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Original Research Article

Study on Quick Score of Hormone Receptor Status of Breast Carcinoma

Siva Jyothsna S.[1]

[1]Assistant Professor, Department of Pathology, NRI Institute of Medical Sciences, Sangivalasa, Visakhapatnam, Andhra Pradesh 531162, India.

Abstract

Introduction: Breast acer is a common cancer in Indian population. The assessment of estrogen receptor (ER) status of breast carcinoma is a routine practice to predict the likely outcome of endocrine therapy. The assessment of progesterone receptor (PR) status along with ER gives a stronger predictive power. The present study was undertaken to look at the prevalence of hormone receptor positive breast carcinomas and to correlate the ER/PR status by Quick score with various tumor characteristics in our local population.

Aim of the study: To assess the value of the Quick Score in terms of correlation with the other proven clinicopathological prognostic parameters of breast carcinoma and to document the prevalence of hormone receptor-positive breast carcinomas in patients attending Government General Hospital, Kakinada, Andhra Padesh.

Materials and Methods: Total 122 cases were studied over a two year period and included 109 MRM and 13 simple mastectomy specimens. Routine hematoxylin and eosin stain and immunohistochemistry for estrogen and progesterone were done on these 122 tissue specimens. Patient age, size, histologic type and grade of the tumor, lymph node status, the ER/PR status with Quick score and the Nottingham prognostic index were studied.

Results: Most of the patients with carcinoma breast were in the 40-60 year age group. Most of the tumors were of size 2-5 cm, histologically were invasive ductal type and also most were of intermediate grade. ER and PR both were negative in 65.5% cases and both were positive in 23.7% cases.

Conclusion: Breast cancers in India show low hormone receptor expression and tend to occur in younger age group. The grade of tumor and NPI showed significant inverse correlation with Quick score of hormone receptor expression. In the present study, the overall grade of the tumor correlated well not only with the proportion of cells stained but also the intensity of staining, the two components of the Quick score that justify the assessment of staining in giving score.

Keywords: Quick Score; ER and PR Immunohistochemistry; Carcinoma Breast in India.

Corresponding Author:

Siva Jyothsna S., Assistant Professor, Department of Pathology, NRI Institute of Medical Sciences, Sangivalasa, Visakhapatnam, Andhra Pradesh 531162, India.

E-mail: jskuro76@gmail.com

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Introduction

Breast cancer is a multifaceted disease comprising of distinct biological subtypes with diverse natural history, presenting a varied spectrum of clinical, pathologicand molecular features with different prognostic and therapeutic implications [1]. Over the last few decades there have been outstanding advances in breast cancer management leading to earlier detection of disease.

The assessment of estrogen receptor (ER) status of breast carcinoma has become the routine practice to predict the likely outcome of endocrine therapy. The assessment of progesterone receptor (PR) status along with ER gives a stronger predictive power [2]. Hormone receptor status of breast carcinoma was earlier assessed by biochemical methods.

The immunohistochemical (IHC) assessment has replaced biochemical assay of hormone receptor status, which can even be done on tiny biopsies. IHC assessment permits visualization of the tumor cells expressing hormone receptors and gives the opportunity to semi-quantify against the cells that are negative for hormone receptors. IHC assessment is considerably less expensive compared to the biochemical assay and can be done on routinely processed tissue sections [3]. With the widespread use of IHC assay, the need for effective quality assurance program has been emphasized. There is no worldwide accepted general agreement on a scoring system for the immunohistochemical evaluation of hormone receptor status of breast carcinoma. The two main features evaluated are the proportion of stained cells and the intensity of the staining. There are several different methods to evaluate the staining like the Allred score (Quick Score), H score, and the proportion of positive cells [4,5]. The present study was undertaken to look at the prevalence of hormone receptor positive breast carcinomas and to correlate the ER/PR status by Quick score with various tumor characteristics in our local population.

