

EVALUATION OF SOME TRACE ELEMENTS IN SERUM AND URINE FROM FEMALES BREAST CANCER (PRE AND POST MENOPAUSE) PATIENTS IN MISSAN CITY, IRAQ

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ABSTRACT

To investigate the concentration and role of certain important elements in 55 patients women diagnosis with breast cancer. The patient groups which divided into 2 groups: (30 patients with premenopausal) and (25 patients with postmenopausal) aged (20-60) years have been examined and formed the initial study group trace metals are essential to normal human homeostasis. When present in an abnormal expression, they contribute in many pathological processes. Our aim was to investigate the serum and urine concentration of some important elements Copper (Cu). Zinc (Zn). Selenium (Se). Aluminum (Al). Chromium (Cr). Lead (Ld). and Magnesium (Mg) of the patients with breast cancer, and (50) healthy control women it is found that there were statistically increased significant of (Copper, Lead. and Aluminum) in postmenopausal patients as compared with controls and pre-menopausal patients with $p \leq 0.001$. While the serum levels of (Zinc. Selenium. Chromium and Magnesium) in pre-menopause patients was increased significantly different from control and postmenopausal patients. There was no significant difference in the serum level of (Zinc. Selenium. Lead and Magnesium) between the groups of breast cancer patients. The urinary minerals that exhibited the levels of (Cu (in pre). Zinc (in post), Selenium (in pre), Lead (in pre and post). Chromium (in post) and Aluminum (in pre – post), a significant difference (increased) from controls

KEYWORDS: Breast Cancer, Pre and Postmenopausal Women, Trace Metals, Urine from Breast Cancer

INTRODUCTION

The breast is an external symbol of beauty and women hood; however cancer of the breast is responsible for the death of millions of women world every year ⁽¹⁾. The breast is collection of glands each adult female breast usually consists of (15-20) glandular lobes covered by amount of adipose tissue ⁽²⁾. Breast cancer is a heterogeneous most frequent disease in women. Female breast cancer is the second leading cause of cancer –related death. While male breast is rare ⁽³⁾. Breast cancer is systemic disease in that cancer cells may start to be disseminated in to blood and lymphatic systems even in early stages or when the tumor size still small ⁽⁴⁾. Early menarche has repeatedly been found to be associated with increased incidence of breast cancer ⁽⁵⁾. One of recent studies stated that breast cancer risk in women whose menstrual period begins before the age of 15 years ⁽⁶⁾. Postmenopausal women had a lower risk than premenopausal women of the same age. Among postmenopausal women the risk increased with increasing age at a menopause, whether menopause occurred naturally ⁽⁷⁾. Breast is constantly responding to the changes in hormonal, genetic, And Nutritional. Psychological and environmental stimuli cause continual cellular Changes ⁽⁸⁾. The alteration of a substance such as element intumormight help to demonstrate to potential of element to tumor and to resolve whether these elements could be used as supplemental biochemical markers' for the diagnosis and prognosis of tumor growth ⁽⁹⁾. Many of major elements are required for normal

or growth. Generality of the elements enter the human body mainly by the circulating blood and leave at most through urine and feces. Other mechanisms of elimination are secretion through sweat, milk, menses or semen⁽¹⁰⁾. It has become well decided that many trace elements play an essential role in of a number of biological processes through their action as activators or inhibitors of enzymatic reaction by contend with other elements and proteins for binding sites, that influencing the permeability of cell membranes. It is therefore reasonable to suppose that these trace minerals would exert action directly or indirectly on the carcinogenic process⁽¹¹⁾.

During the past several years, many have focused attention on the role of study element in serum from breast cancer⁽¹²⁾, this interest was stimulated by a series of early study by our that which is demonstrated in the urine studying the elements and compared them with serum.

Objectives of this study were to find out the level of some minerals (Cu, Zn, Se, Ch, Mg, Cd, Al) by atomic absorption spectrophotometer in serum and compared to urine from the breast cancer patients and controls to study the role of biomarkers in disease diagnosis for obtained the same patients for pre and postmenopausal breast cancer.

