Research Article

A study of the prevalence of Estrogen & Progesterone Receptor markers positivity in female breast cancer cases in Al-Yarmouk Teaching Hospital

Maitham M. Al-Khateeb *D.S, C.A.B.S*[1]*;* ***Louai Abdul Muna'am Al-Hilli*** *M.B.Ch.B*[2]

[1]*Department of General Surgery, College of Medicine, Al-Mustansiriya University, Baghdad, Iraq.*
[2]*Department of General Surgery, Al-Yarmouk Teaching Hospital, Baghdad, Iraq.*

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Address for Correspondence:
Dr. Maitham M. Al-Khateeb
Department of General Surgery, Al-Yarmouk Teaching Hospital, Baghdad, Iraq

Abstract

Background: Breast cancer accounts for 26% of all new cases of cancer among women and is the leading cause of death in those aged between 20-59 years, Estrogen and Progesterone receptors play an important role in the development and prognosis of breast cancer, their measurement by immunohistochemical assay is vital in the hormonal management of breast cancer.

Aims: To estimate the prevalence of Estrogen and Progesterone receptors positivity in female breast cancer cells in Al-Yarmouk Teaching Hospital.

Patients & methods: A prospective study for breast cancer cases in females in Al-Yarmouk Teaching Hospital for the period from **1st** of July **2009** to **31st** of December **2010** , By using immunohistochemical study in the Histopathological department of Al-Yarmouk Teaching Hospital , the results were correlated with age of the patients, tumor size, grade and tumor histopathological type**.**

Results: Total No. of patients studied were fifty. During the study, the age frequency was higher in the postmenopausal group (54%); the Estrogen Receptor (ER) +ve highest content (55.55%) was observed in that group, the Progesteron (PR) +ve highest level (52.17%) was observed in a premenopausal group (46%),Receptor positivity in Tumor size of the favorable type means equal to or less than 5 cm present in (92%) of the cases in which ER+ve level (58.69%) is the highest in that type, while the PR+ve level was (50%) in the unfavorable type means size above 5 cm which represent (8%) of the total cases. Regarding the relation with Tumor Grade the frequency of Receptors positivity in grade II (56%) more than grade I (24%) then grade III (20%), The highest ER+ve level (75%) was observed in grade I while PR+ve highest level (42.85%) in grade II, In studying the histopathological types the most prevalent type is infiltrative duct carcinoma (I.D.C) (62%) which have the highest ER+ve level (64.51%) while PR+ve level (42.10%) was high in infiltrative lobular carcinoma (I.L.C) type.

Conclusions: Breast cancer in this hospital has the same pattern of age incidence worldwide and histopathological type but of the more favorable type with lower Grade of malignancy. This reflects a changing trend towards less aggressive pattern. The variability of ER & PR positive contents in this study needs further studies to compare our results with because the Iraqi cancer registry lacks data on hormonal receptor contents of breast cancer cases.

Keywords: ER, PR, Breast cancer, Immunohistochemistry, Hormone receptors, Histopathological Grades

INTRODUCTION

Breast cancer is the most common site-specific cancer in women and is the leading cause of death from cancer for women aged 20 to 59 years. It accounts for 26% of all newly diagnosed cancers in females and is responsible for 15% of the cancer-related deaths in women,[1,2] Breast cancer remains one of the most dreaded human diseases. [3,4] It was predicted that in the United States in 2008 / 40,480 patients died from breast cancer.[5]

Breast cancer is uncommon under the age of 35 years and rare under the age of 20 years, its incidence continues to rise with age.[6] It has been reported that the increase in breast cancer incidence occurred primarily in women 55 years and older and paralleled a marked increase in the percentage of older women who were examined by mammograms.[7]

The average lifetime risk of breast cancer for newborn U.S. females is 12%,[8,9] and the genetics of the disease is now provoking speculation regarding possible hereditary influences on breast cancer risks that are related to racial or ethnic ancestry.[10,11] These risk factors include increasing age; mutations in breast cancer risk genes (including BRCA1 and BRCA2, PTEN, and p53) and other factors related to a family history of breast cancer;[12] hormonal and reproductive factors, including early menarche, late menopause, nulliparity,[13] and the use of exogenous hormones and; environmental factors, including diet and certain pathologic findings within breast tissue, including previous breast cancer and various premalignant lesions.[14] There are also number of molecular markers that can be correlated with prognosis, including estrogen receptor (ER) status and HER-2 / neu gene amplification.[15]

Histopathological types of breast cancer:[16,17]

Invasive ductal carcinoma: 70-80%. Invasive lobular carcinoma: 5-10%. Medullary carcinoma: 1-5%. Colloid (mucinous) carcinoma: 1-6%.Tubular carcinoma: 2%, 6. Invasive papillary carcinoma: less than 1%.

