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The Role of Total Fucose and Protein Bound Fucose in Patients with breast Cancer

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ABSTRACT

Oligosaccharides, arrangements of sugars related with proteins and lipids, are one the most munificent sorts of atoms and fundamentally different. Fucose is one of the most significant oligosaccharide changes associated with malignant growth and aggravation. Novel advances in glycemic have recognized various sorts of glyco-biomarkers containing L-fucose that are formed to particular kinds of disease. **Aim** of the examination : to assess the job of complete L-Fucose (TF) in patients with demonstrated breast malignant growth and assessed to discover the chance of utilizing such boundaries as a biomarker in the analysis of breast cancer disease patients contrasted with control.

The statistical analyses for the data obtained in the present study showed Significant increase in TSF level in breast cancer patients compared to that of normal controls ($P<0.0005$) and showed significant decrease ($P<0.0005$) in breast cancer who respond well to treatment (clinical remission). Thus TSF may be used as marker of tumor activity during follow-up under treatment. Significant increases in PBF levels in breast cancer patients compared with normal controls ($P<0.0005$), and showed significant decrease ($P<0.025$) in breast cancer patients with clinical remission. Thus serum PBF may be used as a marker of tumor activity during follow-up treatment. Moderate decrease in TP level in breast cancer patients compared with normal controls ($P<0.0005$). and less significance ($P>0.05$) when compared to pathological controls.

Conclusions: The degree of absolute serum fucose and protein bound fucose increment in patients with breast malignant growth illness, this might be acceptable biomarker in analysis and lineup of patients with breast cancer .

Keywords: Fucose , Fucose binding Protein , breast cancer

Introduction

In a number of malignancy patients, Serum fucose levels demonstrated marked differences from normal. contradictory relationships of serum glycoprotein-fucose concentration and the malignancy of tumor [1,2], a vast amount of literature has accrued that specifically shows a higher L-fucose content in the serum glycoproteins of the Malignancy patients, and the diagnostic value of its determination [3,4]. Increased fucose sphingo-lipid levels were also explained in individual of malignancy and transformation, the accumulation of the novel fucosyl-ceramide in some human colon tumors, mentioned previously [5]. Fucosyltransferases levels were. However, found to be increased in patients having different neoplasia [6] , and assay of the level affords a means of diagnosis of neoplasia in these cases ,and of ascertaining the success of surgery , chemotherapy, or radiation[5].

Glycoprotein is now thought that alterations in the outer surface of cancer cells are important to abnormal growth and activities of the cell [4]. These differentiating surface properties can be called tumors marker[2]. Glycoprotein and glycolipids are components of cell surface fundamentals , necessarily to cancer properties[3]. Cancer cells often has an increased concentration of glycoconjugate on outer surface of malignancy cells, which increases the concentration in the blood stream [7]. Various tumors have been proved to have breast, lung and prostate[6] .In certain disease, L-Fucose metabolism also alterations. In some cancer lead to high concentration of serum fucose, and in liver tumor the activity of α-L-Fucosidase in serum increases[2]. explanation an evident connection between high convergence of protein–bound fucose in serum and the presence of malignant breast masses. Beforehand, we clarified strategy for the chromatographic examination of the three unbiased protein – bound sugars (mannose, Galactose, and fucose) in serum and expository information on tests from ordinary ladies and those

with metastatic breast cancer[6]. Convergence of the three carbohydrate were for the most part expanded in hydrolysates of serum from the patients with danger, the expansion in fucose being proportionately higher than mannose and Galactose [2]. Determination of protein-bound carbohydrate of glycoproteins have been used as index to glycoproteins levels[5].

