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Assessment the impact of zinc and copper on GST, ALP in blood of cancer patients

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Abstract: Breast cancer is a highly heterogeneous disease globally. The absolute risk of breast cancer increases with age for women . Study the role of trace elements in the blood such as (copper and zinc) and biomarkers breast cancer diagnosis such as GST, ALP . had been measured in serum of 60 women with breast disease , divided to :. the first group (A) (30) women benign breast tumors , the second group (B) (30)women breast cancer. Evaluated of serum copper and zinc of breast cancer women was determined by atomic absorption spectrophotometry , and the level of (GST)(Glatathione – S- transfers) enzyme and ALP(Alkaline phosphates) enzyme compared with control group. The results showed a significant increase ($p<0.001$) in the level of enzyme (Glatathione – S- transfers) and the (Alkaline phosphates)enzyme in serum breast cancer group , while we observed a slight increase ($p<0.01$) in benign tumor group compared to the control group. However , there was a significant decrease in the level of the zinc element compared with the control group and a rise in the level of the copper element, which means copper /zinc ratio in patients was higher than in ontrolgroup.

Key words : Breast cancer , trace element , zinc, copper , GST,ALP enzyme.

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Introduction:

Breast cancer is a common disease worldwide.Recurrent risk factors for breast cancer have been document . Breast cancer risk increase with age and occurs at much higher levels in postmenopausal women (1). Among women breast cancer is the second most common type of cancer resulting in death, after lung.

Cancer cells divide and grow uncontrollably , forming and invade nearby parts of the body.The cancer may also spread to more distant parts of the body through the lymphatic system or blood stream. Breast cancer is a type of cancer originating from breast , most commonly from the inner ling of milk ducts or the lobules that supply the ducts with milk (2).

In a study conducted by the international Breast cancer , Alkaline phosphatase (ALP), was examined for their sensitivity in detecting breast cancer recurrence . ALP alone was abnormal in a high proportion of breast cancer patients with bone metastases and /or liver metastases. Alkaline phosphatase (ALP) comprises a group of enzymes that catalyses the hydrolysis of phosphate esters in an alkaline environment, generating an organic radical and inorganic phosphate. As with other enzymes , ALP has different iso enzymes . ALP is mainly derived from the liver and bones and in lesser a mounts from intestines , placentas , kidneys and leukocytes. Increase in

serum ALP levels have associated with a variety of hepatic and bone diseases, such as cholestasis and infiltrative liver disease. Serum ALP levels are elevated in patients with metastatic tumours of liver and bone, such as hepatic metastasis of colorectal cancer, bone and liver involvement in breast cancer (3). In patient with malignancies, therefore, an elevated serum ALP level may be an indicator of metastatic disease. However the association of ALP with other clinic opathological characteristics of cancers, such as the status of lymph node hydrogen peroxide and dioxide, hence breaking down the toxic reactive oxygen species (ROS) radical (4).

Glutathione –S- Transferase (GST) is a family of volatile proteins .Motivation and Cytoplasm of many tissues this enzyme plays a key role in reducing the toxic properties of compounds . This type of enzyme stimulates the interaction of the total factors and is guarantied by effective pharmacokinetics (5), Claysin or clotamate followed by Acetylation (Mercapturic Acid) and that the treatment of harmful substances causing oxidation caused an increase in the (G.S.T.) hepatic(6). Copper (Cu) and zinc (Zn) are heavy metals and are recurrently detected in the environment. Various studies have shown that metals are connected with adverse health, possessions, pointing to their importance as a public health (7). Copper is an important element broadly distributed in nature. Increased serum and tumor tissue levels of Cu are also observed in several cancers, although little is known about how the metal might promote disease progression at the molecular level. Epidemiological studies have instituted no distinct association between Cu exposure and cancer (8). Zinc (Zn) is a necessary metal Zinc toxicity is infrequent and occurs only at very elevated exposure levels . Oral Zn supplements do not appear to have significant effects on the incidence of cancer . Actually , Zn deficiency may be connected with increased risk of cancer in humans (9). Zinc on the other hand , present in more than 200 enzymes and transcription factors as a functional component. Therefore , Zinc affects major metabolic processes , as well as instruction of the cell cycle and cell division . The primary symptom of Zinc deficiency is an inhibition of cell growth and proliferation. Additionally , Zn is required for the optimum performance of the immune system (10). The aim of the present study was to Assement serum trace elements such as Cu and Zn on levels of alkaline phosphatase activity and the Glutathione –S-Transferase (GST) in Iraqi women with breastcancer.