Grading is according to modification on the scarff-Bloom and Richardson grading scale scored on the basis of 1-3, Nottingham combined histological grading (NCHG) has resulted in widely accepted reproducible grading system.[18]

Staging: according to the American Joint Committee.[19]

Risk factor biomarkers are those associated with increased cancer risk, these include familial clustering and inherited germ line abnormalities, proliferative breast disease with atypia, and mammographic densities.[20,21]

Steroid Hormone Receptor Pathway:- Breast cancer risk is related to estrogen exposure over time, in postmenopausal women, hormone replacement therapy consisting of estrogen plus progesterone increases the risk of breast cancer by 26% compared to placebo.[22] The identification of steroid receptors in the 1960s and their analysis in breast cancer in the 1970s resulted in evidence that they could aid the identification of cancers that were likely to respond to endocrine treatment.[23] Tumors positive for estrogen or progesterone receptors have a higher response rate to endocrine therapy than tumors that do not express estrogen or progesterone receptors. Approximately 70% of breast carcinomas are hormone receptor- positive; Approximately 60% of patients with ER-positive tumors respond to endocrine therapy, but only 5% - 10% of patients with ER-negative tumors do so, and responses probably result from false-negative receptor results.[24]

Testing for estrogen and progesterone receptors should be performed on all primary invasive breast cancer specimens.[25,26] The tumor hormone receptor status should be ascertained for both premenopausal and postmenopausal patients to identify patients who are most likely to benefit from endocrine therapy.[27]

Breast cancer is the predominant malignancy where oncologists use predictive markers clinically to select treatment options.[28,29]

The determination of ER and PR by Immunohistochemistry is equal to biochemical assays for predicting the response of metastatic disease to Tamoxifen,[30] and is superior for predicting response to adjuvant endocrine therapy. ER is now routinely assessed in breast cancers worldwide, though PR is not always assessed in parallel.[31]

PATIENTS AND METHODS

This is a prospective study which was done in Al-Yarmouk teaching hospital, for the period from **1st of July 2009 to 31st of December 2010**,patients who had mastectomy for breast cancer were studied for the histopthological type of the breast cancer whether **Infiltrative ductal carcinoma (I.D.C)** or **Infiltrative lobular carcinoma (I.L.C)** and By using the **immunohistochemical study (IHC)** we tested the

collected samples for the presence of **ER & PR** in the breast cancer cells in this hospital, all immunohistochemical tests were Examined by the same histopathologist by using :

- ❖ Slices taken from the paraffin cube sample of mastectomized breast cancer patient.
- ❖ Fixing the sample slice on a ***charged slide.***
- ❖ Using **Dako (K5207)** ***IHC-reagent kit (made in Denmark)*** for Antigen antibody reaction on the tissue slices showing the ER/PR positivity on the slide.

We correlated ER and PR positivity with **age** of the patients as(premenopausal and postmenopausal) , **grade** of the tumour **(I,II,III)** according to Scarff-Bloom and Richardson grading scale, **tumor size** as **favorable** below or equal to 5cm (relatively early) and **unfavorable** above 5cm(relatively late), and the **histopathological type** of the tumour . The P-value was calculated.