Serum TP have been investigated by numerous creators for finding and organizing disease and for assessing different restorative methodologies. Plucinsky et al. discovered that there is a to different malignant growths in study. Likewise discovered there is noteworthy increment in serum huge reduction in the mean TP levels in patients with tumor melanoma, carcinoma of the ovary, colorectal disease and bosom disease, and diverse other danger related TP levels in malignant growth patients react well to treatment while it is stay inside low level in non-responder patients[8]. On other hand Kemal et al. discovered that there is a sufficient increment in serum TP levels in patients with renal malignancy when contrasted with typical people [9]. Ruddy etal. demonstrated that there is no noteworthy distinction in absolute protein of bone malignancy. They presume that the abatement in egg whites level related with significant levels in different divisions of all out proteins particularly gamma globulin which give bogus impression that complete protein fixation is with in ordinary [10] .

Methods

Patients: Thirty one woman with newly diagnosis breast cancer(stage I), ten women positive axillary node (stage II) and forty four woman with benign breast tumors were seen at the center of Breast Cancer in Alleluia Hospital for Woman Care form June 2012 till April 2014. The total physical assessment was done to every patient. The last determination was set up by desire of blisters (FNA) to check cytology, histology (biopsy) and Mammography. Other twenty nine normal healthy were considered as normal controls.

Sampling collection: Blood was gathered by venipuncture from the patients. The examples gathered on various days in light of patients visiting emergency clinic in various days. The serum was isolated, centrifuged and put away at - 20°C.

Chemicals: All basic research center synthetic substances and reagents utilized in this examination were of Analar grade except if in any case determined and were gotten from the accompanying organizations: BDH organization ;(UK). H2SO4, L-cysteine, Bio Maghreb organization; absolute protein pack, and Sigma organization ;(USA). L-Fucose.

Statistical analysis: The SPSS 20 was used to analysis the data, T-test score was used , P-value <0.05 were considered significant.

Estimation of Total Fucose: The rule of this strategy relies upon the development of chromogen on adding L-cysteine to test tube holds the example and sulphuric acid[2]. The shading produce framed by Fucose has most extreme ingestion at 396 nm, and about no assimilation at 430 nm. Different sugars, which additionally structure a shading item with cysteine and ingest maximally at 396 nm, have an equivalent retention at 430 nm (Figure (1), so the distinction in assimilation between these two frequencies is the ingestion due to Fucose only[1].
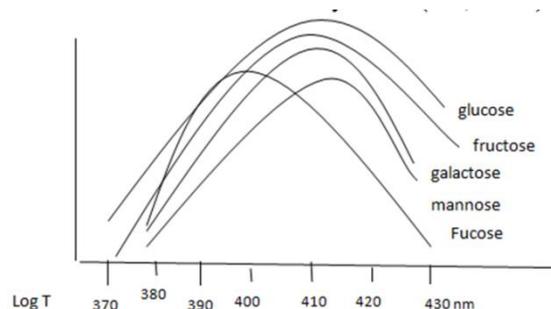

Figure (1): The Absorption of different monosaccharide sugars have an equal absorption at 430,X-axis the wave length nm ,y-axis absorption

Estimation of Protein –Bound Fucose

The protein-bound Fucose in serum was determined according to the method of Winzler [1]. Were precipitated protein from serum by addition of ethanol 95% and then dissolved in 0.1 N NaOH and then developing the Chromogen as fucose determination methods.

Estimation of Serum Total Protein

Cupric ion in an alkaline medium reacted with protein peptide bonds resulting in the realization of a coloured product whose absorbance is measured at 546nm[11].

Results

All out Fucose (TF) level were resolved to evaluate its utility in finding what's more, line up of patients with breast tumor infection. The mean ±SD of TF level expressed as mg /dl in sera of normal controls (12.46±3.64), pathological controls (17.9±4.92), and breast cancer patients negative axillary node (stage I) (22.280±8.1) stage II(20.27±5), also the ranges expressed as mg/dl of serum TSF in normal controls (8.6-21), pathological controls (10.2-30) ,Breast cancer patients stage I (11-47) and stage II(14.6-26).

Table (1) represents the comparism between the means of TF and serum total protein (TP) in sera of normal"controls, pathological controls, and breast cancer patients stage (I) and stage (II). Where the mean serum TF level normalized to the mean serum TP level as (TF/TP ratio) showed significant measure for breast cancer patients stage (I) and stage (II),when compared to normal" controls and pathological controls (2.64)."