Materials and Methods:

The present study was conducted on (90) breast cancer patient and healthy was participating in this study from the city of medicine Baghdad Hospital. They divided into:

1- Control group ,consist of (30) healthy .
2- Group (A) of. 30 benign contract in the Breast , include blood and tissue models.
3- Group (B) of. 30 breast cancer include blood and tissue models.

Procedure to prepare sample (Tissue sample):

Weight the fabric , was placed in the beaker added mixture (5ml of

sulfuric acid , 5 ml nitric acid and 3ml perchloric acid) , heated beaker then left beaker for 24 hours. Added distilled water, heated the beaker, filtered,complet the volume to 100ml with distilled water.

Blood sample:

The samples were collected from healthy and cancer patients was centrifuge, serum samples were separated and stored for biochemical profile analyzed of blood.

Analyzed according to standard kits (11) **A** . Alkaline Phosphatase (ALP).

Determination of Serum Copper and Zinc: B.

Serums Cu. Zn was determined by atomic absorption spectrophotometry according to (12,13) .

Activity of GST enzyme:

Blood samples were collected from the patients; in EDTA tubes. The collected samples were analyzed for biochemical profile of Red blood cell ,Glutathione-S-Transfers enzyme (GST), according to standard kits(14).

Statistical:

Comparison was done between breast cancer with or without metastasis , Cu , Zn and enzymes with control by using "t" test (2001) data were expressed as means ± SD. For the lowest significant between group rates and probability level ($p<0.05$).

Results and Discussion:

Breast cancer is one of the leading cause of death in women in the developing countries , a needs is felt for some simple biochemical investigations for the early detection of cancer and can be assayed in smaller laboratories located at remote areas . In view of this , certain biochemical investigations , which are though considered nonspecific , carried out in breast cancer patients to establish their diagnostic value in cancer with and without metastasis. The diagnostic Neoplastic is (15).

Alkaline phosphatase (ALP) enzyme effective level:

The mean serum levels of the alkaline phosphatase (ALP) activity studied are represented in (Table 1) and (Figure 1) showed the mean serum ALP activity of breast cancer subjects groups (A) and group (B) were significantly higher than those of control group , The increase activity of this enzyme seen in breast cancer and benign cancer subjects may be due to osteolytic bone metastases in breast cancer leading to increase osteoclastic activity and bone resception and the activity of ALP between breast cancer subjects for damages proteins that cause inhibit the enzymes that work together to cause damage by oxidation of cell contents such as nucleic acids, proteins and fats (16,17). The types of interactions are prevented or removed before they can damage the contents of the cell (18).

Table (1): level of alkaline phosphatase in breast cancer patient and control (mean ± SD).

groups	NO.	ALP mmol/L Mean ± S.D	T.test
C	30	46.1 ±16.48	----------
A	30	92.31±32.91	P<0.05
B	30	.37 28 ± 96.95	P<0.001

C= control groups.
A= Serum of women group have benign contract.
B= Serum women group breast cancer.

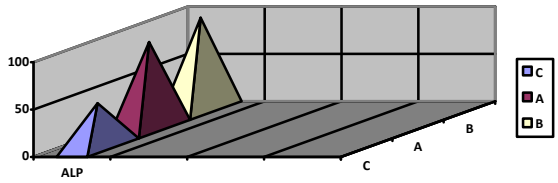

Figure (1): Serum ALP enzyme profile of cancer patient and control .

C= control groups.
A= Serum of women group have benign contract.
B= Serum women group breast cancer .

The Glutathione –S- Transferase specific activity in Red blood cell for cancer patients and control group.

The mean serum levels of the glutathione –S-transferase (GST) activity studied are represented in (Table 2) and (Figure 2). shows the mean serum GST activity of breast cancer subjects groups A and B were significantly higher than those of control group. There are many research and reports that refer to different types of cancers including esophageal cancer , intestinal juices , gastric cancer and its effect on the effectiveness of GST enzyme(19). Therefore, it is necessary to maintain the balance of the presence of GST enzyme and to preserve the contents or components of the enzyme GST enzyme glutathione –S-transferase plays a key role in reducing the toxic properties of foreign compounds entering the body such as some pharmaceutical compounds. This enzyme stimulates the collocation of the plutonium with foreign objects and therefore the products accumulate more solubility in water to dispose of the body (20) .Glutathione as a reluctant is very important in maintaining stability of erythrocyte membranes. Its sulfhydryl group reduces peroxides formed during O_2 transfer and thus it provides protection against free radical injury. Which affected the formation of the base material for the work of the enzyme GST or the transformation of non-cancer by stimulating different effectiveness the equal forms of GST in all cases of breast cancer (21).Regarding the role of minerals and heavy metals in the origination or van cement of breast cancer , emerges one potential conclude that the raised levels of trace elements and heavy metals could have led to the formation of free radicals or additional reactive oxygen spices that unfavorably manipulate DNA , thus causing breast tumor.

