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Association between Immunohistochemical Profile and Clinicopathological Appearance in Breast Cancer: A 7-year Review from Hamadan, Iran

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Abstract

Introduction: Invasive breast cancer is the most common carcinoma in women. Immunohistochemistry classification now plays a key role in prognostic identification and prediction of outcome in this disease. Based on recent gene expression studies, immunohistochemical subtypes are as follows: Luminal A (ER+ and /or PR+, HER2-), luminal B (ER+ and /or PR+, HER2+), HER2+/ ER-, PR-, and basal-like (ER-, PR-, HER2-). These molecular differences have been shown to correlate with clinical features, such as survival, and sensitivity to treatment. In this study we evaluated the association between different subtypes with histological type, grade, tumor stage, lymph node positive ratio, lymph node status, recurrence, and survival.

Patients and methods: We retrieved the clinical records of 580 patients with breast cancer who were treated at Mahdieh Institute of oncology in Hamadan, Iran, between Oct 2004 and Oct 2011, and we evaluated clinicopathological data of these patients.

Results: mean age of patients was 47.22 ± 11.1 years. Of 573 patients, 116 (20.2%) were ER /PR+, HER2+, 257 (44.9%) were ER /PR+, HER2- , 72 (12.6%) were ER /PR- , HER2+ , 124 (21.6%) were ER /PR- , HER2- and 4 were undefined. The estimated median follow up period for all subjects was 4.9 years (range 3 months to 6.9 years). The overall survival for all patients was 88.21% and the disease free survival was 83.7%. The interesting result of this study was the lower incidence of positive axillary lymph nodes in triple negative subtypes. Five-year relative survival rates were higher for patients with ER/PR+ and negative lymph nodes ($p< 0.05$).

Conclusion: This study highlighted the importance of immunohistochemical subtypes. As our patients were good representatives of breast cancer in western Iran and this study showed some differences with literature, further research should be directed at standardization of molecular and immunohistochemical methods in our country.

Keywords: Breast Cancer, chemotherapy, survival, Immunohistochemistry, IHC.

Introduction

Invasive breast cancer is the most common cancer among Iranian women, accounting for 22% of all female malignancies. Breast cancer is a heterogeneous malignancy, no longer seen as a single disease, but rather as a multifaceted disease. Over the last decade, outstanding advances have been achieved in the early detection and treatment of breast cancer, resulting in significant decrease in mortality, as well as improved outcomes [1, 2]. Histological type, grade, tumor size, lymph node involvement, estrogen receptor (ER), progesterone receptor (PR), and HER2 receptor status all affect the prognosis and the probability of response to systemic therapies.

Recent attention has been directed at molecular classification of breast cancer. Nowadays immunohistochemistry classification plays a key role in prognostic identification and prediction of outcome. Estrogen receptor (ER) positivity predicts the response to endocrine treatment such as antiestrogen (tamoxifen) therapy or ovarian suppression. Similarly, human epidermal growth

factor receptor 2 (HER2) positivity is useful for selecting patients for targeted therapy with monoclonal antibody (trastuzumab) against HER2 [3]. Based on recent gene expression studies, Carey et al. updated the definition of immunohistochemical subtypes as Luminal A (ER+ and /or PR+, HER2-), luminal B (ER+ and /or PR+, HER2+), HER2+/ ER-, PR-, and basal-like (ER, PR-, HER2-). These molecular differences have been shown to correlate with clinical features, such as survival, and sensitivity to treatment [4]. For example, the basal-like subtype accounts for about 15% of breast cancer cases and is associated with an aggressive histology, poor prognosis, and unresponsiveness to endocrine therapies [5]. This study reports our attempt to sub-classify breast carcinomas according to specific immunoprofiles, and to evaluate the association between different subtypes with histological type, grade, tumor stage, lymph node positive ratio, lymph node status, recurrence, and survival.

