The Importance of CA15-3 in the Follow up of Metastatic Invasive Ductal Carcinoma Iraqi Women

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Abstract :

Objective: The aim of this study is to find out the efficiency of CA15-3 in the follow up of metastatic invasive ductal carcinoma patients pre and post-treatment.

Method: Seventy five inavasive ductal carcinoma Iraqi patients with metastasis were admitted to Nuclear Medicine Hospital in Baghdad, all were females and have received the same treatment regimen which is represented by FAC: 5-fluorouracil, doxorubicin, cyclophosphamide given 3-weekly for 6 cycles. Those patients have been observed for four months until they completed their course of treatment. Also 15 samples of apparently healthy women were involved as a control. The blood samples (5 mL) were drown from all of studied cases in order to be used for measuring their serum level of CA15-3 (serum CA15-3 levels of patients were measured pre and post treatment) by using Enzyme-linked Immunosorbent Assay (ELISA) technique.

Result: Serum CA15-3 concentration was elevated in 36 patients (highly significant) from the total number of patients that had been considered in the study before starting therapy, 31 patients from them were followed up for four months while they completed their course of treatment. Then; their serum CA15-3 concentrations were measured again, the result showed that the CA15-3 values decreased in 27 patients to be within normal level, but increased in 4 patients more than its primary concentration before starting therapy.

Conclusion: CA15-3 is a sensitive tumor marker for diagnosing metastatic invasive ductal carcinoma and especially for monitoring treatment of those patients.

Keywords: CA15-3, invasive ductal carcinoma, Iraqi women.

Introduction:

Breast cancer is the malignant tumor that forms from the uncontrolled growth of abnormal breast cells. It usually affects tissues involved in milk production (Ductal and lobular tissues).

It is the most common malignancy in women and it remains one of the greatest health threats facing women around the world as we enter the 21st century (1).

Every year, one million new cases are reported world-..

wide, representing 18% of the total number of cancer in women, it has been established that one out of eight women in USA (2) and one out of 10 women in UK (3) will develop breast cancer at some point in her life.

In Iraq, breast cancer cases number were steadily rising after the 1991 war (4), the number of cases were 778 in 1990 which increased to 2154 case in 2001(5) and the number continue to increase sharply year after year till now (6,7).

The most frequent histology types of breast cancer in Iraq are Invasive ductal carcinoma which represent 77.2%, lobular carcinoma 9.8%, Comedo carcinoma 1.8 % and medulary carcinoma 1.5% and the rest percentage represent other histologic variants of carcinoma.

Studies with monoclonal antibodies in the sera and tumor

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tissue of patients with breast cancer have led to description of a tumor associated antigen, detected by monoclonal antibody DF3, a high molecular weight glycoprotein which is expressed with differentiation of mammary epithelium.

A second type of antigen has also been defined in the sera of breast cancer patients by the use of monoclonal antibody 115D8, originating from human milk fat globule membranes.

Subsequent studies have used both monoclonal antibodies DF3 and 115D8 in a bi-determinant immunoradiometric assay which has identified a circulating antigen designated CA15-3 (Carbohydrate Antigen) (8).

Patients and Method:

Patients: Seventy six patients with proved diagnosis of Invasive Ductal Carcinoma and had Metastasis (Not otherwise specified "NOS") on histopathological examination presented to the Nuclear Medicine Hospital in Baghdad.

These patients have been of different age groups, different geographic residencies and have been admitted to this hospital from September/ 2009 to the end of December/ 2009 , all of them have received the same treatment regimen which is represented by FAC (or CAF): 5-fluorouracil, doxorubicin, cyclophosphamide given 3-weekly for 6 cycles. Those patients have been observed for four months until they complete their course of treatment at the end of May/ 2010.

Control: Fifteen apparently normal individuals with comparable age range of patients.

Blood Sampling: Two ml of blood were collected in plain tubes from patients and from control individuals (all of them were female).

Blood samples were drawn at the morning between 9 and 11 a.m. by venapuncture using disposable syringes. The patients were newly diagnosed breast cancer patient with metastatic invasive ductal carcinoma, postoperative and pretreatment, their ages ranging between 24 -65 years old.

Samples collection, Processing and Storage: The blood samples were collected in dry tubes (plane tube) containing no additives, then centrifuged at 3000 rpm for 2-5 minutes to separate serum from blood cells, after that all serum samples were being stored at -20C°. Then assay procedure by using ELISA CA15-3 kits of DRG Company.