Table (2): level of Red blood cell The Glutathione –S- Transferase G.S.T. enzyme in cancer patients and control(mean± SD)

groups	NO.	GSTU/gm Hb Mean ± S.D	T.test
C	30	1.91 ±0.5	----------
A	30	2.51±1.7	P<0.05
B	30	5.11±2.6	P<0.001

C= control groups.
A= Serum of women group have benign contract .
B= Serum women group breast cancer.

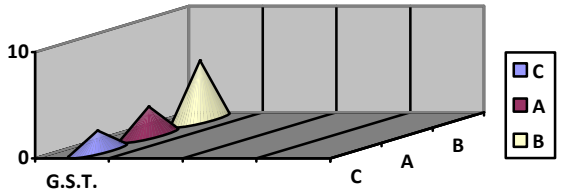

Figure (2): Red blood cell The Glutathione –S- Transferase G.S.T. enzyme profile of cancer patients and control group .

C= control groups.
A= Serum of women group have benign contract.
B= Serum women group breast cancer.

The trace element role including:

Serum copper (Cu) concentration levels:

The results of this study are present in (Table 3) and (Figure 3) showed the mean serum level of Cu metal activity was increased in patients viewing a highly significant distinction in breast cancer patients as compared to the control . Also the level of Cu metal activity was increased in patients viewing a significant distinction , in benign breast cancer patients as compared to the control (22). The elevation in serum Cu concentration may be due to the destruction and necrosis of involved tissues , leading to the release of Cu into circulation. The segregation determination in methods may be the cause for these contradictory results. It has been postulated that blood serum concentration of Cu was significantly higher in patients with breast cancer than in controls(23). The present results are practically parallel to other findings reported, who was found a significant increase in serum Cu levels in breast cancer patients compared with the control Also, (24) reported that serum Cu levels were increased significantly in cancer patients compared with the control group.

Table (3): level of (Cu) metal parameter between breast cancer patients and control (mean ± SD).

groups	NO.	µg/dL Cu Mean± SD.	T.test
C	30	78.0 ±0.24	-------
A	30	136.3±11.8	P<0.05
B	30	168.8.1± 12.40	P <0.001

C= control groups.
A= Serum of women group have benign contract.
B= Serum women group breast cancer.

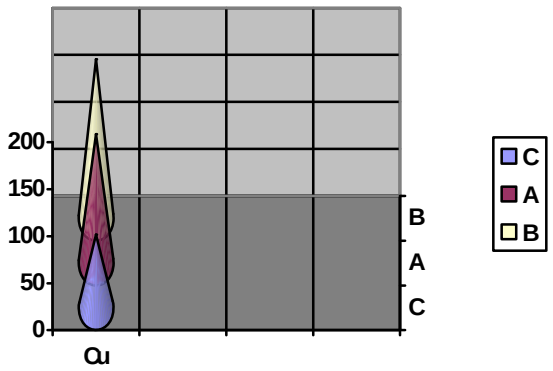

Figure (3): serum (Cu) metal profile between the patients and the control group .

C= control groups.
A= Serum of women group have benign contract.
B= Serum women group breast cancer.

Serum Zinc (Zn) concentration levels:

As demonstrated in (Table 4) the serum level of zinc was significantly lower in women with breast cancer and in benign breast cancer patients as compared to the control . In breast cancer women , serum level of Zn showed no significant differences in Zinc (Zn) in (Figure4). (25). In this study , there was no significant distinction in Zn among breast cancer patient as compared with control group, serum concentration of Zn did not differ significantly in breast and benign cancer and control .The current study found that serum Zn levels in breast cancer patients did not significantly be different with those in the controls. Nevertheless, the means concentrations of Zn were lower in breast cancer patients than in the controls. The means of serum Zn concentrations between the cases and the controls did not be different significantly (26). Some studies , however, found conflicting results . For instance, a study conducted by (27) , found that Zn values were significantly lower in patients with breast cancer than in the controls. A variation in the method of Zinc recognition may be the cause for the differences between the result of the current study and earlier studies.