Aims and Objectives

1. To assess the value of the Quick Score in terms of correlation with the other proven clinicopathological prognostic parameters of breast carcinoma
2. To document the prevalence of hormone receptor-positive breast carcinomas in patients attending Government General Hospital, Kakinada, Andhra Pradesh.

Materials and Methods

No ethical issues were involved in the study. We conducted the study for two years period from July 2009 to June 2011. We received a total of 157 cases which included edge biopsy, trucut biopsy, lumpectomy, simple mastectomy and modified radical mastectomy (MRM) specimens. Only simple mastectomies and MRM specimens were included in the study. All the specimens were fixed in neutral formalin for sufficient period.

Tumor size was assessed during the grossing of specimen after sufficient fixation. Appropriate tumor tissue and all the lymphnodes identified were processed and subjected to hematoxylin and eosin (H&E) staining.

Tumor type was categorized. Tumor grade was assessed on H & E stained slides using the Nottingham Grading system. This grading system includes 3 components – tubule formation, nuclear pleomorphism and mitotic count. Each variable was given a score 1 to 3 and the scores were added to produce the grade.

Tubule formation and nuclear pleomorphism were assessed by interpreting 3 to 5 blocks of tumor tissue according to tumor size.

Mitotic score was determined by the number of mitotic figures found in 10 consecutive highpower fields in the most mitotically active part of the tumor.

Histological grade was assigned as per the Nottingham grading system.

The Nottingham Prognostic Index (NPI) was calculated for each tumor [NPI = 0.2 tumor size in cm + lymph node status (1, 2, 3) + histological grade (1, 2, 3)]. The NPI was also categorized into three prognostic groups (<3.4, 3.4-5.4, >5.4).

Appropriate block containing adequate amount of tumor tissue and internal controls were selected for Immunohistochemical staining. IHC was performed using BIOGENEX automatic staining system (i6000) of Biogenex Laboratories Inc; CA, USA.

Reagents used for ER:Anti Human Estrogen receptor (AM-272-2ME), Clone:ID5, Species:mouse, Proteinconc: 50mg/ml.

Reagents used for PR: Anti Human Progesterone receptor (AM328-5ME), Clone:PR88, Species: mouse, Protein conc: 10-15mg/ml.

IHC procedure was done as per the technical manual supplied with the instrument.

IHC stained slides were evaluated according to the Quick score which takes proportion of cells stained and intensity of staining into consideration. The staining was evaluated on the invasive component only. Best-preserved and best-stained areas of the sections were assessed.

Nuclear staining was assessed for ER and PR. A score for the proportion of stained cells (0 = no nuclear staining, 1 = <1% nuclear staining, 2 = 1-10% nuclear staining, 3 = 11-33% nuclear staining, 4 = 34-66% nuclear staining and 5 = 67-100% nuclear staining) and the intensity of staining (0 = no staining, 1 = weak staining, 2 = moderate staining, 3 = strong staining) were assigned to each tumour. The score for the proportion of cells stained and the score for the

intensity of staining were added to get the total score, which ranged from 0 to 8.However, a score of 1 was not given as the overall Quick Score is the sum of two scores.

Observations and Results

Total 122 cases were studied over a two year period. The specimens included were 109 MRMand 13 simple mastectomies.

Most of the patients were in the age group of 40-60 years.

Tumor size: Tumor size was assessed during the grossing and most of the tumors were in the range of 2-5 cm.

Tumor grade: Tumor grade was assessed by Nottingham grading and most of the tumors were in the intermediate grade.

Lymphnode status: Lymph nodes were received only in 110 cases out of 122 cases. Metastases were identified and scoring was done. The nodes were negative in 35 (31.8%) cases and were scored as 1. Less than 3 nodes were positive in 45 (40.9%) cases and more than 3 nodes were positive in 30 (27.3) cases and were scored as 2 and 3 respectively.

Type of cancer: There were 108 (88.5%) cases of invasive ductal carcinoma, 2 cases of Lobular carcinoma, 6 cases of Medullary carcinoma and 3 cases each of Papillary and Metaplastic carcinoma.

