

(RESEARCH ARTICLE)



Measuring of serum taurine and beta endorphin in women as a pre-early marker for diagnosis of cancer breast

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Abstract

Objective: Uses of serum level of taurine and beta endorphin as early marker for diagnostic of breast cancer.

Method: We selected our patients from Bahia hospital and cancer institute of Cairo university mainly suffered from positive family history of cancer breast ,acute and chronic mastitis and all women presented with benign mass (fibroma/lipoma) and different stages of cancer breast. We measure serum beta endorphin and taurine, beside complete blood picture, liver function ,kidney function and glucose in blood in all of our patients.

Result: The data showed highly significant change in liver &kidney function between frank control and stages of cancer($p>0.05$) that are clearly at latest stages after doses of chemotherapy, but there are slightly change between inflammation and benign.

After we finished all result, we found that there were slightly change of beta-endorphin and taurine at inflammation and benign but further change at stage 1,2 that are continuously change to latest stage

Conclusion: Decreasing of serum taurine level &increasing of serum beta endorphin level considered as a highly sensitive per early marker for breast cancer.

Keywords: Taurine; Beta Endorphin; Breast cancer; Cancer; Biochemical.

1. Introduction

Till now the diagnosis of cancer breast in our country is mainly detected by imaging techniques (mammography, specific tumor marker and biopsy (histopathological examination) and measuring of tumor marker for breast.

From the recent paper we found that breast cancer is the most common type of cancer that lead to death at shorter time due to their stages are rapidly progressive

Because we can't sign the breast cancer at early stage and female need to take chemotherapy. we start our study to can avoid this. So, we use beta-endorphin and taurine as early diagnosis for breast cancer

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Beta endorphin is opioid hormone that plays apian killer and related to stress so at breast cancer all women being under stress and increasing within stages of cancer

Taurine is an antioxidant and play role through down regulation of angiogenesis and enhancement of tumor cell apoptosis, Taurine is called immune marker due to a relation of it with immune system ,But our aim of this study to selected highly susceptible women for cancer breast to avoid the breast cancer

Breast cancer is the most diagnosed cancer among women in the United States and the second leading cause of cancer-related deaths; therefore, prevention of the disease would significantly improve public health [1,6]. An estimated 268,600 new cases of invasive breast cancer occur among women each year, and 41,769 women will die from the disease [1,2,6]. The United States Preventive Services Task Force recommends that clinicians offer chemoprevention as a primary prevention strategy to women at high risk of breast cancer and low risk of adverse effects from these drugs [3,6]. High-risk criteria for breast cancer is defined as a 5-year invasive breast cancer risk of $\geq 1.67\%$ or lifetime risk score of $\geq 20\%$ according to the Gail risk model, which accounts for age, race and ethnicity, benign breast disease, first-degree family history of breast cancer, and reproductive factors [4,5,6]. Carcinoma of the breast is the most prevalent cancer among Egyptian women, it constitutes 29% of Cairo National Cancer Institute cases, and it is usually diagnosed at an advanced stage [7, 8,9]. There are several hypotheses regarding breast cancer etiology, including carcinogenesis by steroid hormones, chemical carcinogens and oxidative stress. Epidemiological studies suggest that a diet that is rich in antioxidants may help to prevent the development of breast carcinoma [10,11,9]. The lifetime risk for women of being diagnosed with breast cancer is currently between 1 in 7 and 1 in 8, the risk is even higher for women with certain risk factors, such as a strong family history or known BRCA1 or BRCA2 mutations [12,9]

. In Egypt, breast cancer is the most common malignancy in women, accounting for 38.8% of cancers in this population, with the estimated number of breast cancer cases nearly 22,700 in 2020 and forecasted to be approximately 46,000 in 2050

The most common malignancy that leads to death in women worldwide is the breast cancer [13,14]