MATERIALS AND METHODS

Participants

Fifty five (55) newly diagnosed breast cancer patients who didn't have family history of cancer were recruited from surgical oncology clinics at the AL-Sadder hospital teaching and Al-mamonah oncology center for chemo and radiotherapy. Massin. Southern of Iraq.

All study participants were asked to complete a self – administered questionnaire that included questions on demographic factor medical history and health related behavior. In order ensure uniformity all patients and controls were chosen from the same ethnic (Missan) city and living in the same southern part of Iraq. Patients or controls with inflammations, women on regular haematinics or taking supplements women with diabetes mellitus. Hypertension, hepatitis jaundice. pregnancy or breast feeding subjects were exclude from this study.

Subjects

A total of fifty five women with diagnosed with breast tumor aged (20-60) years have been examined and formed the initial study group. These patients were subdivided in to two groups:-

Group 1

Consisted of with premenopausal 30 patients with premenopausal.

Group 2

Consisted of 25 patients with postmenopausal.

About forty healthy women with ages matching to the patient groups were included in this study as the control group (healthy) aged (21-54) years.

OUTCOME MEASURES

Height

This was measured and reading was recorded in meter.

Weight

This was estimated in kg.

Body Mass Index

This was calculated from the using the formula:-

$$\text{BMI (Kg/ m}^2\text{)} = \text{Weight (kg) / Height}^2 \text{ (m}^2\text{)}.$$

Collection of Blood Samples and Measurement of Trace Element Levels

Five mL of venous blood was obtained in to vacutainer plain tubes from patients. The blood sample was allowed to clot and retract after which it was centrifuged in (400 Searle / England) centrifuged machine at 4000 rpm for 10 min to obtain serum which was stored at – 20 C0 until ready for assay.

Collection of Urine Samples

Fresh urine samples were obtained from each cancer patients at Two point during the 1 (pre or to post menopausal).

Serum and the urine concentration of (Copper. Zinc. Selenium. Aluminum. Chromium. Lead. And Magnesium) were determined by atomic absorption spectrophotometer and flameless atomic absorption spectrophotometer ((Shimadzu Model 670. GU.7)

Statistical Analysis

Statistical evaluation was carried out with SPSS 19.0. The total patient group and the control group were compared with students (t) test. In all subgroups, parameters were subjected to Pearson correlation analysis ($p \leq 0.05$) was considered significant; data are a presented as Mean \pm S. D in the tables.

RESULTS

In this work the result represented the measurement of trace elements levels in serum and urine in a total of (55)breast cancer patients and (40) apparently healthy age – matched controls were recruited. The mean age of the breast cancer patients was 49.91 ± 2.48 years and that of controls was 50.28 ± 1.78 years.

Table 1- compares the demographic and anthropometric measurement of breast cancer patients with controls (healthy). The mean age. Mean BMI percentage. Body fat (PBF).age at (menopause. first pregnancy).number of smoker and number of miscarriages were highly significantly in (age of menopause, P.B.F) in pre or postmenopausal differently from the controls

Table 1: Comparison of Anthropometric Indices in Postmenopausal Women with Breast Cancer and Control

Parameters	Patients		Control	*P-Value
	Premenopausal	Postmenopausal		
Age(Yrs)	43.6±1.87	50.51±1.20	38.3±1.60	0.05
BMI(Kg/m2)	25.5±1.90	27.41±1.13	27.33±1.05	0.05
P.B.F	26.3±2.61	28.39±1.40	28.17±1.48	0.05
Age of menopause	40.51±2.0	49.72±1.59	21.87±0.31	0.05
Age at first pregnancy	22.52±2.04	24.20±1.66	48.56±1.55	0.05
Smoker	3	10	/	0.05
Miscariage/Abortion	5.98±0.65	6.71±1.89	3.52±0.37	0.05

* = $p < 0.05$ is significant

BMI = body mass index

PBF = percentage body fat

Table 2- presents a comparison the mean levels of trace elements in subject (cases) and controls. There were statistically increased significant of (Copper, Lead. and Aluminum) in postmenopausal patients as compared with controls and pre-menopausal patients with $p < 0.001$. While the serum levels of (Zinc, Selenium. Chromium and Magnesium) in pre-menopause patients was increased significantly different from control and postmenopausal patients.

