LEVEL OF TOTAL TUMOR PROTEIN 53 IN THE SERA OF IRAQI BREAST CANCER PATIENTS

Haider A. Hassan
Department of Biology, College of Science, University of Baghdad. Baghdad- Iraq.

Abstract

Breast cancer is one of the most common cancers in women. It occurs when abnormal cells in the breast divide uncontrollably and form tumors. Mutation of tumor suppressor gene P53 is a common event in this disease.

Level of total tumor protein 53 has been estimated in fifty serologic samples of Iraqi breast cancer patients and (50) of benign breast tumors as patients control in addition to (50) serologic samples which belong to apparently health volunteers. All these patients were attending to the Teaching Hospital of Baghdad and Al-Elwia hospital during the period between October/2007 and (April) 2008. They were diagnosed as having breast cancer by clinical and laboratory investigation as ultrasound waves test and fine needle aspiration (FNA). In addition to TP53, some hematological tests as hemoglobin, pocket cells volume, platelets count and white blood cells count were occurred. The current study revealed that (22%) out of total malignant breast cancer tumor patients were positive, while (8%) out of benign breast tumors patients were positive for TP53. The mean value of TP53 concentration in patients with breast cancer, benign tumors, and apparently healthy groups were (16.47 ± 31.81 pg/ml), (4.13 ± 4.94 pg/ml) and (2.27 ± 0.52pg/ml) respectively, with significant difference between malignant and benign cases.

The evaluate TP53 test by application of Receiver Operator Characteristic (ROC) showed that the accuracy of TP53 was (57%), however its specificity was proposed to be (68%) in comparison with (53.33%) sensitivity.

مستوى البروتين الورمي ٥٣ الكلي في مصول مريضات سرطان الثدي العراقيات

حيدر احمد حسن
قسم علوم الحياة، كـ ـ ـلية العلوم، جـ ـ ـامـ ـ ـعة بغداد. بغداد- العراق.

الخلاصة

يعد سرطان الثدي واحدا من اكثر الامراض السرطانية شيوعا لدى النساء. ويحدث عند انقسام خلايـا غير طبيعية من خلايا الثدي بصورة غير مسيطر عليها لتكون اوراما. وحدوث الطفرة الجينية في الجـين المثبط للورم (P53) يعد من الاحداث الشائعة في هذا المرض.

تم تقدير مستوى البروتين الورمي (TP53) الكلي في خمسون عينة مصلية لمريضـات سـرطان الثـدي العراقيات و(٥٠) عينة مصلية لمريضات اورام الثدي الحميدة كعينة ضبط وسيطرة للمرضى اضافة الـى (٥٠) عينة سيطرة لنساء متطوعات يبدون اصحاء.كن يراجعن مركز الكشف المبكر لسرطان الثـدي فـي مستشفى بغداد التعليمي ومستشفى العلوية في بغداد للفترة من تشرين الاول/٢٠٠٧ الى (نيسـان) ٢٠٠٨. وكذلك تم اجراء الفحوصات الدموية (فحص انضغاط كريات الدم وفحص الهيموكلوبين و تعداد كريات الدم البيض و تعداد الصفيحات الدموية).

ظهر باستخدام التحليل الاحصائي لهذه الدراسة ان (٢٢%) من مجموع مريضات سرطان الثـدي و(٨%) من مريضات اورام الثدي الحميدة قد اظهرن نتيجة ايجابية لهذاالفحص وان هناك ارتفاعا معنويا في مستوى البروتين الورمي ٥٣ في مصول مريضات سرطان الثـدي (١٦.٤٧± ٣١.٨١ بيكـوغرا/مـل)مقارنـة بمجموعة سيطرة الاصحاء (٢.٢٧ ± ٠.٥٢ بيكو غرام/مل) وسيطرة المرضى (٤.١٣ ± ٤.٩٤ بيكـو غرام/مل)(قيمة الاحتمالية <٠.٠١٢). وعند اجراء فحص ال ROC ظهر ان قيمة دقة الفحص accuracy كانت (٥٧%) في حين بلغت حساسية الفحص (٥٣.٣٣ %) وقيمة تحديد الفحص specificity (٦٨ %). نستنتج من ذلك انه بالامكان الاستفادة من قيمة البروتين الورمي ٥٣ كمؤشر ورمي يساعد فـي تشـخيص الاصابة بسرطان الثدي.