- 1) Taurine (Tau), the most abundant free amino acid in humans that has numerous potential health benefits through its antioxidant and anti-inflammatory properties
- 2) Taurine (Taurine; 2-aminoethanesulfonic acid) an essential non-protein containing sulfur amino acid, involved in a wide range of physiological processes, among the most abundant organic molecules in human body [15,25]. Present in high concentrations in the liver, also in enormous amounts in the brain, retina, heart and platelets [16,25]. The best food sources are meat and fish [17,18,25]. Taurine involved in cell volume regulation, enhances stability of membranes and directly stabilizes membrane proteins and modulates inflammation [19,20,25]. Moreover, studies showed that taurine involved in apoptosis regulation [21,22,25]. Lately, it has been used as an antipyretic and anti-inflammatory agent, to treat liver and Gallbladder disease, Cardiovascular disease, Diabetes and Cataract [23,24,25].
- 3) The antioxidant Taurine is found to display antineoplastic effects through down regulation of angiogenesis and enhancement of tumor cell apoptosis. It has been found that progressive inhibition of apoptosis and induction of angiogenesis may contribute to tumor initiation, growth and metastasis in the pathogenesis of breast cancer (9). Taurine (2-aminoethanesulfonic acid) is a free amino acid found ubiquitously in millimolar concentrations in all mammalian tissues. Are mainly synthesis in the liver tissue. Taurine exerts a variety of biological actions, including antioxidation, modulation of ion movement, osmoregulation, modulation of neurotransmitters, and conjugation of bile acids, which may maintain physiological homeostasis. Recently, data is accumulating that show the effectiveness of taurine against diabetes mellitus, insulin resistance and its complications, including retinopathy, nephropathy, neuropathy, atherosclerosis and cardiomyopathy, independent of hypoglycemic effect in several animal models. The useful effects appear due to the multiple actions of taurine on cellular functions. This review summarizes the beneficial effects of taurine supplementation on diabetes mellitus and the molecular mechanisms underlying its effectiveness. (26)

Endorphins are endogenous morphine, neuropeptides, produced in the pituitary gland in response to stress and pain. There are three types of endorphins: beta-endorphins, enkephalins, and dynorphins binds to mu, kappa, and delta receptors situated on the nervous system and immune cells. Cancer is a major threat to mankind killing millions of people around the world annually. There have been recent advancements in the field of surgery, chemotherapy, and radiotherapy, still the prognosis of cancer patients has not improved much with increasing morbidity. We can't kill

cancer cells without killing normal cells. Cancer cells and normal cells work alike. The aim of the review was to determine the anticancer activities of beta-endorphins. (11)

Chronic psychological stress is one of the predisposing factor for cancer along with depression ,frustration ,fear, hatred results in release of CRH (corticotrophin releasing hormone) from hypothalamus activates hypothalamic-pituitary adrenal axis through sympathetic nervous system activity of ANS release neuro hormones such as cortisol ,ACTH ,and noradrenaline

We have a relationship between the nervous system and cancer [27,28]and this relationship demonstrates the connection between the psychological characteristics of humans due to stressful situations in life and specially at cancer evolution.

- The recent study shows that neurobehavioral stress can promote the progressive at cancer breast [28,29,30,31,32].
- Psychological factors can alter immune and endocrine function, stress can affect the growth of tumors.
- Similarly, beta endorphins release and increased at chemotherapy to recover the feeling of Bain [28,33]
- Breast cancer is one of the best known to influence the different stressors involved in coping with disease [28,34]

2. Material and method

2.1. Patients

THREE -HUNDRED EGYPTIAN PATIENT (MALE AND FEMALE) who attended national cancer institute ,Cairo university; most of them were referred from private clinic and the other patient are very simple so all of them come to the institute with chronic diagnosis so after doing biochemical analysis and ultra sound will complete with tumor marker test and mammogram that indicated that most of patient that enter cancer institute are lately stage from cancer and that so sadly

- Because this reason we take 40 sample only from cancer institute and take the other from Bahia hospital
- Bahia hospital are hospital for early check up to female, so we collected 40 sample from it after ethics approval
- Adding all above we choose 20 sample from normal women that will be a control but divided to 2 group according family history
- We will discuss now the group that we will study on our research
- The prospective study involves fifth group of a total 100 female candidates
- all groups consisted of 100 women. aged 38-58 years old
- We will take all groups after approval ethics that are include
- Frank control with negative history (10)
- Frank control with positive history (10)
- Inflammation group from Bahia hospital (10)
- Benign group from Bahia hospital (10)
- Malignant stage 1 from Bahia (20)
- Malignant stage 2 from cancer institute (20)
- Malignant stage 3 from cancer institute (20)
- this work approved by ethical committee of national cancer institute and Bahia hospital
- 2-2 sample collection and tests.