RESULTS

The total number of cases collected during the period of the study was **50** cases. Regarding the **age** of patients in correlation with **receptor positivity** as seen in Table .1 were as follows; patients were divided into two groups, **premenopausal** (below or equal to 50 years), total No. **23** cases equal to **46%** and **postmenopausal** (above 50 years), total No. 27 cases equal to **54 %**:

- **ER+ve** tumor content in premenopausal cases were **11** (47.82) out of these 23 cases.
- **ER+ve** in postmenopausal cases were **15 (55.55%**) out of these 27cases.
- **PR+ve** in premenopausal cases were **12 (52.17%**) out of these 23 cases**.**
- **PR+ve** in postmenopausal cases were **12 (44.44%**) out of these 27 cases**.**

The total ER+ve cases were **26** equal to **52%** of total 50 cases, while total PR+ve cases were **24 cases** equal to **48%** of total 50 cases, the P-Value for ER/PR +ve cases with age was **> 0.05**.

The **size** of breast cancer cases in correlation with **receptors positivity** seen in Table .2 were as follows;

Number of cases below or equal to 5cm Favorable (**relatively early**) type was **46** equal to **92%** and cases above 5cm Unfavorable type(**relatively late**) was **4** equal to **8%**, in relation to receptor positivity:

- **ER+ve** in cases below or equal to 5cm was **27 (58.69%**) of these 46 cases**.**
- **ER+ve** in cases above 5cm were **2 (50%)** of these 4 cases**.**
- **PR+ve** in cases below or equal to 5cm **were 19 (41.30%)** of these 46 cases**.**
- **PR+ve** in cases above 5cm were **2 (50%)** of these 4 cases**.**

Table 1. Correlation of ER and PR Positivity to Age of patients

Age	ER+ve		PR+ve		Total No.	
	No.	Percent %	No.	Percent %	No.	Percent %
Premenopausal (Equal or below 50 Yrs)	11	47.82	12	52.17	23	46
Postmenopausal (Above 50 Yrs)	15	55.55	12	44.44	27	54
Total No.	26	(52%)	24	(48%)	50	100%
P-Value	>0.05					

The total ER+ve cases were **29 (58%)** out of total 50 cases, while total PR+ve cases were **21 (42%**) out of total 50 cases, the P-Value calculated for ER and PR +ve cases in relation to size of the cancer was **> 0.05**.

The relation between **tumor grade** and **receptor positivity** as seen in Table .3 were as follows:

Grade I cases **12 (24%)**, **Grade II** cases **28 (56%), Grade III** cases **10 (20%),** while for receptor positivity;

- **ER+ve** in Grade I were **9** cases **(75%)** of this grade cases**.**
- **ER+ve** in Grade II were **16** cases **(57.14%**) of this grade cases**.**
- **ER+ve** in Grade III were **6** cases **(60%)** of this grade cases**.**
- **PR+ve** in Grade I were **3** cases **(25%**) of this grade cases**.**
- **PR+ve** in Grade II were **12** cases **(42.85%**) of this grade cases**.**
- **PR+ve** in Grade III were **4** cases **(40**) of this grade cases**.**

The total ER+ve cases were **31** in all grades (62**%)** out of total 50 cases, while total PR+ve cases were **19** in all grades (**38%**) out of total 50 cases, The P-Value when calculated for ER/PR +ve cases was **> 0.05.**

The relation between **tumor histopathological type** and **receptor positivity** as seen in Table .4 were as follows;

I.D.C cases **31 (62%)**, while **I.L.C** cases **19 (38%**) out of total 50 cases, for receptor positivity the results were:

- **ER+ve** in IDC were **20** cases **(64.51%)** out of these 31+ve cases**.**
- **ER+ve** in ILC were **11** cases **(57.89%**) out of these 19 +ve cases**.**
- **PR+ve** in IDC were **11** cases **(35.48%**) out of these31+ve cases**.**
- **PR+ve** in ILC were **8** cases **(42.10%**) out of these 19 +ve cases**.**

The total ER+ve cases were **31 (62%**), while total PR+ve cases were **19 (38%**) out of the total 50 cases, The P-Value calculated was **> 0.05.**

Table 2. Correlation of ER and PR positivity to tumor size

Tumor size	**ER +ve**		**PR +ve**		**Total No.**	
	No.	Percent %	No.	Percent %	No.	Percent %
Below or Equal to 5 Cm (Favorable)	27	58.69%	19	41.30	46	92
Above 5 Cm (Unfavorable)	2	50%	2	50	4	8
Total No.	29	(58%)	21	(42%)	50	100%
P-Value	>0.05					