However there was no significantly difference in the serum level of (Zinc. Selenium. Lead and Magnesium) between the groups of breast cancer patients.

Table 2: Mean (Mean ± SD and Range) of Serum Trace Elements in Breast Cancer Patients Compared with Healthy Controls

Parameters	Patients		Control	P-Value
	Premenopausal	Postmenopausal		
Copper (ugLdl)	26.0±3.60	32.2±5.21	22.08±2.05	0.033
Zinc (ugLdl)	8.78±3.50	7.25±2.31	17.38±0.5	0.05
Selenium	1.62±0.02	0.88±0.03	0.28±0.04	0.03
Lead	70.60±4.77	72.52±3.88	52.03±1.71	0.03
Chromium	6.22±2.84	4.03±1.71	3.25±0.11	0.005
Magnesium	0.71±0.25	0.65±0.05	1.85±0.13	0.005
Aluminum	44.01±3.51	69.33±4.62	10.3±2.60	0.032

Table 3- shows the urinary minerals that exhibited the levels of (Cu in pre. Zinc in post, Selenium (in pre), Lead (in pre and post). Chromium (in post) and Aluminum (in pre – post), a significant difference (increased) from controls.

Table 3: Mean (Mean ± SD and Range) of Urine Trace Elements in Breast Cancer Patients Compared with Healthy Controls

Parameters	Patients		Control	P-Value
	Premenopausal	Postmenopausal		
Copper (ugLdl)	21.38±4.24	19.16±6.45	18.21±3.71	0.05
Zinc (ugLdl)	55.39±5.90	101.3±1.21	60.40±2.51	0.05
Selenium	0.98±0.19	0.62±0.24	0.25±0.18	0.05
Lead	79.70±6.38	62.90±8.66	16.31±2.58	0.05
Chromium	4.60±1.75	8.48±2.44	5.39±1.29	0.05
Magnesium	2.88±0.91	1.89±0.93	5.79±1.76	0.05
Aluminum	52.77±4.08	56.03±5.85	7.94±1.95	0.05

Further more. Table 4 explication the model coefficients computed from multiple regression analysis for all elements studied in this study. These indicated that these elements in serum were significantly inversely correlated

observed in the outcome variable in urine.

Table 4: Regression Table between Tumors Parameter Index (Dependent Variable) and Trace Elements (Independent Variable) in Breast Cancer Patients

Trace elements	R	R2	B	P - value
Copper	0.781	0.482	-0.781	0.001
Zinc	0.528	0.357	-0.528	0.045
Selenium	0.29	0.036	- 0.29	0.001
Chromium	0.273	0.192	-0.273	0.001
Magnesium	0.281	0.06	- 0.281	0.001
Aluminum	0.655	0.548	-0.655	0.045
Lead	0.201	0.082	-0.201	0.05

DISCUSSIONS

Breast cancer disease is a common tumor in female's worldwide knowledge elements metabolism and their disturbances in tumor cell or tissue⁽¹³⁾. Might help to elucidate the relation of the elements to tumor and to decide whether these elements could be used as additional biochemical marker for the diagnosis and prognosis of tumor development.

It will be referred to that elements partake on biology processes in the organisms which induce the activation or inhibition of enzymatic reactions between elements and more metal protein tying positions, also adjustments in the corrosive for cell division membrane⁽¹⁴⁾.

There are few studies concerning the determination of elements level in blood and urine of breast cancer patients, most of them deal with Zn, Cu levels due to the role of these elements in the antioxidant defense system of the organism⁽¹⁵⁾. No examination is superior to the research center performing it. Correct explanatory methods must be utilized and each step in the collection and preparation of sample and the urine analysis. Gathering tubes uncommonly outline for designed.

This study demonstrated that anthropometric also demographic indices need aid comparable on breast cancer patients and controls. Therefore suggesting that lifestyles and diet would those same in the two assemblies about subjects.