Table 1: ER and PR status (n=122)

ER+/PR+	ER+/PR-	ER-/PR+	ER-/PR-	ER+	ER-	PR+	PR-
29 (23.7%)	7 (5.7%)	6 (4.9%)	80 (65.5%)	36 (29.5%)	86 (70.5%)	35 (28.7%)	87 (71.3%)

Table 2: Quick score for ER and PR

Quick Score	ER		PR	
	Frequency	Percent	Frequency	Percent
0	86	70.5%	87	71.3%
2	0	0%	0	0%
3	7	5.7%	5	4.1%
4	8	6.5%	6	4.9%
5	15	12.3%	12	9.8%
6	3	2.4%	6	4.9%
7	2	1.6%	4	3.3%
8	1	0.8%	2	1.6%
Total	122	100%	122	100%

Table 3: Age and ER/PR status

Age(Years)	ER+/PR+(n-29)	ER+/PR- (n-7)	ER-/PR+ (n-6)	ER-/PR- (n-80)
21-30	3	0	1	5
31-40	7	0	3	18
41-50	10	4	1	27
51-60	5	3	1	25
61-70	3	0	0	2
71-80	1	0	0	3

Table 4: Age with Quick score of ER and PR

ER	Age	0 (n=86)	2 (n=0)	3 (n=7)	4 (n=8)	5 (n=15)	6 (n=3)	7 (n=2)	8 (n=1)
	21-30	6	0	0	1	2	0	0	0
	31-40	21	0	2	0	3	1	1	0
	41-50	28	0	2	5	6	1	0	0
	51-60	26	0	3	1	3	1	0	0
	61-70	2	0	0	1	0	0	1	1
	71-80	3	0	0	0	1	0	0	0
PR	**Age**	**0 (n=87)**	**2 (n=0)**	**3 (n=5)**	**4 (n=6)**	**5 (n=12)**	**6 (n=6)**	**7 (n=4)**	**8 (n=2)**
	21-30	5	0	0	0	2	1	1	0
	31-40	18	0	2	2	2	1	2	1
	41-50	31	0	0	2	6	2	1	0
	51-60	28	0	3	1	1	1	0	0
	61-70	2	0	0	1	1	0	0	1
	71-80	3	0	0	0	0	1	0	0

Table 5: Age with proportion and intensity of cells stained for ER

Age	Proportion of cells stained for ER						Intensity of staining for ER			
	0 (n=86)	1 (n=0)	2 (n=22)	3 (n=9)	4 (n=4)	5 (n=1)	0 (n=86)	1 (n=10)	2 (n=14)	3 (n=12)
21-30	6	0	2	1	0	0	6	0	2	1
31-40	21	0	7	1	1	0	21	2	1	4
41-50	28	0	10	4	0	1	28	3	7	4
51-60	26	0	2	2	1	0	26	4	3	1
61-70	2	0	1	1	2	0	2	1	1	2
71-80	3	0	0	0	0	0	3	0	0	0

Table 6: Age with Proportion and intensity of cells stained for PR

Age	Proportion of cells stained for PR						Intensity of staining for PR			
	0 (n=87)	1 (n=1)	2 (n=16)	3 (n=10)	4 (n=6)	5 (n=2)	0 (n=87)	1 (n=7)	2 (n=9)	3 (n=19)
21-30	6	0	2	1	1	0	6	0	0	4
31-40	21	0	6	1	2	1	21	3	2	6
41-50	28	0	4	6	1	0	28	1	2	6
51-60	26	1	3	1	1	0	26	2	3	1
61-70	2	0	1	1	0	1	2	1	1	2
71-80	3	0	0	0	1	0	3	0	1	0

Table 7: Tumor size with ER/PR status

Tumor size(cm)	ER+/PR+(n=29)	ER+/PR-(n=7)	ER-/PR+(n=6)	ER-/PR-(n=80)
<2 cm	0	0	1	0
2 – 5cm	21	7	4	62
>5 cm	8	0	1	18

