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Breast cancer classification according to immunohistochemistry markers: subtypes and association with recurrence in an oncology hospital database, Baghdad.

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Abstract:

BACKGROUND: Breast cancer is comprised of a series of complex and heterogeneous subtypes with difference in clinical behavior and outcomes. The immunohistochemistory defined subtypes have a predictive significance and prognostic value in breast cancer. There are limited data regarding immunohistochemistory defined subtypes among Iraqi breast cancer patients. The objective of this study was to study the prevalence of immunohistochemistory defined subtypes and to identify their associations with the risk of recurrence.
PATIENTS AND METHODS: A study included 150 patients with breast cancer attending Baghdad oncology teaching hospital between June 2019 and December 2019 (50 cases with negative recurrence history and 100 recurrence cases) for whom data including age, stage, grade, histopathological type, date of recurrence for recurrence cases and date of last follow-up for cases with negative recurrence history were collected. The breast cancer subtypes defined using immunohistochemical measures of hormone receptors and human epidermal growth factor receptor 2 and classified into four major subtypes: luminal A, luminal B, HER2-positive, and triple negative. the association between these subtypes and the recurrent history was evaluated by Chi-squared test.
RESULTS: The mean (±SD) age was 48.4 (±10.8) years. The immunohistochemistory defined subtypes of cancer was shown: luminal A in 79(52.7%)patients, 24(16%)patients had luminal B, 15(10%)patients had HER2 positive and 32(21.3%)patients had triple negative breast cancer. there were a significant association between immunohistochemistory defined subtypes and recurrent history (p=0.012).
CONCLUSION: Tumor profiling using molecular subtypes is a promising agent to identify a cases at high risk of recurrence.

Introduction:

Breast cancer is one of the most common cancer type in women and it affects millions of women around the world.1 Breast cancer recently seen as a multifaceted disease, as the cancers with same clinical and pathological appearance may relatively have diverse behaviors.2 The clinical and histological presentation of the cancers may not be enough to determine the underlying complicated genetic alterations and the biological episodes that participated in cancer development and progression.3 There are many subtypes with different biological features that lead to various response to treatment modalities and had different clinical outcomes.3 The distinct and multiples molecular subtypes of breast cancer have been identified using standard immunohistochemical (IHC) markers based on hormone receptor and human epidermal growth factor receptor 2 (HER2) status.4, 5 As the recurrence event are complex and many factors affecting on it, including clinical and histological characteristics of primary cancer and treatment regimes(with/without postoperative adjuvant radiotherapy, chemotherapy and/or hormone therapy).6 A perception of the likelihood of tumor recurrence will support clinical decision making and adequate follow-up. More concern most be given to identify recurrence of tumors in patients with more severe breast cancer subtypes, such as the triple negative subtype, by routine postoperative follow-up. Although the relation between breast cancer molecular subtype and recurrence has been studied in many countries,7-9 limited information is available for Iraqi patients, Therefore, the main objective of the study was to analyze the prevalence

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Of breast cancer subtypes and to determine the association between the molecular subtypes defined by hormone receptors and HER2 and the risk of recurrence.

PATIENTS AND METHODS:

A total 150 histologically confirmed invasive breast cancer patients were attending Baghdad oncology teaching hospital between June 2019 and December 2019 (50 cases with negative recurrence history and 100 recurrence cases) included in this study. The study included cases diagnosed over different periods of time from 1st of January 2012 to 30th of December 2017, with exclusion patients with bilateral breast cancer, patients with denovo metastasis, patients with second primary cancer, inflammatory breast cancer, male patients and patients with carcinoma in situ. The following information were collected: age at diagnosis, tumor characteristics in form of tumor stage, tumor grade, histopathological type, estrogen receptor (ER) and progesterone receptors (PR) status and human epidermal growth factor receptor 2 status (HER2) , date of recurrence for recurrence cases and date of last follow-up for non recurrence cases. Molecular subtype classification was done based on immunohistochemical surrogates for ER, PR and HER2 status and definitions were as follow: Luminal A (ER+ and/or PR+, HER2-), Luminal B (ER+ and/or PR+, HER2+), Triple negative breast cancer (TNBC) (ER-, PR -, HER2-), HER2+ (ER-, PR-, HER2+).4, 10 The recurrence was defined as the reoccurrences of carcinoma either locally, regionally or distally. Statistical analysis was done with IBM SPSS version 23. Categorical variables was introduced as frequency and percentage, while continuous variables was introduced as mean and standard deviation (SD), chi- square test used to find association between four groups of molecular subtype and recurrence history. As appropriate, Kaplan-Meier survival analysis was carried out to estimate mean disease-free survival (DFS) which was the time from the date of the first diagnosis to the date of the recurrence and the group differences in disease free survival time were analysis by a log-rank test. The level of statistical significance was set to <0.05, to reject the null hypothesis.