Patients and methods

We retrieved the clinical records of patients who were diagnosed with breast cancer between Oct 2004 and Oct 2011 at Mahdieh Institute of Oncology in Hamadan, Iran. Patients with bilateral breast cancer, those with a history of another primary cancer, and those who had received neoadjuvant chemotherapy were excluded. Clinicopathological data, including age, size, grade, histology, lympho-vascular invasion, perineural invasion, axillary lymph node status, hormone receptor and HER2 status, P53 index, loco-regional recurrence, distant metastases, disease free survival (DFS), and overall survival (OS) were recorded in a database.

Patients had been treated with either mastectomy or breast conservation surgery followed by -if indicated- systemic adjuvant chemotherapy, external beam radiotherapy, and endocrine therapy (for endocrine responsive tumors). The histological classification was based on the criteria set by World Health Organization.

Table 1. Baseline characteristics of the patients

Patient's characteristics	No of patients (%)
T stage	
T1	12.5
T2	56.6
T3	23.8
T4	7.1
N stage	
N0	29
N1	42.5
N2	17.4
N3	11
Cancer type	
Invasive Ductal	87.6
Invasive Lobular	5.3
Ductal and Lobular	2.7
Medullary	2.7
DCIS	1.7
Surgery	
Mastectomy	76.7
Breast conserving	23.3
Radiotherapy	90.1
Chemotherapy (adjuvant)	95.98
Hormone replacement therapy	65.1
Tumor subtypes	
ER/PR+ , HER2 +	20.2
ER/PR+ , HER2-	44.9
ER/PR- , HER2+	12.6
ER/PR- , HER2-	21.6

Survival time was calculated from the date of surgery until time of death or confirmation of survival at the end of the observed interval. For the evaluation of ER/PR, the cut off positivity was 10% tumor cells. A positive HER2- stain was determined by membranous staining of tumor cells equal to 3+ or fluorescence insitu hybridization (FISH) with more than two copies of the HER2 gene. T and N at diagnosis were coded according to the American Joint Commission on Cancer (AJCC) staging, 6th edition [6].

Results

The final analysis included 573 patients with breast cancer. Mean age of the patients was 47.22±11.1 years (23-93 years). Baseline characteristics of patients, including tumor subtypes, are presented in table 1. Of 573 patients, 116 (20.2%) were ER /PR+, HER2+, 257 (44.9%) were ER /PR+, HER2- , 72 (12.6%) were ER /PR- , HER2+, and 124 (21.6%) were ER /PR- , HER2- and 4 remained undefined. Differences in baseline characteristics between the four subtypes are presented in table 2.

Patients with ER/ PR+ , HER2- subtypes were more likely to have perineural invasion (30.3%), and patients with positive ER/PR and HER2 showed more lymphovascular invasion ($p<0.05$). Ninety-three patients (16.2%) developed recurrence; of these, 77 (13.4%) had distant recurrence, 15 (2.6%) local recurrence and 1 (0.17%) both local and distant recurrence. Table 3 shows sites of recurrence in association with different subtypes.

The estimated median follow up period for all patients was 4.9 years (3 months to 6.9 years).

Discussion

Age is an important risk factor for breast cancer. The mean age of all patients in all groups was between 46 and 50 years, which was less than that of other published data. In many regions of the world, especially in developed countries,

Table 2. Baseline characteristic by tumor subtypes

	ER/PR+,HER2+	ER/PR+, HER2-	ER/PR-HER2+,	ER/PR-, HER2-
Age (years±SD)	46.04±11.67	49.5 ± 9.19	47.85 ± 10.58	46.04 ± 11.45
T stage (%)				
T_1	16.7%	17.7%	9.9%	9.6%
T_2	58.3%	51%	60.6%	57.4%
T_3	20.4%	24.5%	16.9%	28.7%
T_4	4.6%	6.8%	12.7%	4.3%
N stage				
N_0	26.7%	29.2%	29.6%	36.3%
N_1	49.1%	43.2%	49.3%	37.9%
N_2	18.1%	16.7%	14.1%	14.5%
N_3	6.1%	10.9%	7%	11.3%
LVSI	40.4%	39.8%	43.5%	40.2%
PNI	26.6%	30.3%	25.4%	25.2%
P_{53}				
+	69.4%	42.7%	51.2%	35.6%
-	30.6%	57.3%	48.8%	64.4%
Cancer type				
Invasive Ductal	93%	86.8%	89.1%	84.4%
Invasive Lobular	2%	6%	4.7%	7.4%
Medullary	1%	2.6%	1.6%	4.1%
DCIS	2%	2%	3%	0%
LCIS	2%	2.6%	1.6%	4.1%
Surgery				
Mastectomy	80%	80.9%	78.5%	89.5%
Breast conserving	20%	19.1%	11.5%	10.5%
Radiotherapy	86.2%	96.4%	95.7%	94.8%
Systemic chemotherapy	93%	86.8%	95.7%	98.3%
Hormone therapy	100%	100%	0%	0%