The frank control that enrolled at our study we take a blood sample from them to measure biochemical analysis ,CBC ,ALT, AST, albumin ,total bilirubin ,creatinine ,blood urea,sodium,potassium,CEA,CA19.9 and finally measured (taurine beta endorphin),The other sample we measure also biochemical analysis, tumor marker, mammograms, CBC, kidney function and liver function before chemotherapy and before its addition to (taurine beta-endorphin)

3. Sampling assay

3.1. Taurine assay

Venous blood samples were extracted from all patients for estimation of serum taurine levels after fasting for at least 12 hours. Otherwise, measuring of serum AST (Aspartate Transaminase), ALT (Alanine Transaminase), Blood urea, Creatinine, Albumin, Bilirubin, Sodium, Potassium, CEA and CA19.9 was not needed during the fasting period.

Serum taurine was determined by High Performance Liquid Chromatography (HPLC) according to the pre-column extraction and derivatization methodology of McMahon et al. In the present at work, we use the Shimadzu HPLC model LC-10AT. Serum ALT, AST, Albumin, Bilirubin, Creatinine, Urea, Sodium, Potassium. Were performed using Cobas C111 (Roch), CEA and CA 19.9. determination kits were purchased Cobas e411 (Roch).

3.2. Beta-endorphin assay

Venous blood sample was obtained after overnight fast in tubes without anticoagulant. The samples could clot and centrifuged at 3000 for 10 min, at 4 c to obtain the serum. Serum samples were collected, rapidly frozen in liquid nitrogen and kept on -80 c until usage for assays. Sample was measured by a human Beta-endorphin ELISA kit (CUSABIO) According to manufacturer instructions. The sensitivity of detection is 15.6 pg./ml intra-assay coefficient of variation is <8% Inter-assay coefficient of variation is <10%

3.3. Statistical analysis

The data will be analyzed using Microsoft Excel 2010 and statistical package for social science (SPSS version 24.0) for windows (SPSS IBM, Chicago, IL). Continuous normally distributed variables were represented as mean \pm SD with 95% confidence interval, while nonmoral variables were summarized as median with 25 and 75 percentile and using the frequencies and percentage for categorical variables; a p value <0.05 will be considered statistically significant. To compare the means of normally distributed variables between groups, the student's t test was performed, and Mann-Whitney test will be used in non-normal variables and χ^2 test or Fisher's exact test will be used to determine the distribution of categorical variables between groups All comparisons with P-values below 0.05 were considered significant.

4. Result

This investigation included one hundred patient that were measured kidney function test and liver function test and CBC and taurine and beta endorphin

- We will discuss every ratio at separate table to can demonstrate our result
- We measured all function test before and after chemotherapy to discuss the effect of therapy

In table 1, We make CBC at all patient

Tables 1 Illustrated that for HB there is a highly significant change ($p < 0.01$) between all groups and control groups but no significant different ($p > 0.05$) between inflammatory group and benign group preoperatively while postoperatively no significant changes ($p > 0.05$).

Study cases	Age	RBC	WBC	LYMPHOCYTE	PLATELET	HB before	HB after
Normal range	-----	4.3-5.7	4.5-11	18-44	150-440	11.5-15.5
Control	26-37	4.5-5.6	5.1-7.4	20-33	238-346	14.3 \pm 1.17
inflammation	39-67	4.1-5.0	7.5-12.8	22-35	350-583	11.5 \pm 0.7
Benign	48-57	4.0-4.8	3.5-7.2	23-39	127-285	10.2 \pm 0.5
Malignant 1	53-79	3.9-5.3	5.2-8.3	36-40	390-460	9.8 \pm 0.7	9.66 \pm 0.7
Malignant 2	37-65	3.9-4.5	6.3-10.2	38-45	310-530	8.6 \pm 0.5	8.2 \pm 0.5
Malignant 3	22-55	3.2-4.0	7.5-13.6	40-49	420-620	7.5 \pm 0.9	6.9 \pm 1.0