Table 8 Tumor size with Quick score of ER and PR

ER	Tumor size(cm)	0 (n=86)	2 (n=0)	3 (n=7)	4 (n=8)	5 (n=15)	6 (n=3)	7 (n=2)	8 (n=1)
	<2 cm	1	0	0	0	0	0	0	0
	2 – 5cm	66	0	7	6	11	2	2	0
	>5 cm	19	0	0	2	4	1	0	1
PR	**Tumor size (cm)**	**0 (n=87)**	**2 (n=0)**	**3 (n=5)**	**4 (n=6)**	**5 (n=12)**	**6 (n=6)**	**7 (n=4)**	**8 (n=2)**
	<2 cm	0	0	0	0	1	0	0	0
	2 – 5cm	69	0	5	4	7	6	2	1
	>5 cm	18	0	0	2	4	0	2	1

Lymphnode status:

Lymphnodes were received along with specimen in only 110 cases.

Table 9: Lymph node status with ER/PR Status

Lymph node status	ER+/PR+(n-25)	ER+/PR- (n-6)	ER-/PR+ (n-5)	ER-/PR- (n-74)
No positive nodes	6	5	2	22
<3 positive nodes	7	0	2	36
>3 positive nodes	12	1	1	16

Table 10: Lymph node status with Quick score of ER and PR

ER	Lymph node status	0 (n=79)	2 (n=0)	3 (n=6)	4 (n=7)	5 (n=12)	6 (n=3)	7 (n=2)	8 (n=0)
	No positive nodes	24	0	5	2	4	0	1	0
	<3 positive nodes	38	0	0	2	2	1		0
	>3 positive nodes	17	0	1	3	6	2	1	
PR	**Lymph node status**	**0 (n=80)**	**2 (n=0)**	**3 (n=4)**	**4 (n=5)**	**5 (n=10)**	**6 (n=6)**	**7 (n=4)**	**8 (n=1)**
	No positive nodes	27	0	0	1	4	2	1	0
	<3 positive nodes	36	0	3	1	2	2	0	0
	>3 positive nodes	17	0	1	3	4	2	3	1

Tumor grade:

Table 11: Tumor grade with ER/PR status

Tumor grade	Total n=122	ER+/PR+(n-29)	ER+/PR- (n-7)	ER-/PR+ (n-6)	ER-/PR- (n-80)
Low	46 (37.7%)	21	4	4	17
Intermediate	66 (54.0%)	7	2	2	55
High	10 (8.1%)	1	1	0	8

Table 12: Tumor grade with Quick score of ERand PR

ER	Tumor grade	0 (n=86)	2 (n=0)	3 (n=7)	4 (n=8)	5 (n=15)	6 (n=3)	7 (n=2)	8 (n=1)
	Low	21	0	4	5	12	2	2	0
	Intermediate	57	0	3	2	3	0	0	1
	High	8	0	0	1	0	1	0	0
PR	**Tumor grade**	**0 (n=87)**	**2 (n=0)**	**3 (n=5)**	**4 (n=6)**	**5 (n=12)**	**6 (n=6)**	**7 (n=4)**	**8 (n=2)**
	Low	21	0	3	5	10	3	4	0
	Intermediate	57	0	2	1	1	3	0	2
	High	9	0	0	0	1	0	0	0

Nottingham Prognostic Index:

Table 13: NPI with ER/PR status

NPI	Total n=110	ER+/PR+(n-25)	ER+/PR- (n-6)	ER-/PR+ (n-5)	ER-/PR- (n-74)
<3.4	14 (12.7%)	5	2	1	6
3.4 – 5.4	81 (73.6%)	18	3	3	57
>5.4	15 (13.6%)	2	1	1	11

Most of the tumors in our study had intermediate NPI score.