Introduction

Among women worldwide, breast cancer is the most common cause of cancer death [1]. The number of cases worldwide has significantly increased since the 1970 [2], some 45% of the more than 1 million new cases of breast cancer diagnosed each year [3]. Because the breast is composed of identical tissues in males and females, breast cancer also occurs in males, though it is less common [4]. The likelihood of developing breast cancer is low before age 35, but the risk rises after that [5]. It occurs when abnormal cells in the breast divide uncontrollably and form tumors. Heredity and radiation are the known causes of breast cancer. Poisonous substances (like beryllium, benzene, and chromium) in the environment, high levels of estrogen, high fat diet also may cause breast cancer [6].

The pathogenesis of breast cancer is unknown, but number of factors are associated with an increased risk which includes genetic factors which involved oncogenes and tumor suppressor genes which include:

1. Breast cancer predisposition gene-1 (BRCA-1).
2. Breast cancer predisposition gene-2 (BRCA-2).
3. Tumor protein 53 gene (TP53) [7].

   The TP53 tumor suppressor gene is one of the most commonly mutated gene in human cancers. It can exert antiproliferative effects, but equally important, it regulates apoptosis [8]. TP53 is a tumor suppressor gene that is mutated in more than 50% of tumors [9]. Furthermore about 25% of breast cancer appear to have either mutation in or loss of protein that starts the TP53 pathway [10]. The half-life of a normal p53 protein is short (20-60 minutes), but with some mutations, it increases and reaches up to 6 hours [11].

Materials and methods

Two study groups were investigated, which included fifty patients with breast cancer group P and 50 patients with benign tumors group as patient control (PC), and 50 of apparently healthy control group (HC). Their age group ranged between 18-69 years. All groups sera was submitted to immunological test to estimate the TP35 level using solid phase sandwich Enzyme Linked Immuno Sorbent Assay (ELISA) and to some hematological tests which include hemoglobin (Hb), pocket cells volume (PCV), total white blood cells (WBC_S) count, Platelets count. [12, 13].

Statistical analysis

Descriptive statistical which involved statistical tables including observed frequencies with their percentages and graphical presentation were done by using (bar-charts) for comprison of dichotomous variables and a value for P less than 0.05 was considered significant., furthermore inferential statistics which used to accept or reject the statistical hypotheses were done by using (SAS) system program/2001[14].

Results

I. Clinical and Demographical Picture of Studied Groups;

Some of the clinical features and the demographical picture of patients in comparison with controls have been listed in table 1.

II. Level of TP53 among the sera of the studied groups:

Level of TP53 has been measured in pg/ml using ELISA technique for its estimation among the sera of the studied groups. The result is listed in table 2.

Table 1: Clinical and Demographical Picture of the Studied Groups

Characteristics	(P)	(PC)	(HC)
Age mean (years)	50.9 ± 11.8	38.8 ± 11.8	25 ± 5.6
TP53 (Pg/ml)	16.47 ±31.81	4.13 ± 4.94	2.27 ± 0.52
Hb (mg %)	11.21 ± 0.9	12.24 ± 0.1	12.5 ±0.3
P.C.V (%)	37.5 ± 3.9	38.5 ± 3.8	39.8 ±2.07
W.B.C_S count (Cell/c.mm.)	5990.9± 2301.51	5060 ± 955.28	5100 ± 551.66
Platelets count (Cell/c.mm.)	182736.36 ± 48597.20	213120.23 ± 65878.30	19505.82 ± 35865.14
Total No.	50	50	50

Table 2: Mean distribution of immunol-ogical test TP53 level (Pg/ml) among studied group.

Studied groups		N	Mean	Std. Deviation	Comparison of Significant	
					P-value	Sig.
TP53 Pg/ml	Control	50	2.27	0.52	-	-
	Benign	50	4.14	4.94	0.773	NS
	Malignant	50	16.47	31.81	0.012	S
	Total	150	Benign Vs Malignant 0.029			S

III. Correlation between TP53 and other parameters:

Pearson correlation has been applied to study the correlation between level of TP53 and tumor stages and type of tumor. The results are listed in table 3.

Table 3: Mean of TP 53 levels (Pg/ml) among parameters of breast cancer (Malignant) patients.