Table 1 Hb is represented as mean \pm SD, while RBCs, PLT, WBC, LYMPHO, are represented as median and interquartile range (25%-75%). Groups 3 that are benign initially are significantly different from the control group (1,2). Groups bearing 4 that are inflammation initial are significantly different from the benign group. Groups bearing 5 that are malignant 1 initially are significantly different from the benign tumor group. Group bearing 6 that are malignant 2 initially significantly differ from malignant 1. Group bearing 7 that are malignant 3 initial are different from malignant 2.

Table 2 the kidney functions in a diverse group of patients, show that urea and creatinine values recorded highly significant changes in all groups of patients related to, frank control group ($p < 0.01$),

The kidney functions in different groups of patients. Data are expressed by difference between low and high level of each group, illustrate the kidney function test before and after chemotherapy

Table 2 CREAT 1, BUN1, Uric acid 1(before chemotherapy) and CREAT2, BUN, Uric acid (after chemotherapy).

Study cases	Age	CREAT1	CREAT2	BUN 1	BUN 2	Uric Acid1	URIC ACID2	Sodium	Potassium
Normal range	----	0.6-1.6	10-12.5	2.4-6.1	135-150	3.5-5.6
Control	26-37	0.6-1.1	7-11.5	2.7-5.9	132-143	3.5-4.5
Inflammation	37-67	1.1-1.26	13-17.6	2.3-4.5	133-149	4.0-4.8
Benin	39-83	0.9-1.3	18-22	4.2-5.6	133-142	3.1-4.7
Malignant 1	39-73	0.9-1.4	0.8-1.32	25-30.6	10-12.5	6-7.1	10-12.5	130-135	3.2-4.6
Malignant 2	37-63	1.3-1.46	1.2-1.42	30.5-36.7	25-30.6	7.2-8.2	25-30.6	129-130	3.0-3.9
Malignant 3	22-55	1.42-1.6	1.2-1.5	39.5-45.2	30.5-36.7	8.2-10.9	30.5-36.7	122-131	2.9-3.2

Table 3 the liver function in our groups, illustrate the LIVER function test before and after chemotherapy

Table 3 AST 1, ALT 1, T. BILL 1(before chemotherapy) and AST 2, ALT 2, T. BILL 2 (after chemotherapy).

Study cases	Age	T. Bill	T. BILL2	AST	AST 2	ALT	ALT 2	AST/ALT Ratio
Normal range	----	(0-1.2 mg/dl)	(0-40 U/I)	(0-33 U/I)	
Control	26-37	0.3-0.5	26.3-34.3	25.6-32.4	0.04117
Inflammation	37-67	0.4-0.5	17-25.8	18.8-24.6	0.07
Benin	39-83	0.4-0.5	18-26.2	18.5-24.3	0.02804
Malignant 1	39-73	0.4-0.6	0.4-1.5	13.4-18.2	10-22	12.1-17.1	13-36	0.08219
Malignant 2	37-63	0.5-0.6	0.6-1.7	23.3-51	12-55	27.7-43.9	25-74	0.03626
Malignant 3	22-55	0.6-0.7	1.3-2.6	43.7-67	24-105	32.4-67.2	12-77	0.01103

From all above data that are collected in the 6 tables we concluded that the chemotherapy has side effect on the internal body so if we can find a solution it early predicts of cancer, we can avoid this effect

that are we were tray to doing it through out study do we will be discussed now the data we are founded to added at the last researcher about the same point

when we are measuring the HB, liver and kidney function before and after chemotherapy, we found the chemotherapy have negative effect on them that leads to chronic anemia, liver fibrosis, kidney failure and more than them so made another data to can suffering this effect through measuring beta endorphins and taurine at all group to can improve our conclusion they will illustrate at the end

now we will discuss the data of beta endorphin and taurine

Table 4 Data of beta endorphin and taurine

Group	Age	Number	beta-endorphin	taurine
Control	26-37	20	1.4± 0.07	116.1± 5.8
Inflammation	37-67	10	1.7± 0.17	77.5± 7.7
Benign	39-83	10	2.1± 0.21	49.1± 4.9
malignant 1	39-73	20	3.2± 0.16	37.4± 1.8
malignant 2	37-63	20	4.6± 0.23	18.7± 0.9
malignant 3	22-55	20	9.4± 0.47	6.6± 0.3

this table discusses the serum level of beta-endorphin and taurine at all group with mean ± SE related to the ages of cases and number of each group