Table (1): Comparism of the mean levels of total Fucose (TF) as mg/dl and total protein (TP) as g/dl in sera of normal controls , pathological controls and breast cancer patients stage I and stage II.

Group	n	TF (mg/dl)	TP (g/dl)	TSF/TP ratio (mg/g)
Normal controls	29	12.46±3.64	7.9±0.69	1.576
Pathological controls	44	17.9±4.92	6.77±0.83	2.64
Breast cancer (stage I)	31	22.28±8.1	6.93±0.76	3.21
Breast cancer(stage II)	10	20.27±5	7.26±0.56	2.79

The bio-statistical calculation and studies t-test for TF level in sera of normal controls pathological controls and breast cancer patients. The TF level from both breast cancer patients stage I ,stage II and pathological controls were significantly increased (P<0.0005) when compared to normal controls.

Table (2): Sensitivity, specificity, predictive values of positive and negative tests and efficiency test of TF were considered in using 12.45 mg/dl as cut-off value.

Sensitivity*	93.0%
Specificity*	72.0%
Positive Predictability	77.0%
Negative Predictability	91.0%
Efficiency*	83.3%

Serum Protein-Bound Fucose:
Serum PBF level was measured to evaluate its usefulness in diagnosis and follow-up of patients with breast cancer. The mean ±SD of PBF levels (mg/dl) in sera of normal controls were (8.0±1.5), pathological controls (11.11±3.98), breast cancer patients stage I (12.17±5.43), and stage II (12.83±5.8). Also the range of serum PBF were in normal controls (6--14.3), pathological controls (4.2—16.9) , breast cancer patients stage I (5—24) , and stage II (6.67-19.3).

Table (3) represents the comparism between the means of PBF and serum total protein (TP) in serum of normal controls, pathological controls , breast cancer patients stage I and stage II , where the mean serum PBF level normalized to the mean TP level as (PBF/TP ratio) showed significant measure for breast cancer patients stage I, and stage II, when compared to normal controls and pathological.

Table (3): Comparism of the mean levels of serum protein-bound fucose as mg/dl and total protein (TP) as g/dl in serum of normal controls , pathological and breast cancer patients stage I , and stage II.

Group	n	PBF (mg/dl)	TP (g/dl)	PBF/TP ratio(mg/g)
Normal controls	29	8.0±1.5	7.9±0.69	1.01
Pathological controls	44	11.11±3.98	6.77±0.83	1.64
Breast cancer stage(I)	31	12.17±5.43	6.93±0.76	1.76
Breast cancer stage(II)	10	12.83±5.8	7.26±0.56	1.78

The bio-statistical calculation and student t-test for PBF level in sera of normal controls, pathological controls, breast cancer patients stage I and stage II. The serum level PBF level form breast cancer patients stage I , stage II and pathological controls were significant increase (P<0.0005) when compared to normal controls.

Table (4) show Sensitivity , specificity , predictive values of positive and negative tests and efficiency test of serum PBF were using 8.02 mg /dl as cut-off value.

Table (4) : Predictive values of serum protein –bound Fucose (PBF) level in breast cancer patients using 8.02 mg/dl as cut-off value.

Sensitivity	74.0%
Specificity	48.0%
Positive Predictability	60.0%
Negative Predictability	63.0%
Efficiency	61.6%

Discussion

Variations in serum glycoprotein concentration and TF concentration are distinctive of many pathological conditions, including malignancy[6] has emboldened the study of fucose as a probable tumor markers in blood. In more common studies, total serum fucose (TF) level has been increased in malignant disease as result of increasing in carbohydrate bound to the protein when compared to controls. Several theories have been put forward in an try to explain the increased plasma glycoprotein levels observed in various disease states, but none of these can be accepted as being adequately substantiated. working with experimental tumors, assumed that the increase in protein-bound carbohydrate arises as a result of deploymerization of the ground constituent of connective tissue adjacent to the tumor, with subsequent liberation of these compounds into the circulation.[12] .