Table 14: NPI with Quick score of ER and PR

ER	NPI	0 (n=79)	2 (n=0)	3 (n=6)	4 (n=7)	5 (n=12)	6 (n=3)	7 (n=2)	8 (n=0)
	<3.4	7	0	2	7	4	0	1	0
	3.4 – 5.4	60	0	4	0	5	3	1	0
	>5.4	12	0	0	0	3	0	0	0
PR	**NPI**	**0 (n=80)**	**2 (n=0)**	**3 (n=4)**	**4 (n=5)**	**5 (n=10)**	**6 (n=6)**	**7 (n=4)**	**8 (n=1)**
	<3.4	8	0	0	1	2	1	1	0
	3.4 – 5.4	60	0	4	3	8	5	1	1
	>5.4	12	0	0	1	0	0	2	0

The NPI was inversely proportional to the Quick score of ER and PR.

Discussion

The hormone receptor status of breast carcinoma can predict the response to adjuvant endocrine therapy. The prevalence of hormone receptor-positive breast cancer in Asian countries has been found to be lower than the western world where more than 50% tumors express hormone receptors. Christopher et al. [6]. have documented a prevalence of 76-78% of hormone receptor-positive breast cancers in the United States from 1992 to 1998 with a rise in the prevalence over the years. In our study, the ER and PR were positive in only 23.7% cases (Table 1).

Majority of the patients in present study were within 31-50 years (57.4%) (Table 6). In a study by Shet et al. [7] they also encountered similar observation(60%) in this age group.

In the present study, most of the cases had tumor size between 2-5cm (77%) and 68% of the cases had lymphnode positivity and 22.1% presented with tumor size>5cm.Other Indian study by Shetet al. [7] where they encountered 39% of patients presenting with tumor size of 5cm. Similar results were obtained by Mudduwa et al. [5] in their study

in Srilankan population (74% had 2-5cm size). This probably could be due to absence of National breast screening program in our country.

The majority (54.1%) of the patients included in our study had intermediate grade tumors (Table 12). Among the three parameters taken to determine the grade,we found the majority of tumors had higher score for tubule formation and nuclear pleomorphism,but did not show significant mitotic activity which contributed to lowering the grade on the whole.The low mitotic activity in our study could be due to prolonged ischemic interval and improper fixation which leads to loss of mitotic figures.

The NPI was between 3.4-5.4 in majority (73.6%) of our cases (Table 13).This correlated with the findings of Mudduwa et al. [5]. Majority of carcinomas in our study were Invasive duct cell carcinoma, which constituted 88.5%

In our study we found four categories with respect to ER and PR expression (Table 1).

1. 65% cases were negative for ER and PR.
2. 23.7% were positive for both ER and PR.
3. 5.7% of cases showed only ER positivity.
4. 4.9% showed only PR positivity.

The number of studies performed on this topic of ER and PR expression is much less in the Asian communities compared with the Western world. Desai et al. [8] have documented a prevalence of 32.6% for ER-positive and 46.1% for PR-positive breast cancers.Our study documents a prevalence of 29.5% for ERpositive and 28.7% for PR positive breast cancers (Table 1). It indicates that the majority of the breast carcinomas in the study sample would not respond to endocrine therapy.

The causes for high proportion of dual negativity in our study may be related to preanalytical and analytical factors such as prolonged ischemic interval, improper fixation or inefficient antigen retrieval. This could be one of the causes for low positivity. We have used sodium citrate for antigen retrieval whilst study shows that EDTA is superior antigen retrieval solution irrespective of heating method that was used.We used the microwave based EZ retriever system V2.1 of Biogenex for antigen retrieval whilst it has been found that pressure cooking is more convenient and cost effective method.

Clubbing the ER and PR positive tumors with only ER positive tumors,the total ER positivity in our study was 29.5% compared to 50.5% expression by Shetet al. [7] in their study.

When the two components of Quick score were applied for interpreting ER and PR positivity,we found that proportion of cells stained was similar for both ER and PR, but intensity of staining was weak with ER compared to PR (Tables 2, 5 and 6). We used the monoclonal antibody provided by Biogenex for assessing both ER and PR.We could not find cause for this difference in intensity of staining.