At table 4

we found that serum level of beta endorphin starts with normal range at group 1(control) and start to slightly increase gradually to the final group 6(malignant 3) Thar are clear at diagram B

beta-endorphin is slightly change from group 1 to group 2, and gradually to group 3 but it duplicated at group 4 and triple duplication or more than triple duplication of its level at group 6,

Otherwise we found that the level of serum taurine start normal range and decrease gradually to latest stage that shown at diagram A

The level of serum taurine is affected with immune state and we found that it highly sensitive due to the stage of cancer It rapidly decrease from group 4,5&6

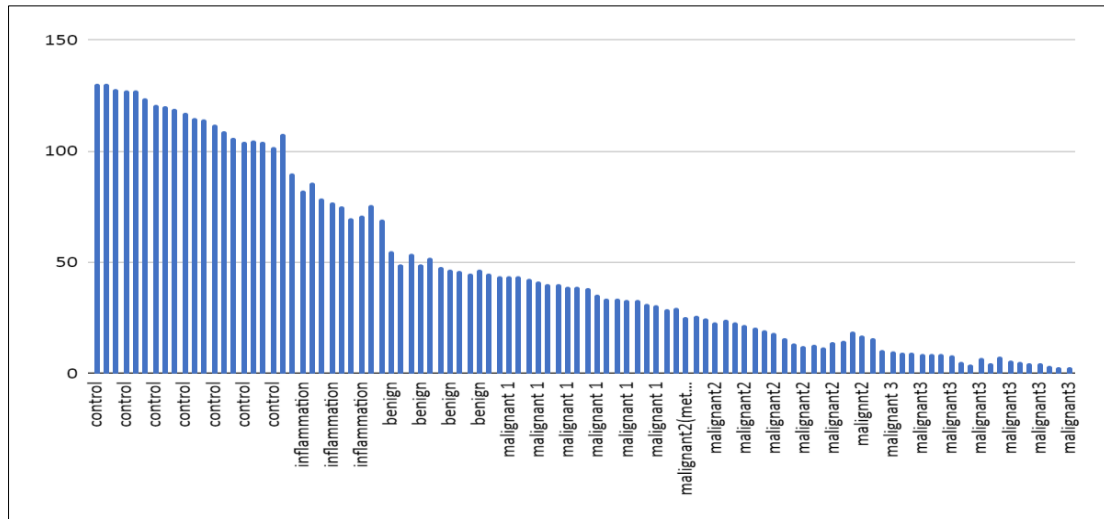


Figure 1 Histogram between taurine at all group we are studying it

X -axes are cases we are studying, y-axes are the level of taurine

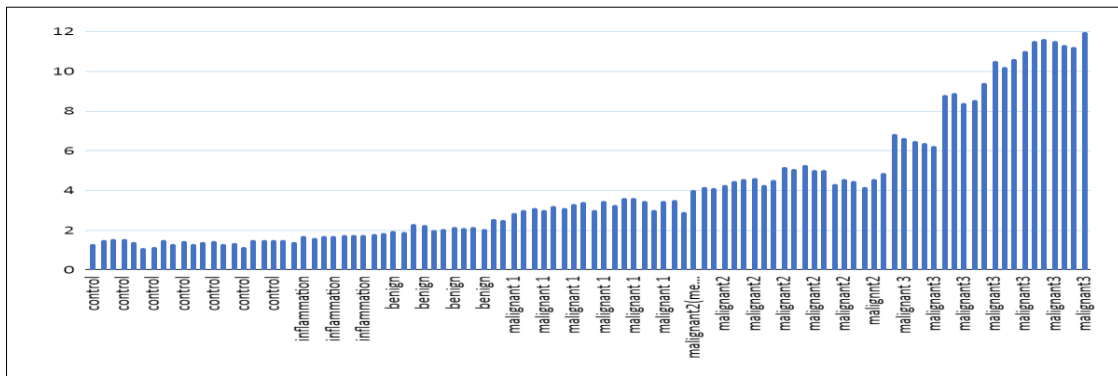


Figure 2 Relation between beta-endorphin and all group we are studying it

x-axes are the cases we are studying; y-axes are the level of beta-endorphin.