RESULTS:

A one hundred fifty patients with breast cancer enrolled in this study. The molecular subtypes of cancer was shown that luminal A in 79(52.7%) patients, 24(16%) patients had luminal B, 15(10%) had HER2 positive and 32(21.3%) had TNBC, figure -1-

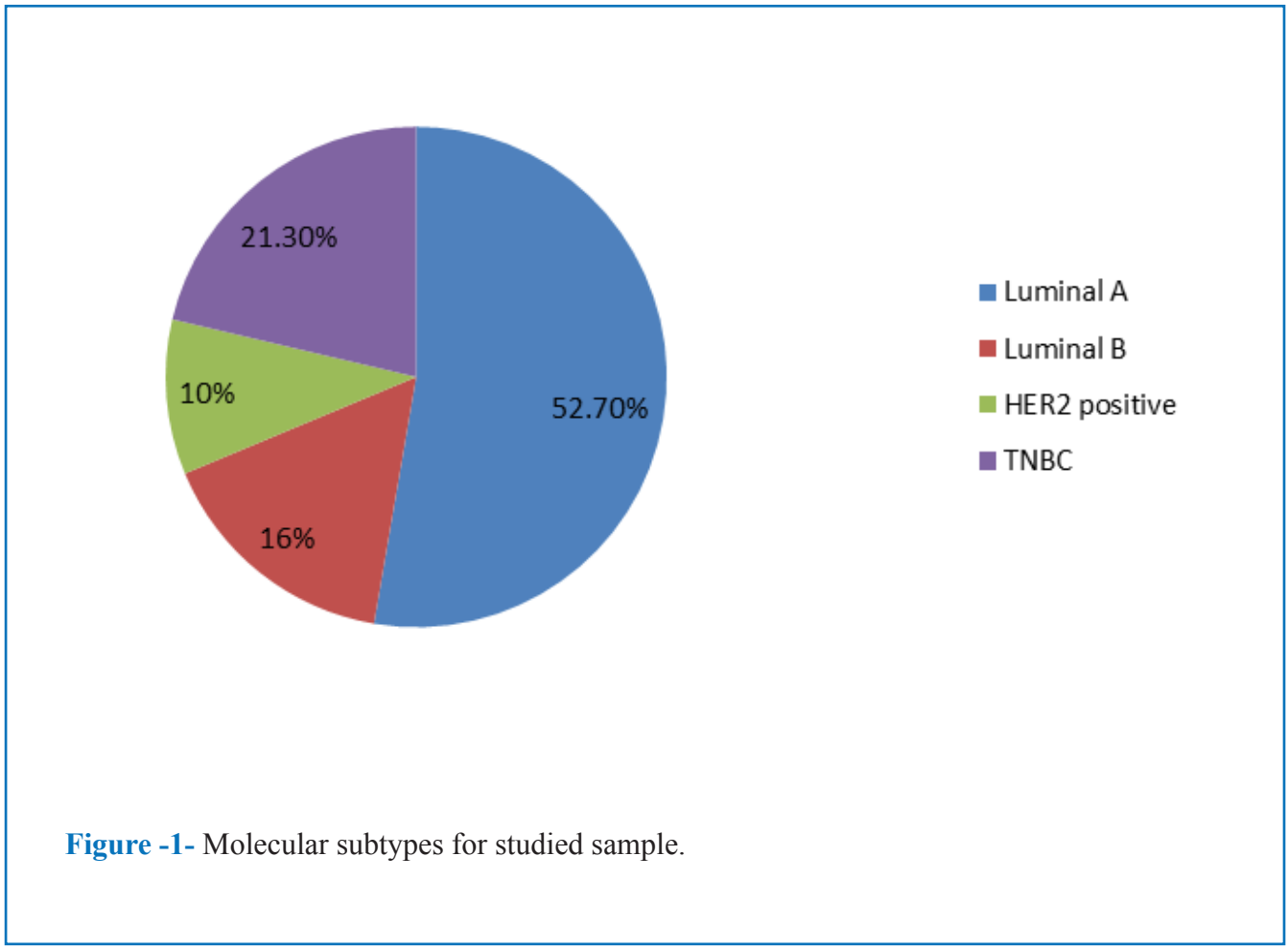

Figure -1- Molecular subtypes for studied sample.

The grade of cancer and molecular subtypes were associated with recurrence history (p=0.028, 0.012 respectively), table -1-.

Table -1- The clinicopathological findings in relation to recurrence history.

Variables			Recurrence history		Total	P value
	Negative	Positive				
Age	Mean (±SD)		48.3(±9.3)	48.5(±11.5)	48.4 (±10.8)	0.95#
Stage		Early	30(60%)	44(44%)	74 (49.3%)	0.065*
Late		20(40%)	56(56%)	76 (50.7%)		
Grade		High	8(18%)	33(33%)	41 (27.3%)	0.028*
Low-intermediate		42(84%)	67(67%)	109 (72.7%)		
Histopathological finding		Invasive carcinoma of NST	43(86%)	81(81%)	124 (82.7%)	0.44*
Others		7(14%)	19(19%)	26 (17.3%)		
Molecular subtypes		Luminal A	32(64%)	47(47%)	79(52.7%)	0.012*
		11(22%)	13(13%)	24(16%)		
Luminal B		3(6%)	12(12%)	15(10%)		
HER2+ve		4(8%)	28(28%)	32(21.35)		
TNBC						

#Student T test, *chi-square test, significant≤0.05. NST= Invasive carcinoma of no special type, TNBC=triple negative breast cancer. HER2+ve= human epidermal growth factor receptor 2 status positive.

The mean(±SD) fallow up period for non-recurrence case was 56.4(±15.3) months with range between24-84 months and the mean (±SD) disease free survival (DFS) for recurrence case was 19.7(±11.9) months with range between 3-60 months. The mean (95% CI) disease free survival for patients with luminal B was the highest and the Mean (95% CI) disease free survival for patients with TNBC was the lowest and the log rank p value was .005, table -2- , figure -2-.

Table -2- The mean disease free survival according to molecular subtypes.

Molecular sub-types	Disease free survival				*P* value
	Mean ((months	Std. Error	Confidence Interval 95%		
			Lower Bound	Upper Bound	
Luminal A	44.6	3.695	37.4	51.9	005.
Luminal B	45.6	5.545	34.7	56.5	
HER2 positive	25.5	6.259	13.2	37.8	
TNBC	25.3	3.021	19.4	31.2	
Overall	40.3	2.590	35.2	45.4	