Table.3 Correlation of ER and PR positivity to tumor Grade

Grade	**ER +ve**		**PR +ve**		**Total No.**	
	No.	Percent %	No.	Percent %	No.	Percent %
I	9	75	3	25	12	24
II	16	57.14	12	42.85	28	56
III	6	60	4	40	10	20
Total No.	31	(62%)	19	(38%)	50	100%
P-Value	>0.05					

Table 4. Correlation of ER& PR positivity to Tumor histopathological type

Histopathology	ER +ve		PR +ve		Total No.	
	No.	Percent %	No.	Percent %	No.	Percent %
I.D.C	20	64.51	11	35.48	31	62
I.L.C	11	57.89	8	42.10	19	38
Total No.	31	(62%)	19	(38%)	50	100%
P-Value	>0.05					

DISCUSSION

Our work depend mainly on Immunohistochemistry (IHC) markers, ER may be the best example of tumor biomarker with an assay that drives therapeutic decision-making. Since ER and PR represent a well established predictor of response to endocrine therapy in breast cancer, ER and PR assays have been routinely used in the selection of appropriate therapy for breast cancer patients for more than 30 years.[28] However, it is well known that up to 30%- 40% of breast tumors with positive hormone receptor status do not respond to endocrine therapy.[29]

In this study the total number of cases collected during the period of the study was **50** cases and this is not a high figure for surveying a sector of population, the reason for that is the non availability of the IHC Kit in hospital at the time of the study which we were obliged to purchased from the private market and the long waiting list and restricted list cases in general surgery theaters of the Hospital because of the renewal work.

Regarding the correlation between age of patients and the positivity of hormone receptor content, there was higher frequency of female breast cancer in our study at the postmenopausal age **27** cases(54%), in which the **ER+ve** content was **15** cases **(55.55%**) of postmenopausal cases, while the **PR+ve** content is a little higher in percent in premenopausal cases **12 (52.17%**), than postmenopausal cases 12 **(44.44%**), this is consistent with a studies done by **Zhabg,1997,**[32] and **AlFallouji,1998**[16] Both of these studies showed that the **5th** decade has the highest frequency of cases followed by the **6th** decade. That was supported by an international study by **Kerby I. Bland,2004**[7] Which showed that the **5th** and **6th** decade equal to **78.2%** of his collection, that was rather similar to what was reported in the **Iraqi Cancer Registry center;1994, 1995, 1997, 2004.**[33] The results of a local study done by **Assad Aljanabi, 1997,**[34] which was done in Al-Najaf/Iraq showed that the highest frequency of breast cancer was displayed in the **4th** decade, **Nissan et al;2004**[35] found that Palestinian women found to be younger at the time of diagnosis and it is well known that breast cancer is diagnosed at a younger ages in developing countries.

Regarding the size of breast cancer cases in correlation with receptor positivity as seen in (**Table 2)** it is observed that tumor size which is **5cm or less** (Favorable prognosis),[36] is found in the majority of cases, **46 cases** (**92%**) out of total cases, In this group the ER+ve found in 27 cases **(58.69%),** While cases **above 5cm(**Unfavorable prognosis) found in **4** cases (**8%)** out of total cases; In this group ER+ve cases found to be equal to PR+ve cases 2 for each (**50%**) , which indicate that the female education regarding early consultation in breast clinics of general hospitals is improving for early detection of breast cancer in our hospital and this is similar to a local study done by **N.Alasheeri; 1999,**[37] in Sulaymania Medical Complex/ North of Iraq, where tumor size of **2-5cm** constitute **52%** of cases while tumour sizes more than **5cm** constitutes **19.7%**, A regional study done by **Nagi et al; 2006**[38] in Jordan, And an international study done by **Carr et al; 2000** [39] showed that 33% of their cases were diagnosed with T_1 and 40% T_2, on the contrary in a Pakistanian study done by **Ashok Kumar Tanwani; 2009,**[40] showed that tumors more than **5cm** in size were the most common which indicates late presentation, while in our study we noticed a trend following the European and American styles in presentation as observed by **Falkson G. et al;1996,**[41] who displayed that the most common type of breast tumor were **2cm** size. This result illustrates

that the females education and awareness is improving in our country and trying to approximate the western figures, this can be proved by a wide scale study in different centers in the future.