Copper (Cu) is biologically essential trace element that is required for the synthesis of hemoglobin white blood cells maturation bone strength and as integral compound of certain for protein as ceruloplasmin⁽¹⁶⁾. Which are principally concerned with oxidation reactions? Angiogenesis the growth of a tumor blood supply. is essential for tumor growth. Molecular processes of angiogenesis require copper as essential cofactor (17). Also reliably large amounts about copper have been found in breast cancer tissue. In these study serum levels of copper were significantly increased in postmenopausal for breast cancer group when compared to controls. While no significant difference was observed between controls and per – menopause breast cancer groups. Our discussion on this thing in higher serum Cu levels in postmenopausal breast cancer it may be indicate metabolic effects concerning that depend on the tumor state. In addition the Cu in serum exists generally in the manifestation in ceruloplasmin more than 90%, since the serum Cu also ceruloplasmin levels would normally nearly associated⁽¹⁸⁾. According to some studies that benefit us increasing concentration of Cu and ceruloplasmin breast cancer patients and as well as the extent of their influence in Cu absorption, transport metabolism or excretion inflammation infection with many different type of cancer espial in breast cancer. Our study the results obtained was in agreement with some reported that patients with breast cancer in postmenopause had high serum Cu than other study groups⁽¹⁹⁾. Urine Cu presents a significant difference between pre and

postmenopause breast cancer patients and controls that showed in table (3).

Zinc

Is apart overt cell in the body and manifestations of over 300 enzymes that have functions of body hormones to cell growth ⁽²⁰⁾. The epidemiological information on the relation between Zn and growth of tumor are restricted. Furthermore, in vital works about Zn would its contribution into cellular division protective function against free radical and reactive oxygen. Therefore those cell membrane will be characteristically secured by metallo – enzymes against OHRadicals, this indicated that the Zn might restrain carcinogenesis ⁽²¹⁾. On other hand Zn is needed for growth and likewise as a component of the Zn finger protein it play controlled of cell division and oncogenic actuation there maybe likewise a portion proof to an opposite cooperation between Zn and breast cancer. Referring to the results we have obtained in relationship with Zn concentration as shown in the table (1).

In the pre and postmenopausal a significant decreased in serum Zn was observed among pre – postmenopanse as compared with control. This finding is agreement with some studies who found that Zn concentration was decreased in breast cancer patients. Through scientific references know that found interference of Zn with intestinal absorption of copper. for this reasoning any absorbed Cu could displace Zn due to copper that high affinity to metalbthionen as protein. This guide whom we found in our results through increased copper concentration and lack of concentration of zinc as support what we found in our study. With regard to the discussion of urine in all study groups the decreased excretion of Zn in breast cancer compared with control group table (2). Another mechanism suggested the possibility of excretion to extraction to Zn from the circulation by the tumor tissue to be used proliferation. The observed decreased Zn level in our study also agrees with Borrellaetal, ⁽²³⁾.

Selenium

Is an essential trace element and has been shown to be a natural component in the enzyme glutathione peroxidase and other proteins most selenium compounds are water soluble and can efficiently be taken up in the intestine as well as non soluble compounds can be taken up by the lungs. Several of scientific inquiry suggests that an increased risk of cancer occur as a result of statute concentration of Se in the diet. After such a review and we achieved results that showed a significant decreased was in serum Se concentration in pre and post menopause groups as compared with controls with $p < 0.03$. While when returning to the analysis between premenopausal and postmenopausal of breast cancer note the lack of statistically significant differences were too high.

As for the urine of study groups and in particular pathological have we found a significant slow increased compared with control group. In some studies an opposite companionship between serum Se levels and neoplastic has been observed in different malignancy types it need additionally been postulated that those methodology underling tumor improvement car lead to an uptake of Se by the malignant cells showing the human defense mechanism. The lower serum Se levels observed in our study could be attributed to either lower Se intake or to sequestration of this element by the tumor cells ⁽²⁴⁾. This finding is in agreement with the reports (Charalbpowls and Rejali) ⁽²⁵⁾.

