MDA(mg/dl)	6. 1±0.21	3.1 ±0.11	P≤0.05

4. Discussion

In this work, we found that the level of vitamin D is very low in the group of female patients when measured with healthy subjects, which means the consumption of this vitamin in patients, there were found differences between levels of vitamin D in control group and disease group of malignant tumor depending on statistical values , and it was observed that vitamin D in control group was increased because the normal values of vitamin D ranged between 30 and 100 ng/mL as sufficiency as we shown in table (1) [17].

In table (1) we note values of vitamin E for group of patients and healthy subjects (mean± SD). A lot of researchers have found that women who eat a vitamin E are not at very high danger of improving breast cancer, but it was found in many women after menopause that taking vitamin E did not have the same important effect in reducing the incidence of breast cancer [18, 19].

Several studies indicated that the use of radiation therapy for patients with advanced breast cancer leads to a low level of vitamin A and this is consistent with our study. The level of vitamin A for patients and healthy people can be observed as in table (1) [20].

The mean levels of MDA were (4.8 ±0.31) mg/dl, (6.1 ±0.21) mg/dl in the control group and malignant group respectively showed in the table (1). The purpose of the present study was to evaluate malondialdehyd in the pathogenesis of breast tumor patients and healthy group there were statistically significant serum MDA levels in breast tumor groups than in control group (p-value< 0.05) as shown in table (1). It was found that the biomarker of oxidative stress (Malon Didehyde) and as a result of the low level of antioxidants in patients with breast cancer increases significantly. The high oxidative stress is one of the most important factors that help the growth of breast cancer [21].

We conclude and according statistical values of GOT and GPT has been showed in table (2) while statistical values of GPT and GOT demonstrated no significant distributed between control and malignant groups, however mean ± SD of GOT for control (19.4 ± 7.23) U/L, and patients (19.54 ±6.17) U/L also statistical value for GPT control and malignant are (19.61 ± 10.55) U/L and (15.25 ±9.81) U/L respectively.

The results that obtained show the normal levels of GOT and GPT, so there are other reasons contributed to lower vitamin D levels may be due to inadequate sunlight exposure [22].

(Mean ±SD) of body mass index in sera of patients and healthy subject appear in table (2) as we show. Obesity and high body mass index, especially after menopause, increases the risk of breast cancer as a result of high level estrogen, and this stimulates to fatty tissue being a fundamental supplies of estrogen rather than having ovaries [23, 24].

When we measured the level of estradiol hormone for the two groups of patients and healthy people, we obtained the results indicated in Table (2).

It is clear to us a significant decrease in the level of the hormone in patients compared to healthy subjects. This is consistent with many studies and research that indicate the absence of a significant relationship in the high level of the hormone estradiol in patients with malignant breast cancer compared to healthy group [25].

In table (3), we notice a highly significant increase in concentration of vitamin D in group of patients after taking the treatment (vitamin D3 pills) for a month compared to the same group before taking treatment, and we also notice a significant increase in the level of vitamins, E and A concentration for the same group after taking medication for a month compared with its results before taking the treatment, while we notice a significant decrease in the level of MDA concentration after taking medication for a month [26]. The increase in concentration of vitamins D, E, and D leads to an increase in the rate of recovery in patients with cancer. An increase in concentration of vitamin E and A leads to a decrease in oxidative stress and therefore a decrease in the level of MDA as a biomarker of oxidative stress because vitamins E and An efficient antioxidants work to reduce the level of MDA [27].

5. Conclusion

In this work, we conclude that the level of vitamins plays an important role in development or reduction of breast cancer, as well as the oxidative stress represented by malondialdehyde, while we did not notice the important role of the level of liver enzymes.

The normal level of the hormone estradiol is of great importance in reducing the development of breast cancer in addition to maintaining a normal level of

body weight.

Ethical Clearances

Taken from the ethical and research committee in university of Baghdad.

Conflict of Interest

None declared.

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