Table (4) : level of (Zn) metal parameter between breast cancer patients and control (mean ± SD).

groups	NO.	µg/dL Zn Mean± SD.	T.test
C	30	84.68±9.72	--------
A	30	69.3 ±8.2	P<0.05
B	30	60.9±6.8	P<0.001

C= control groups.
A= Serum of women group have benign contract.
B= Serum women group breast cancer.

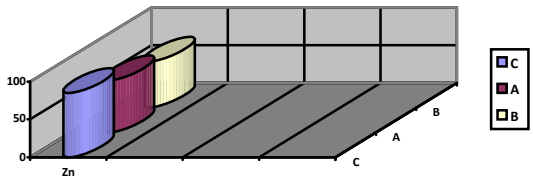

Figure (4): serum (Zn) metalprofile between the patients and the control group .

C= control groups.
A= Serum of women group have benign contract.
B= Serum women group breast cancer.

Breast tissue Copper (Cu) concentration level:

The mean tissue levels of the Copper (Cu) activity studied are represented in (Table 5) and (Figure 5). It was observed a significant decrease in the level of Cu when compared to the control. On the other hand significant increase in copper concentration is observed in tissue sample compared to their benign cancerous group (28). A study reported that the Cu induces apoptosis by P53 dependent and independent pathways.

Table (5): Concentration (µg/g) of studied metal (Cu) in tissue sample of cancerous and benign cancer group .

Group	NO.	Cu µg/g Mean± SD.	T.test
A	30	8.51±4.35	-------
B	30	16.1±6.54	P<0.05

A= Serum and tissue of women group have benign contract.
B= Serum and tissue of women group breast cancer.

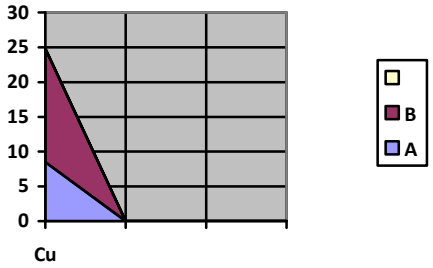

Figure (5): Tissue sample(Cu) metal profile in tissue sample of cancerous and benign cancer group.

A= Serum and tissue of women group have benign contract.
B= Serum and tissue of women group breast cancer.

Breast tissue Zinc (Zn) concentration level:

The results of this study are present in (Table 6) and (Figure 6) .show the mean tissue level of Zn metal activity was significant increase in Zn concentrations were detected in tissue samples of cancerous patients compared to their corresponding non-cancerous one in patients viewing a lower significant distinction, in benign breast cancer patients as compared to the control, while zinc has no significant difference between studied groups(29) .Our study is in contrast to a study which raised the possibility that relatively high level of Zn in benign breast tissue may be associated with a modest increase in risk of subsequent breast cancer In postmenopausal women, the mean values Zn was significantly decreased ($p<0.001$) in patients as compared to healthy subject. Zn is required for growth and , as a component of the Zn finger proteins; it plays a pivotal role in controlling of cell division and on cogenic activation . There is also some evidence for an inverse association between Zn and breast cancer. The Total mean serum protein concentration in patients.

Table (6): Concentration (µg/g) of studied metal (Zn) in tissue sample of cancerous and benign cancer group .

Group	NO.	Zn µg/g Mean± SD.	T.test
A	30	54.6±6.67	----------
B	30	66.9±7.72	P<0.05

A= Serum and tissue of women group have benign contract.
B= Serum and tissue of women group breast cancer.

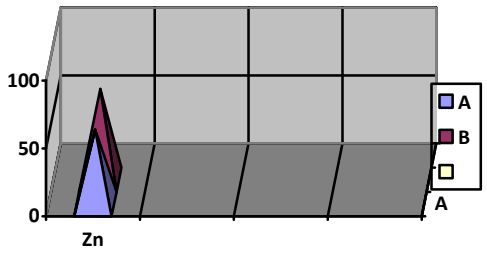

Figure (6) : Tissue sample(Zn) metal profile in tissue sample of cancerous and benign cancer group.

A= Serum and tissue of women group have benign contract.
B= Serum and tissue of women group breast cancer.

Conclusion:

The present study focused on evaluation of serum biochemical of profile of breast cancer patients , that women with breast cancer have increased ALP and glutathione –S-transferase (GST) enzymes activity than in apparently healthy women .Serum trace element (Cu and Zn) are significantly difference in breast cancer patients compared to controls , The alteration of these biochemical parameters is an indication that their measurement may be useful tools in monitoring treatment and disease progression in breast cancer subjects.

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