5. Discussion

Cancer is the major cause of death world, so we made ore study to can prevent the latest stage of it.

our study were ONE hundred patient selected to improvement uses of taurine and beta endorphin as markers for early diagnosis to breast cancer so we start with normal chemical analysis for blood sample that include (BLOOD GROUP, KIDNEY FUNCTION ,LIVER FUNCTION,SPECIFIC TUMOR MARKER,MAMMOGRAM)after we finish this analysis we found correlation between uses of chemotherapy and efficiently worked of body system

so, we measure taurine and beta endorphins to avoid this.

Our study is women that have a variation between frank control, control with positive history, inflammation, Benin, malignant 1(localized), malignant 2(transfer to nearest tissue) and finally malignant 3(transfer to faraway place).

So, we can discuss all data at all group to be more accurate at our conclusion

- The age of frank control was (26-37).
- The age of inflammation was (39-67).
- The age of Benin was (48-57).
- The age of malignant 1(53-79).

- The age of malignant 2 was (37-65).
- The age of malignant 3 was (22-55). Biochemical tests were also performed before and after chemotherapy treatment.
- We will now discuss all the results we found.

Biochemistry profiles of blood measures the chemical substances released from body tissues or are produced during the breakdown (metabolism) of certain substances. The analysis of blood chemistry provides important information about the function of the kidneys, liver and other organs. In the present study, the liver functioning (LFT) and kidney functioning (KFT) were assessed to check the level of different components. (13,14)

The level of creatinine in serum is considered more sensitive kidney function test than BUN. As kidney impairs the only cause of elevated creatinine. In this study, no statistically significant association was found in creatinine during different courses of chemotherapy in contrast to other studies. Observed the increased value of creatinine level ranges between 1.0 and 2.0mg/dl. (13,14)

5.1. At our study

Creatinine does not have any change between frank control, inflammation and benign compare to the normal range, but it slightly changes at carcinoma stages (malignant1,2,3) and still within normal range

So, Creatinine does not consider any sign for inflammation or breast cancer because any change on it if founded still in the normal range.

the measurement of Bun of blood urea is more accurate parameter for comparison at all group

because it's not changes at control, BUN is started to slightly change at group 2 (inflammation stage) & gradually increases at group 3 (benign stage) And suddenly duplicated at group 4,5 (malignant1,2), & continuously increased to group 6 (malignant 3)

So, we can conclude that BUN is affected with inflammation and carcinoma, especially the latest stage. From the collected data we found that Uric Acid is gradually changes at group 1,2,3 (control, inflammation, benign) but still at the normal range And start increasing from group 4 (malignant1) and continuously at group 5 (malignant 2) until travel to highly increasing at the latest stage group 6 (malignant 3) that are the latest stage.

There are no obvious changes, it may be decreased & return to increase at all different group but still at the normal range.

As the mean values of ALT and AST had no significant correlation with age. But data showed significant changes in kidney functions in all diabetic nephropathy stages compared to frank control group ($p < 0.05$). That showed impaired in renal function as increment of blood urea in most groups of patients. It was reported a high frequency of dyslipidemia in patients with diabetic nephropathy than in those without diabetic nephropathy [26]

But we found that the liver function at all group due to measurement the level of alanine aminotransferase (ALT), aspartate aminotransferase (AST), AST/ALT Ratio and Total Bilirubin (T.B) .

The level of ALT, AST, T.B, AST/ALT Ratio were analyzed for proper function of liver that are we will discuss. There are no significant change of total bilirubin at all group we detected because their all the inside normal range, we found that no change in the level of both at group 1,2,3 (control, inflammation, benign) there are mode ratable in the normal range of them.