Statistical Analysis:

1- Descriptive statistics:

A. Statistical tables including observed frequencies with their percentages.

B. Summary statistic of the readings distribution (mean, SD).

C. Graphical presentation by (bar charts).

2 - Inferential statistics:

These were used to accept or reject the statistical hypotheses, they include the followings:

Chi-Square ($\chi2$).

Note: The comparison of significant (P-value) in any test were:

S= Significant difference (P<0.005).

HS= Highly Significant difference (P<0.001).

NS= Non Significant difference (P>0.005).

Result and Discusion:

Patient's ages ranged from 24 to 65 years old. Overwhelmingly, breast cancer incidence and death rates generally increase with age. During 2002- 2006, 95% of new cases and 97% of breast cancer deaths occurred in women aged 40 and older (9).

Regarding patients group, the mean age was 47.1 years old. It was obvious that the distribution of the patients was significantly occurred between 50-59 years age groups ($\chi2$ = 7.033, p < 0.001) as shown in figure (4-1).

In agreement with the results of the present study, a recent study in Iraq show that the incidence of breast cancer has significantly occurred after age of 40 6,7, also a group of researchers both in USA and Australia have found that the incidence of breast cancer increasing sharply after the age of 40 (10,11).

In contrast to the results obtained in this study, another study has found that during 2002-2006, women aged 75-79 had the highest incidence rate, 441.9 cases per 100,000. (9), these findings may be explained by the fact that the increasing life expectancies in western countries have resulted in an increase in the proportion of the population that is elderly. For instance, the percentage of the US population that was aged 75 or more years has increased from 2.6 percent in 1950 to 5.9 percent in 2000 (12).

Tumor marker study (CA15-3)

Serum tumor markers are widely used in patients with breast cancer, the majority of authors agree on the fact that tumor markers are useful since, elevated tumor marker levels almost certainly indicate an advanced stage in assessing the disease extent and serve as an early indicator of the success or failure of therapy in the monitoring of the treatment for metastatic breast cancer. Sensitive and specific detection of breast cancer tumor marker CA15-3 in human serum is an important step toward successful evaluation of clinical treatment and prediction of breast cancer recurrence (13).

The evaluation of serum CA15-3 concentration was involved in 75 patients and 15 healthy controls that included in this study, the result showed high elevation of serum CA15-3 levels in breast cancer patients in comparison to healthy group which is considered statically highly significant (P<0.01), the median and range of serum CA15-3 concentration were 36.57 and 4 – 194.4 (U/mL) for patients compared to 17.54 and 5 – 31.8 (U/mL) respectively for controls.

These results were confirmed by authors like Gion et al. (1999) who reported in their study which carried out in Venice/Italy on 150 breast cancer patients and 83 health controls, that serum CA15-3 concentrations were increased significantly in breast cancer patients compared to control group, the median CA15-3 level was 31.6 (U/mL) for patients compared to 18.2 (U/mL) for health controls, also Duffy, (1999)(14) confirmed that patients with primary breast cancer or metastatic breast cancer show elevated CA15-3 levels, while Kumpulainen et al.(2002) (15) concluded that in healthy people, CA15-3 concentrations are usually below 30 units/mL.

Furthermore, Bon et al.(1996) (16) in a large randomized study involved 938 apparently healthy women, has found that the median values of serum CA15-3 concentration was 17(U/mL). The current data showed that there were large differences among patients in their serum CA15-3 concentrations, with reference to the control group, serum concentrations of CA15-3 were increased in 48% of patients (36 patients from total number of 75 patients gave serum CA15-3 values more than upper normal serum CA15-3 values (31.8 U/mL) as registered by healthy controls in the present study), although some of those patients gave abnormal high values of CA15-3 which were (65, 69 and 75 U/mL), but there were 2 patients gave noticeable extremely higher CA15-3 concentrations (188.8 and 194.4 U/mL).

The remaining 39 patients gave serum CA15-3 values below 31.8 (U/mL) which consider within normal level, 2 patients from them gave very low serum CA15-3 concentrations which were (4 and 6 U/mL).

However, as the serum CA15-3 concentration increase in breast cancer patients as the patient outcome worsen and vice versa, as the CA15-3 concentration decrease as the patients outcome become better, as suggested by Gion et al.(1999)(17), who stated that the increase in serum CA15-3 concentration correlate positively with developing stages of breast cancer disease, also Keyhani et al.(2005) (18) observed a strong correlation between elevated CA15-3 levels and the progression of breast cancer.