We also encountered 4.9% of only PR positive tumors. In their study of 1944 cases, Rakha et al. [9] found a small proportion of cases (3.4%) to be ER negative and PR positive. Shet et al. [7] also reported occurrence of similar hormonal status in their study of 11,780 cases. Both the studies commented that such tumors occurred in a younger age group and were of higher grade. In the present study, we found most of the cases (3/6,50%) occurring in 31-40 years age group but majority of the cases (4/6,66%) were of lower grade unlike that reported in the earlier studies. The high proportion of intermediate grade tumors in our study could possibly explain this discrepancy in PR positivity.

Correlating the Quick score values of ER and PR with other parameters like age (Tables 3 and 4), size of tumor (Tables 7 and 8), lymphnode status (Tables 9 and 10), grade (Tables 11 and 12), and NPI (Tables 13 and 14) we found that both the components of Quick score correlated well with tumor grade and NPI for ER while for PR both the components correlated well with the grade of the tumor only. With PR positivity only the proportion of stained cells correlated with the NPI whilst intensity of staining did not. Other parameters like age, tumor size and lymphnode status did not show correlation with overall Quickscore for both ER and PR. This is similar to findings obtained by Mudduwa et al. [5] in their study.

The majority (77%) presented with a size between 2 and 5 cm and 22% presented with a size > 5cm which is similar to other Asian studies. The majority (68%) of patients included in the present study had lymph node metastasis by the time they sought treatment. Hence, the majority do not present in the early stage in our country.

Table 15: Comparison of receptor positivity with other Asian studies

Study	ER positivity	PR positivity
Shet et al[7]	52.4%	45.2%
Desai et al[8]	32.6%	46.1%
Mudduwa et al[5]	45.75	48.3%
Azizun-Nisa et al[10]	32.7%	25.3%
Present study	29.5%	28.7%

The majority (54.1%) of patients included in our study had intermediate- grade tumors, which explains the low prevalence of hormone receptor expression. It is often observed that high-grade tumors tend to be negative for hormone receptor expression. The present study shows more of intermediate grade tumors with low mitotic activity seen in the tumor tissue which may be due to prolonged ischemic interval between surgical resection and fixation causing necrosis of the tissue and cessation of mitotic activity.

In the present study, the overall grade of the tumor correlated well not only with the proportion of cells stained but also the intensity of staining, the two components of the Quick Score that justify the assessment of the intensity of staining in giving a score.

The NPI was between 3.4 and 5.4 in the majorityof cases. The NPI correlated well with the hormone receptor status when the cut-off point was 2. The European Working Group for Breast Screening Pathology [11] found that there was a poor concordance between observers in assigning a score to tumors except in very strongly positive or completely negative tumors. Therefore, a Quick score of 0 and 7-8 can be relied upon for therapy. The same study concluded that technical and interpretative variations were responsible for the variation in the assessment of the Quick Score in low and moderate levels of hormone receptor expression [11].

A significant proportion of the present study sample (12.2% for ER and 9% for PR) expressed low levels of receptors according to the subcategories suggested by the European Working Group for Breast Screening Pathology [11] (Quick Score 2-4). The Quick Score of this group of tumors could have probably been subjected to variation. Therefore, inclusion of a weakly positive control to the routine staining procedure is suggested to reduce the technical variation. Because the author had strictly adhered to the guidelines given by the United Kingdom National Health Service Breast Screening Programme in assigning a score, interpretative variation was expected to be minimal in this study. Inter-observer variation was eliminated at the beginning.

Conclusion

In our study of 157 cases, 122 cases were analyzed for hormonal receptor status which revealed a high incidence of hormone negative tumors. This is in par with other studies which show that breast cancers in India show low hormone receptor expression and tend to occur in younger age group. The grade of tumor and NPI showed significant inverse correlation with Quick score of hormone receptor expression.

In the present study, the overall grade of the tumor correlated well not only with the proportion of cells stained but also the intensity of staining, the two components of the Quick score that justify the assessment of staining in giving score.

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