Lead and Chromium

Lead furthermore chromium need aid non – essential toxic metals that have affinity forsulphohydryl active site of enzyme and proteins. These interactions are part of natural metal detoxification process. Which result in the metabolic

inactivation of Se in sufficiently high exposure levels Pb and Cr might additional time process a state associated on Se state associated on Se insufficiency thereby aborting the cancer – protecting effects.

Our study returned a significant increased in the serum and urine levels of Pb and Cr in the breast cancer patients.

The chronic exposure to cadmium and lead can lead to breast cancer especially in females and these metals are usually the result of smoking and pollution smoking from car exhaust or through food sources entering the body's digestive tract. After reviewing some of the studies on chromium affect the creation of breast cancer gene alternator (oncogene). Also can inhibit the effect of restraining tumor protein (P53) (Tumor suppressor protein). This protein contains zinc. Combined with a cysteine those compounds will be the ability of chromium on the thiol group ⁽²⁶⁾. After this survey the totally of things on the Pb and Cr note the significant increase in the levels of Pb and Cr in the breast cancer patients observed in our study could have contributed to the pathogenesis of breast cancer.

Magnesium

Concerning the magnesium that is essential for all biosynthetic processes glycolysis and energy dependent membrane transport ⁽²⁷⁾.

Magnesium level in serum of breast tumor patients as compared with healthy controls (table 2). significant decrease was observed in serum Mg level in pre and post menopause groups as compared with control. As seen on the urine concentration of Magnesium. The level of Mg is further decrease in post menopause group as compared with premenopause patient $p \leq 0.001$.

Magnesium deficiency can paradoxically increased the risk or protect against cancer yet we will find that just as severe dehydration or asphyxiation can because death. Magnesium deficiency can directly lead to cancer. Therefore we could see that Magnesium deficiency would lead to physiological decline in cells setting the stage for cancer ⁽²⁸⁾.

Aluminum

Aluminum is ubiquitous in the environment for the general population exposure to aluminum most likely occurs through the consumption of food (mainly processed food) water and medicinal such as antacids, buffered analgesics anti diarrheal agents. Or ant ulcerative medication. The intake of Aluminum from food and water is low a specially compared with that consumed by people medicinal and antiperspirants as cosmetics (29)

Based on the observation of a high incidence of breast cancer in the upper external guardant. Adjacent to the usual area of application of deodoratants and antiperspirants, some scientific teams have advanced the hypothesis of a possible link between antiperspirants and breast cancer (30). In this study there is significant difference in aluminum level between patients and control groups in pre – post menopause patients. the serum level of Al, significantly increased in patients group than in control ($p < 0.001$) table (2).

Aluminum is poorly absorbed following either or inhalation exposure and is essentially not absorbed dismally. Absorbed aluminum is excreted principally in the urine and to lesser in the bile. Therefore can be measured in the urine and to feces. There are insufficient data to relate aluminum exposure levels with blood or ruin levels (31). Based on what has been discussed above, we have analyzed the aluminum element in both the blood and urine and found in the following table (2). The level of aluminum (Al) in serum and urine sample were significantly higher in two patients groups (pre and

post menopause breast cancer) than control group, although its level was significantly lower in per menopause than post menopause ($P \leq 0.001$). Those purposes for the disproportionate deposition of aluminum could relate to physiological mechanisms components not yet understood it might additionally consistent with local absorption of aluminum from long-term antiperspirant use in that of the human body (muscle). Similar to our findings in study carried by Exley et al. was found higher levels of aluminum in serum breast cancer (32), conversely that we found another study from House et al. 2013. Who did not show any statically significantly differences in aluminum level in women with breast cancer. A study by Anane et al. found increased level of aluminum in the urine of mice exposed to amount of aluminum chloride containing antiperspirant cream applied daily to 4cm² shaved for 130 days (33). Through this conclusion we found that agree the results obtained in this study.

CONCLUSIONS

There is growing evidence environmental contaminants such as metals play a role in disease, such as cancer. Based on a relatively small number of studies this literature review has uncovered important deficiencies and gaps in the current literature that assesses the link of the incidence of breast cancer to metal in serum and urine from this patient groups.

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