Regarding age groups of patients in comparison with their serum CA15-3 concentrations, the result of this study show that the serum CA15-3 values of patients increased sharply in the patients with ages more than 59 years, compared with patients below this age, this considered statistically significant (P<0.05). This may be due to that the aggressiveness of breast cancer disease increase with increasing ages of patients, as CA15-3 concentration increase proportionally with breast cancer disease progression, so the CA15-3 was increased in those patients significantly. CA15-3 concentration should be monitored only in patients with a documented elevation of the marker, as high values of plasma CA15-3 are detected in at most 70-80% of women with metastatic breast cancer. If the publication of data had focused only those subjects with elevated pre-treatment concentrations, a better concordance between marker alterations and tumor response would have been found (14).

In the present study the serum CA15-3 concentration has been increased (pre-treament) in 36 from 75 patients, 31 patients from those of elevated CA15-3 concentrations were followed up for four months until they complete their course of treatment, then serum CA15-3 concentration was measured again for those 31 patients.

The average plasma CA15-3 concentration for breast cancer patients pre-treatment has been 54.7 U/mL compare to 24U/mL and 17.5U/mL for breast cancer patients post-treatment and healthy control group respectively ; this consider statistically highly significant (P<0.01).

Serum CA15-3 concentration has been decreased in 27 patients (highly significant) after completion of their course of treatment and became within normal level (bellow 30 U/mL), so this consider as an indicator for good responding to treatment and good prognosis.

Supporting to this suggestion, the authors concluded that 66% of patients with chemotherapy-induced disease regression exhibited a decrease in CA15-3 concentration, 73% of those with stable disease displayed increasing concentrations (19).

The remaining followed up 4 patients in this study showed an increase in their serum Ca15-3 concentrations (highly significant) to be more than its primary concentrations before starting therapy, so this borne in mind two probabilities:

The first one is that those patients might getting worsen (not responding to the treatment).

The second probability is that those 4 patients may have a symptomatic recurrence despite taking therapy, as reported in data collected from seven different studies, were combined; of 352 patients 67% showed elevated CA15-3 concentrations either before or at the time of recurrence, while of the 1320 patients without evidence of recurrent disease, 92% had normal CA15-3 concentrations, the mean lead time from marker elevation to clinical diagnosis of relapse ranged from 2 to 9 months (19).

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اهمية المَعلم السرطاني CA15-3 في تقدم سرطان الثدي لدى عينة من النساء العراقيات

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الخُلاصة:

يعد سرطان الثدي من اكثر انواع السرطانات التي تصيب النساء في العالم ، وقد تناولت الكثير من الدراسات والبحوث العوامل المسببة لهذا المرض من أجل التوصل للتشخيص المبكر والعلاج الناجح له.

تناولت هذه الدراسة حصراً النوع الخبيث والأكثر انتشاراً لسرطان الثدي وهو " سرطان القنوات المنتشر " حيث تركزت على بحث العلاقة بين هذا السرطان وتركيز المعلم السرطاني كما تناولت العلاقة بين تركيز هذا المعلم و اعمارعينة من مريضات سرطان الثدي العراقيات.

اجريت هذه الدراسة على 76 مريضة ادخلن لمستشفى الطب والإشعاع الذري في بغداد ، بالاضافة الى 15 امرأة سليمة (سيطرة)، حيث تم سحب عينة دم (2 مل) من جميع الحالات (المرضى والسليمات) لغرض استخدامها في قياس المعلم السرطاني المذكورة سابقا باستخدام تقنية

وجد زياده في تركيز المعلم السرطاني عند 36 مريضة من العدد الكلي للمريضات اللاتي ادخلن في هذه الدراسة قبل البدأ بأخذ العلاج، وتم متابعة 31 منهن طوال فترة العلاج التي استمرت اربعة اشهر، ثم تم بعدها قياس تركيز المعلم السرطاني من جديد لهؤلاء المريضات (المتابعات)، واكدت النتائج ان تركيزه قد انخفض لدى 29 مريضة ليكون ضمن المستوى الطبيعي، بينما ظهرت زيادة في تركيزه لدى 4 منهن ليكون اعلى من تركيزه الابتدائي قبل بدأ العلاج.