

ORIGINAL ARTICLE

Assessment of the Activity of Lactate Dehydrogenase, Gamma-glutamyl Transpeptidase and Alkaline Phosphatase in Breast Cancer Patients with and without Lymph Node Metastasis

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Abstract

Background: Breast cancer has the highest cancer incidence in women and is one of the leading causes of mortality globally. Lactate dehydrogenase (LDH) and other enzymes are vital metabolic enzymes that are associated with cancer development, invasion, and metastasis.

Aim: The present study aimed to evaluate the LDH, Gamma-glutamyl transpeptidase (GGT) and Alkaline phosphatase (ALP) levels in the circulation of patients of breast cancer in different stages and tried to correlate levels with the disease stage and duration of breast cancer with and without lymph node involvement.

Methods: LDH, GGT, and ALP were analyzed by means of the spectrophotometer, Biosystem device and reagents, serum samples were used for measurement.

Results: GGT and LDH had a significant difference with P.value 0.022 and 0.001 respectively between stages II and III, while ALP didn't show a significant difference.

Conclusion: This data concluded that enzyme markers like serum LDH and GGT could be sensitive, specific and cost effective biomarkers for diagnosing carcinoma of the breast and for monitoring its progression.

Keyword: LDH, GGT, ALP, Breast cancer.

Introduction

Breast cancer continues to be a disease with tremendous public health significance⁽¹⁾. It remains the leading cause of cancer mortality for women in both developed countries (30%) and developing countries (14%)⁽²⁾ where its incidence rates have been continuously rising³. It is a disease characterized by prevalence, aggressiveness, unequal geographic distribution, and an increasing incidence throughout the world^(4,5). Environmental factors, including a nutritious diet, either alone or in combination, have been incriminated in the increase in cancer incidence rates and in its progressive spread into all regions of the world. Many experimental, clinical and epidemiological studies have revealed that a number of factors influence breast cancer etiology. Hormonal factors^(6,7), reproductive factors, genetic factors, lifestyle, and dietary factors⁽⁸⁾. Primary prevention of breast cancer is still not available except by extreme measures such as a prophylactic mastectomy for women who are genetically at high risk, so efforts to promote early detection continue to be the major focus in fighting breast cancer. The goal of early detection is to diagnose and treat breast cancer patients at an early stage when the prognosis for long-term survival is best. Prognosis is generally more favorable for women with an early-stage disease than for those with more advanced disease as early detection is associated with decreased mortality⁽⁹⁾.

Aberrant energy metabolism is a common feature of cancer⁽¹⁰⁾. In normal cells, when oxygen is available, pyruvate generated during the breakdown of glucose is utilized to produce energy through oxidative phosphorylation. In contrast, tumor cells prefer pyruvate metabolism via the anaerobic pathway regardless of oxygen availability, leading to inefficient fuel production and formation of lactate. This anomalous metabolic preference is known as the Warburg effect or ‘aerobic glycolysis’^(11,12). Lactate dehydrogenase (LDH) is the enzyme responsible for the conversion of pyruvate to lactate during glycolysis⁽¹³⁾. It is expressed in all tissues and its A and B subunits, coded by two different genes LDH-A and LDH-B, combine to construct five iso-enzymes (LDH1 to LDH5) with selective distribution among tissues and in serum⁽¹⁴⁾. In addition, LDH is known as a

marker for tissue injury, inflammation, hemolysis, and myocardial infarction^(15,16,17). Elevated LDH levels are seen in cancer patients, and its prognostic value has been shown in several malignancies such as germ cell tumors, lymphoma, melanoma, and renal cell carcinoma^(18,19,20,21). Recent evidence linking LDH-A and the oncogene c-MYC suggests that metabolic derangements may occur before the tumor becomes macroscopic, that is before it is clinically detectable⁽²²⁾. The role of these early metabolic disturbances in the context of cancer survival remains poorly understood. Currently, there is also a lack of information on how serum LDH contributes to cancer-specific survival in long-term follow-up⁽²³⁾.

Gamma-glutamyltransferase (GGT) is a cell-membrane bound enzyme⁽²⁴⁾, widely present on the external surface of most cells⁽²⁵⁾, with high secretory activity like liver, kidneys and pancreas⁽²⁴⁾. GGT is responsible for the glutathione (GSH) metabolism, catalyzing the degradation of extracellular GSH and further promoting amino-acid recovery for subsequent intracellular GSH synthesis⁽²⁶⁾. Intracellular GSH acts as an antioxidant, neutralizing free radicals and so plays a decisive role in protection against oxidative stress during cell metabolism⁽²⁵⁾. Therefore, GGT and GSH are increasing in circumstances of oxidative stress like carcinogenesis⁽²⁶⁾ and have been implicated in numerous other diseases like cardiovascular disease, diabetes, metabolic syndrome, malignancies and is associated with an increase in all-cause mortality^(27,28).