In the correlation between Tumor Grade and receptor positivity we noticed that in the majority of patients the receptor positivity is higher in **Grade II** (**56%**) followed by **Grade I (24%), Table.3**, Cases with **grade I and Grade II** were **40 cases** (**80%)** of total cases (favorable type), The majority of breast cancers in this hospital belong to the moderately and well differentiated type , This may indicate a changing character of breast cancer in this city which should be proved by further studies. While in a regional study done by **Sundquist et al; 2002,**[42] at West bank of Gaza found that **Grade III cases** present in **38%** of postmenopausal patients but the percent will rise to **64%** if young patient were considered , **Alkuraya et al; 2005,**[43] on comparing Saudi and Swiss breast cancer patients found that **Grade III** cases accounted for **65%** in the former and **32%** in the latter, **M. Siddiqui,2000,**[36] **Nemoto et al;1980** (44) and **Koscienly et al;1994**[45] in their studies showed that *"the larger the tumors the higher the histopathological grade"* which makes the cancer less favorable regarding surgical treatment and adjuvant therapy.

In the relation between Tumor histopathology and receptor positivity we noticed that most of breast cancer cases in this hospital were of **I.D.C** type (**62%**) of total cases, ER+ve expression was (**64.51%**) ,Which is higher than PR+ve expression (**35.48%**) in these cases, while **I.L.C** cases represented (**38%**) of the total cases in which ER+ve expression was (**57.89%**) and PR+ve expression was (**42.10%**) of this type, That is close to an international study done by **Abrektsen, 2010**[46] who showed that the **I.D.C** was diagnosed in (**81.53%**) and **I.L.C** in (**6.3%**).

We can also notice in our study that in **I.D.C** the ER+ve value (**62%**) is higher than PR+ve value (**38%**) ,When compared to other studies, we found in a local study done by **N.Alwan 2010,**[47] correlating between ER and PR positivity and female breast cancer stage showed that ER+ve tumors were diagnosed in **166** cases (**65.1%)** while PR+ve cases were **115** cases (**45.1%)** in stage II disease, these figures were a little higher than our study results and this depends on the histopathologist technique and experience in differentiating between suspicious positive expression and clear positive expression also the comparative results were done in a different center with a higher number of cases in Alwan's study.

An international study carried out by **S.Taucher et al; 2003,**[48] in which cases collected from pre-chemotherapeutic female patients with breast cancer showed from total of **325** cases ER+ve tumor content was **80%,** PR+ve **72%** and this is quite higher figure than our results which can also be explained by the fact that the difference in hormone receptor content of breast cancer could be attributable to the different centers and techniques in various institutes, And in Iraq we need to perform further wider scale multicentre study to know the actual national figures.

This study is done for the first time in **Al-Yarmouk teaching Hospital**, that is why we found only one local study with different methodology.

The benefit of biomarkers contents in breast cancer cases in such a study is the importance of receptor positivity in the Immune therapy, as adjuvant therapy, as for women with stage IV breast cancer; however, women with hormone receptor–negative cancers with symptomatic visceral metastasis or with hormone-refractory cancer may receive systemic chemotherapy.[49]

These benefits are limited to patients whose tumors express ER, PR, or both. A newer adjuvant therapy option for postmenopausal women consists of aromatase inhibitor therapy: these drugs function by inhibiting the peripheral conversion of androgens to estrogen, thereby lowering estrogen levels.[50]

Conclusion

The age incidence of breast cancer patients in our study is the same as international figures the fifth and sixth decades and not as it used to be one decade younger.

Tumor grading is decreasing in this hospital where the majority were grade II and grade I (favorable type), while other regional and international studies previously showed grade II and grade III (unfavorable type) in the majority of cases.

In the vast majority of the cases the tumor size is in the favorable group (5cm or less) and this also indicates changing trend towards patient's early consultation and awareness of the disease.

The major histopathological type in this study is I.D.C which goes with the local, regional and international figures but with higher I.L.C percentage.

Our results for ER and PR contents in breast cancer cells in Yarmouk teaching hospital can be used as a local reference for further studies.

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