Although AST & ALT are decreasing at group 4 (malignant 1), there are duplicated at group 5,6 (malignant 2,3), So, AST & ALT levels highly increased in patient with chronic diseases especially carcinoma. There are no significant change for ALT/AST ratio at group 1,2,3 (control, inflammation, benign).

The ratio of AST/ALT was generally decreased at chronic inflammation especially group 5,6 (malignant 2,3), So, AST/ALT/ Ratio not only increases at baseline but also predict the future development of breast cancer.

AST/ALT Ratio is most accurate parameter at liver function for diagnosis the stages of cancer breast.

The data of table 6 discusses the liver function after chemotherapy by measuring the level of AST1, ALT1, T.B 1 (before chemotherapy) and AST2, ALT2, T.B 2 (after chemotherapy).

So, after chemotherapy we found that (AST2, ALT2, T.B 2) are increases at group 4 (malignant 1) but still within the normal range at group 5,6 (malignant 2,3), there are highly increasing than (AST1, ALT1, T.B 1) and also more than normal range.

beta endorphins produce analgesia by inhibiting substance p, a neurotransmitter of pain in the peripheral nervous system through presynaptic μ (mu)-receptor binding. Beta endorphins produce euphoria, rewarding and analgesic effect by inhibiting GABA neurotransmitter and stimulating dopamine release after binding to μ receptors in the central nervous system () from all above data that are collected in the 6 tables we concluded that the chemotherapy has side effect on the internal body so if we can find a solution it early predicts of cancer, we can avoid this effect that are we were tray to doing it through out study do we will be discussed now that data we are founded to added at the last researcher about the same point when we are measuring the HB, liver and kidney function before and after chemotherapy we found that chemotherapy have negative effect on them that leads to chronic anemia, liver fibrosis, kidney failure and more than them.

so made another data to can suffering this effect through measuring beta endorphins and taurine at all group to can improve our conclusion they will illustrate at the end

6. Conclusion

By using our study, we observed a relation between serum level of taurine and beta-endorphin with the different group that are we discusses on table 7 and showing obviously at Diagram a & b

that result indicates that beta endorphin is highly recommended to study development of breast cancer and with us is highly indicated it to can pre diagnosis of cancer

beta endorphin is already responsible for Baine relieve so at carcinoma development it lead to increase of secretion of beta-endorphin

otherwise at the level of serum taurine, when we are measuring it at different stages of our study, we found it start at normal range at group 1 (control) and start to gradually be decreasing to the final stage group 6 (malignant 3)

the serum level of taurine is slightly changing from group 1,2,3

but it suddenly higher decreasing from group 4 and continuously decreasing rapidly to the last group 6 (malignant 3)

that indicated that taurine is highly sensitive that affected with acute, chronic inflammation, so we can use it to flow the development of disease.

so, the relation between them is irreversible.

Decreasing of serum taurine level and increasing of serum beta-endorphin level considered as a highly sensitive pre early marker for cancer breast.

confirming this conclusion, the early decreasing of serum taurine may encourage us for measuring its level in all women specially after age 40 every 6 months & in patient with positive family history of cancer breast, recurrent inflammatory mastitis (attack of mass) or benign (fibroma or lipoma) so, that every woman above 40% must measuring taurine and make mammography addition to beta endorphin,

other attractive point in this work as the paralysis between increasing in serum beta-endorphin in all examined group to decreasing in serum taurine.

at our research we investigated to uses it as early diagnosis for cancer breast due to we study the different level at all group .so, we success to improvement we can uses level of serum taurine and beta endorphin as early diagnosis for breast cancer

lastly, the marked decreasing in serum taurine & increase in serum beta-endorphin in any women suffering from any complication in the breast must be given special dose of taurine as a highly protective immune substance to guard against any complication of breast.

Compliance with ethical standards

Acknowledgment

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Statement of ethical approval

Bahia -Research Ethics committee, Cairo -Egypt. Bahia center for early detection and treatment of breast cancer national research Centre, Bahia RB protocol nuclear, 202210240039, 24/10/2022

Statement of informed consent

Informed consent was obtained from all individual participants included in the study.

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