Table 3: sites of recurrence in association with different subtypes.

	ER/PR+ , HER2+	ER/PR+ , HER2-	ER/PR- , HER2+	ER,PR , HER2-
Metastatic at presentation	7.8%	3.5%	5.6%	1.6%
Local recurrence	2.7%	2.04%	5.7%	3.3%
Distant recurrence	18.5%	7.3%	18.8%	21.4%
Liver	15%	11.1%	23%	38.4%
Brain	15%	22.2%	30.7%	30.7%
Lung	10%	22.2%	15.38%	38.4%
Bone	70%	66.6%	38.4%	30.76%
Time to recurrence (month)	27.83 ± 18.24	26.35 ± 22	24.53 ± 20.3	25 ± 20.35

the maximum disease incidence is around 65-79 years or above 60 years [7]. A possible explanation of this result –apart from racial differences- could be the younger age of menopausal onset, or over-treatment with hormonal drugs like OCPs and estrogens, in Iranian women. Patients with ER/PR+, HER2- subtypes were more likely to be older ($p<0.01$).

Tumor stage plays an important role in prognosis. Advanced tumor stage is directly related to an increasing probability of local recurrence and death. Subjects with ER/PR-, HER2+ subtypes were more likely to be diagnosed with T4 stage ($p=0.05$).

A high percent of patients presented with axillary lymph node involvement at the time of diagnosis (ranged from 63.7% to 73.3% in different subgroups). An interesting result from this study was the lower incidence of positive axillary lymph nodes in triple negative patients, which was in contrast to the majority of previously published data [8, 9, 10]. ER/PR+ cases with involved axillary lymph nodes showed a lower incidence of distant metastases in contrast to ER/PR- cases ($p<0.05$).

This demonstrated that lymph node positivity had a more significant prognostic value in triple negative patients than in other subtypes.

Over expression of p53 has been correlated with a higher likelihood of recurrence and poorer prognosis in the patients. Different studies have reported p53 to be positive in up to 70% of the patients [11-12]. In the present study, p53 positivity was more prevalent in ER/PR+, HER2+ patients (69.4%, $p< 0.05$) than in other subtypes.

The basal-like breast cancers are ER/PR- and HER2- negative (sometimes called triple-negative). They account for about 15% of all breast cancer patients and are associated with shorter survival [13]. In this study, 21.6% of all cases were triple negative. Distant recurrence, especially in liver, lung, and brain, presented more commonly in the basal-like subtype.

The time to recurrence differed between the four subtypes; however, the difference was relatively small.

Five-year survival rates were higher for patients with ER/PR+ and negative lymph nodes ($p< 0.05$). Women with ER/PR+ breast cancer typically received hormonal therapy (tamoxifen or letrozole) in 100% of cases; but trastuzumab therapy was not prescribed to most HER2 positive subtypes because of the cost; this issue makes the interpretation of survival and disease-free survival rates difficult. Our study showed that ER/PR+ subtypes were the major subtypes of breast carcinoma, with better survival and disease-free survival rates than other subtypes.

Conclusion

This study highlighted the importance of immunohistochemical subtypes. As our patients were good representatives of breast cancer in western Iran and this study showed some differences with literature. Further research should be directed at standardization of molecular and immunohistochemical methods in our country.

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