Alkaline phosphatases are a group of phosphatidylinositol-anchored membrane proteins with wide substrate specificity^(29,30). It comprises a group of enzymes that catalyze the hydrolysis of phosphate esters in an alkaline environment, generating an organic radical and inorganic phosphate⁽³¹⁾. Like other enzymes, ALP has many iso-enzymes. In healthy adults, this enzyme is mainly derived from the liver, bones and in lesser amounts from intestines, placenta, kidneys and leukocytes⁽³²⁾. An increase in serum ALP levels is frequently associated with a variety of diseases such as extrahepatic bile obstruction, intrahepatic cholestasis, infiltrative liver disease, hepatitis and cancer. However, the elevation of ALP less than three times the normal level is generally considered to be non-specific and insufficient to provide a definite diagnosis⁽³³⁾.

Materials and methods

Fifty women diagnosed with breast cancer were involved in this cross-sectional study, they attended Khartoum Oncology Hospital for diagnosis, treatment and follow up. They were diagnosed and classified in groups according to the grade, grade I, II and III. Approval of the study to be performed was obtained from each patient and hospital administration as well. Under hygienic condition blood was withdrawn in plane containers from each subject considering labeling and data obtained from each one, blood allowed for clot formation and subsequent serum separation which was preserved at -20 for later chemical analysis. Chemical analysis targeted enzymes, GGT, LDH, and ALP, they were measured by means of spectrophotometer method, using Biosystem device, reagents were Biosystem trademark, they were ready to use.

Results

A total of 50 women with an established diagnosis of breast cancer were involved in this study, the mean+SD of the duration of the disease was (3.52+1.89) years and their age mean+SD was (38.1+6.31) years as in table 1.

Table (1): Age and duration of breast cancer

Parameters	Minimum	Maximum	Mean±SD
Age (Years)	26.00	49.00	38.10±6.31
Duration (Years)	1.00	8.00	3.52±1.89

Blood samples were sent for measurement of GGT, LDH, and ALP. The results obtained were analyzed statistically and the p value of 0.05 consider significant. Data gave significant difference for both ALP and LDH with P-value 0.000 and 0.036 respectively and no significant difference with GGT with p-value 0.298 as in table 2.

Table (2): Mean±SD of ALP, GGT and LDH among breast cancer women and control

Parameters	Case Mean±SD	Control Mean±SD	P-value
LDH (U/L)	455.50±54.54	337±95	0.000
ALP (U/L)	88.52±20.88	70±25	0.036
GGT (U/L)	38.56±16.39	41±7	0.298

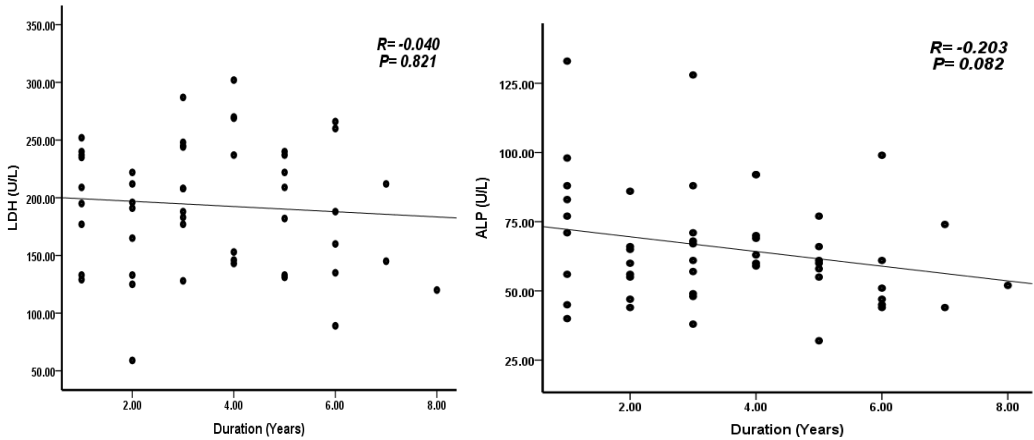
In this study a highly significant increase in mean serum of ALP and GGT was observed in carcinoma breast patients when compared to controls (P < 0.000). Enzymes levels among the three grades of breast cancer brought significant difference for LDH at grade II and III as p.values 0.022 and 0.001 respectively, but ALP didn't show any difference among the three grades while GGT has an only significant difference at grade III as p.value 0.022 as in table 3.