The results presented in this study revealed highly significant elevation in TF level in breast cancer patients stage I, stage II, pathological controls, and normal controls (P≤0.0005, P≤0.0005, and P≤0.0005 respectively) also there was significant between breast cancer patient stage I and pathological controls (P<0.0005) while there was no significant difference in TF level when compared to pathological controls (P<0.1).

The presence of higher ratio of TF/TP in breast cancer patients stage I, Stage II than that of normal healthy controls and pathological controls showed significant measure for breast cancer patients when compared to normal controls and pathological controls as shown in table (1). many authors explain the role of TP in staging cancer and for estimating various therapeutic approaches. Plucinsky et al. found that there is a significant decrease in the mean TP levels in patients with tumor melanoma, carcinoma of the ovary, colorectal cancer and breast cancer, and different other malignancy associated to other cancers in study.

Table (2) shows the criteria of diagnosis validity (sensitivity, specificity, positive predictability, negative predictability and efficiency tests) of TSF at breast cancer patients compared to that of pathological controls using 12.45 mg/dl as cut-off value (clinical decision) and were (93.0%, 72.0%, 77.0%, 91.0%,and 83.3% respectively). so that we can differentiate the non-malignant disease (pathological controls) from malignant disease (breast cancer patients) by using the TF level. Increased in TF level in patients with clinical remission is compatible with tumors regression .

Conclusion

In general we can conclude from this work that : TSF has higher diagnostic validity values in the current study which make the test may be useful as diagnostic tool to identify recurrence and TSF levels measurement provides that: Showed a significant increase in total serum fucose in the present breast cancer patient which was not reported previously. Valuable information for diagnosis can be used for monitoring disease status, The progress of the disease, Response to therapy.

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تقدير الكمي للفوكوز الكلي والفوكوز المرتبط بالبروتين في المرضى الذين يعانون من سرطان الثدي

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الملخص

متعدد السكريات ترتبط بالبروتينات والدهون، هي واحدة من أكثر أنواع الذرات وفرة ومختلفة اختلافًا جوهريًا. الفوكوز هو أحد أهم التغيرات المرتبطة بنمو الاورام السرطانية وتفاقمتها. الدراسات الحديثة المقامة على نسبة السكر في الدم تعرفت على أنواع مختلفة من المؤشرات الحيوية للكاربوهيدرات التي تحتوي على الفوكوز الذي يتكون لأنواع معينة من الأمراض. الهدف من الفحص: تقييم وظيفة الفوكوز الكامل (TF) في المرضى الذين يعانون من نمو سرطان الثدي وتقييمها لاكتشاف فرصة استخدام مثل هذه البارامير كمؤشر بيولوجي في تحليل مرضى سرطان الثدي مقارنة بالسيطرة.

أظهرت التحليلات الإحصائية للبيانات التي تم الحصول عليها في هذه الدراسة زيادة معنوية في مستوى TSF في مرضى سرطان الثدي مقارنة مع الضوابط العادية (P <0.0005) وأظهرت انخفاضًا كبيرًا (P <0.0005) في سرطان الثدي الذين يستجيبون جيدًا للعلاج. يمكن استخدام TSF كعلامة على نشاط الورم وبالتالي أثناء المتابعة تحت العلاج. زيادات ملحوظة في مستويات PBF في مرضى سرطان الثدي مقارنة بالضوابط العادية (P <0.0005) ، وأظهرت انخفاضًا كبيرًا (P <0.025) في مرضى سرطان الثدي بعد العلاج . وبالتالي يمكن استخدام PBF في المصل كعلامة على نشاط الورم أثناء متابعة العلاج. انخفاض معتدل في مستوى TP في مرضى سرطان الثدي مقارنة بالضوابط العادية (P <0.0005). وأقل أهمية (P> 0.05) عند مقارنتها بالضوابط المرضية.

الاستنتاجات: زيادة مستوى الفيوكوز الكلي في الدم وكذلك الفيوكوز المرتبط بالبروتين في المرضى الذين يعانون من مرض سرطان الثدي