Parameters		No.	Mean	SD	Comparison of significant	
					P-value	Sig.
Stages	I	10	22.32	33.77	-	-
	II	31	9.09	20.22	0.246	NS
	III	6	32.31	49.66	0.535	NS
	IV	3	41.54	68.19	0.351	NS
	Total	50	II Vs III = 0.1 NS		Vs IV= 0.09 NS	
			Vs IV =0.67 NS			
Type of tumor	IDC	45	17.45	33.25	-	-
	DCIS	3	1.927	0.91	0.423	NS
	LCIS	2	15.56	16.36	0.935	NS
	Total	50	DCIS Vs LCIS		0.645	NS

IV. Effect of the disease duration on the level of TP53:

It was proposed that TP53 level may be affected by the disease duration. This effect has been demonstrated in table 4.

V. Validity of TP53 Estimation:

ROC test were applied to detect sensitivity, specificity and accuracy as in table 5.

Table 4: Mean of TP 53 levels (Pg/ml) among duration group of breast cancer (Malignant) patients.

Parameters		No.	Mean	SD	Comparison of significant	
					P-value	Sig.
Stages	I	10	22.32	33.77	-	-
	II	31	9.09	20.22	0.246	NS
	III	6	32.31	49.66	0.535	NS
	IV	3	41.54	68.19	0.351	NS
	Total	50	II Vs III = 0.1 NS		II Vs IV= 0.09 NS	
			III Vs IV =0.67 NS			
Type of tumor	IDC	45	17.45	33.25	-	-
	DCIS	3	1.927	0.91	0.423	NS
	LCIS	2	15.56	16.36	0.935	NS
	Total	50	DCIS Vs LCIS		0.645	NS

Table 5: Validity tests (%) for immunological parameters.

parameters	Validity tests (%)		
	Sensitivity	Specificity	Accuracy
TP53 level (Pg/ml)	53.33	68	57

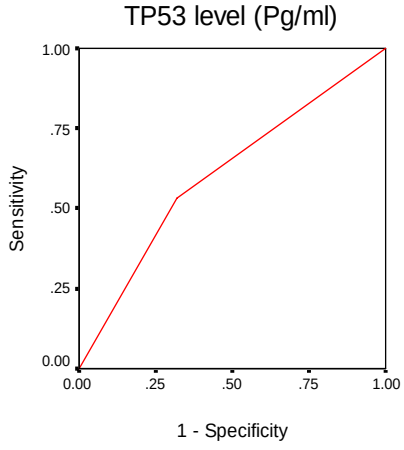

Figure 1: Validity tests (%) for immunological parameter.

Discussion

Generally, this study showed the demographical features which indicated that the mean of age of the majority of patients were within or above the menopause duration (50.9 ± 10.9 years) with highly significant difference in comparison with patients control (38.8 ± 11.8 years), this result is comparable to some extent with that of Lebanon previous study (49.8 ± 13.9) [15]. This result may be du to careless previous tumor or lesion (as cyst) in breast and let it without treatment or even without diagnosis and then convert to malignant tumor with prognosis of age.

The positivity of TP53 was observed in 11(16.5 %) of 50 Iraqi breast cancers, these results are similar to those of Michael A. Levesque et. al, 1998 [16]. Also we noticed that all hematological tests Hb, P.C.V., $W.B.C_S$ count and platelets count are within normal.

Studies suggest that, the presence of TP53 is strongly associated with P53 protein accumulation in tumors indicating that this immune response is triggered by the accumulation of P53 in the tumors [17]. In present study, we detect increase TP53 in 11(22%) of 50 sera patients with breast cancer.

The mean of TP53 of malignant cases was 16.47 ± 31.81 pg/ml in comparison with 4.13 ± 4.94 pg/ml and 2.27 ± 0.52 pg/ml which belong to disease and healthy control respectively, there is significant different between malignant benign breast tumors, these result is to those of Michael [16].

Regarding tumor stages, this study revealed that the production of TP53 is increased with progression of stages, so we noticed that the patients with stage has highly concentration of TP53, but there is no significant difference between stages. These result may be due to increase the number of mutated cancer cells which lead to increase accumulation of TP53 protein.

Furthermore, the highly concentration of TP53 is present in the intraductal carcinoma with no significant difference between types of tumor.

According to this study, it was clear that the TP53 concentration increased with disease duration which may be due to increase the accumulation of TP53 due to increase its half-life when mutation of TP53 gene occur, but there is no significant difference between duration periods which may belong to the small sample size of each period.

Normal hematological results were clear to all patients which mean there is no effect on the hematopoiesis.

It was important to evaluate TP53 may be as a diagnostic and predicator test by applied ROC test. It seemed to be that the sensitivity of this test was **53.33%** and conversely its specificity **68%** and accuracy **57%** which mean the dependence on this test is not enough to diagnose breast cancer.

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