Table (3): LDH, ALP, and GGT among the three grades of breast cancer

Parameter	Grade I (Mean±SD)	Grade II (Mean±SD)	Grade III (Mean±SD)
LDH (U/L)	128.66±14.04	186.00±42.28	204.86±59.14
P-value		0.022	0.001
ALP (U/L)	65.33±5.50	62.44±23.57	67.44±20.33
P-value		0.234	0.112
GGT (U/L)	28.66±13.31	34.27±12.06	42.24±18.24
P-value		0.324	0.022

Significant difference as p.value <0.05

Correlation of enzymes level and duration of breast cancer show no significant correlation, but only GGT has a significant negative correlation as in figure 1, 2 and 3.



Figures (1) and (2): Show correlation analysis between duration and LDH (left one) and duration and ALP (right one).

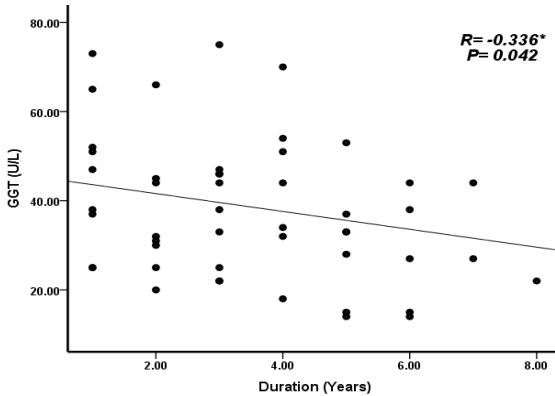


Figure (3): Shows the correlation between duration and GGT

Discussion

Breast cancer is the second most common cancer worldwide after lung cancer, the fifth most common cause of cancer death, and the leading cause of cancer death in women. The global burden of breast cancer exceeds all other cancers and the incidence rates of breast cancer are increasing ⁽³⁴⁾. In this study measured parameters (LDH, GGT, and ALP) were conducted among breast cancer women in Khartoum oncology hospitals, they were sorted to 3 stages I, II and III, data obtained compared with normal range gave significant difference LDH for grade II and III.

For enzymes levels among the 3 grades of breast cancer, grade I showed no change or no difference as the other grades have, a significant difference for LDH for both grades II and III as p values 0.022 and 0.001 respectively, GGT brought

significant difference with grade III with p. value 0.022 and ALP did not bring any difference among the three grades while GGT has only significant difference at grade III as p.value 0.022, this in agreement with a study concerned about the same manner and enzymes, referring that these enzymes play potential roles in tumor growth and have prognostic relevance in a diverse array of tumors, it found increased level of the 3 enzymes ALP, LDH and GGT than normal limits and mentioned that LDH increased had poor effects on both overall survival and distant metastasis-free survival with P 0.009 and 0.035 respectively and ALP had poor effects on both overall survival and local recurrence-free survival (P = 0.037 and 0.039, respectively⁽³⁵⁾). Considering finding of ALP in this study (p-value 0.23 and 0.11 for both grades II and III), it disagrees with other Indian study measured ALP only among breast cancer patients and observed ALP has increase level found among women with grade IV with P value <0.001, revealing that ALP can be used for differentiation of stages of breast cancer⁽³⁶⁾. Somehow our study agrees with other study included 102 confirmed cases of carcinoma breast with and without metastasis and 25 healthy non-pregnant females. They were evaluated for blood levels of LDH, ALP, and GGT and other parameters before and 21 days after mastectomy. A significant increase (p<0.001) was observed in LDH level in cancer patients without metastasis in comparison to normal control subjects. Patients with metastasis had further elevated (P)<0.001) levels of ALP and GGT as compared to non-metastatic patients. Mastectomy in both the cases i.e. with and without metastasis resulted in a non-significant decrease in all the biochemical parameters suggesting that longer follow up could confirm post surgery decrease in the biochemical parameters⁽³⁷⁾.

Conclusion

Enzymes measured in this study (LDH, ALP, and GGT) were found elevated and yet have an obvious significant difference with a progression of the disease to further grades.

Funding

This study is self-funded.

Conflict of Interest

The authors declared no conflicts of interest.

Acknowledgment

We gratefully acknowledge colleagues at Al-Amel Oncology Center and the staff for their help and support.

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