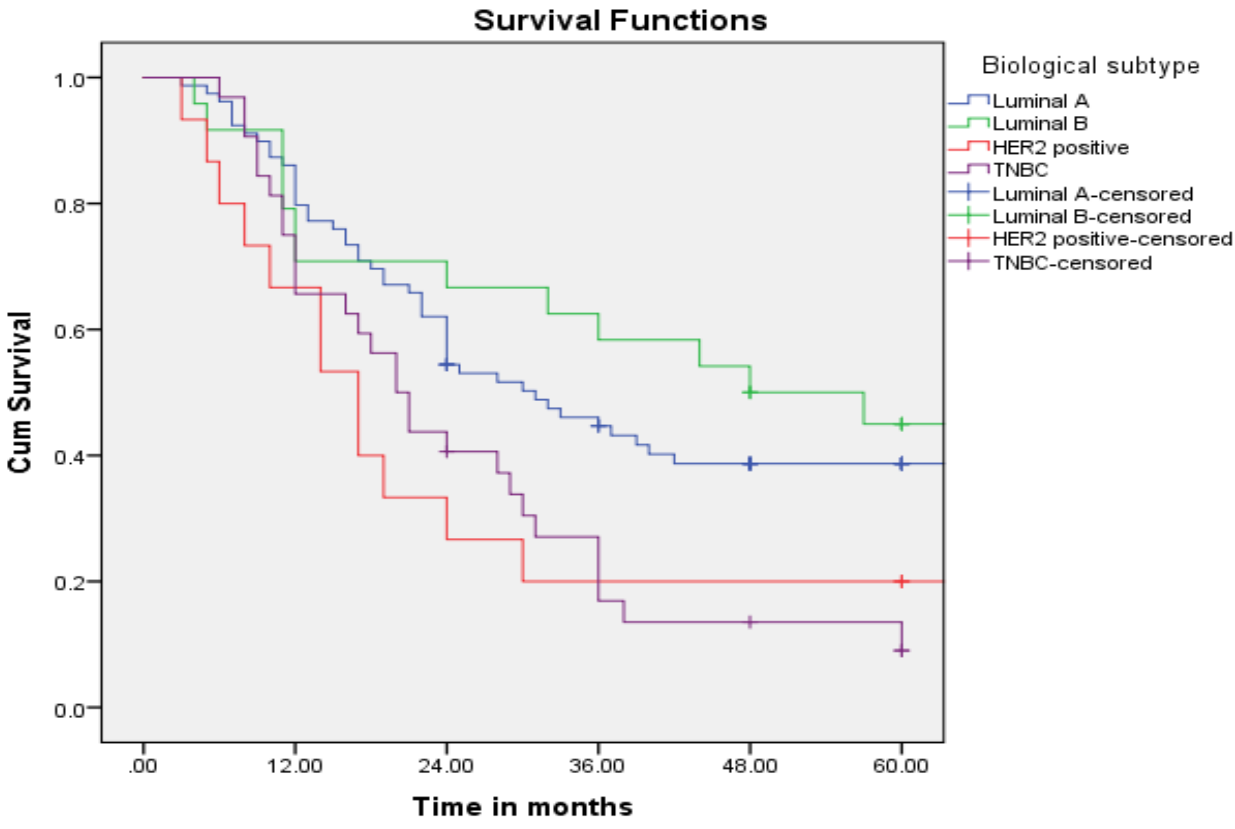

Figure -2- Kaplan- Meier survival analysis according to molecular subtypes.

DISCUSSION:

Worldwide, Breast cancer is the most common cancer in women. In Iraq, Breast cancer ranks first among cancer in women.11 Several molecular subtypes of breast cancer have been determined depending on hormonal receptors and HER2 status, there have been several previous classifications that either classify breast cancer into two subtypes depending on estrogen receptor status only, which has been low informative in terms of outcome, and the additional HER2 status to classification was offers improved and significant therapeutic assistance, others was separating the breast cancer into triple negativity and other12, this way of classification is simplistic and informative but may be misleading by grouping the luminal A, luminal B with HER2 positive. IHC- based classification on both hormonal receptor status and Her2 status provides prognostic and therapeutic information non realizable when depend on one of them alone, our oncology hospital classified the breast cancer based on IHC into four molecular subtypes out of the eight possible molecular subtypes that will emerge if the classification is based on PR expression (ER+/PR+ vs. ER+/PR− tumors) and this classification used commonly in other centers.13 We think that this classification is more practical, easy, informative and clinically useful, this classification became the base for making treatment plan and was implicated in many breast cancer treatment guidelines as in ASCO14 and NCCN.15

This study including of 150 women patients with histologically confirmed invasive breast cancer where the molecular subtypes of breast cancer was evaluated. The most common type in this study was luminal A subtype which represent 52.7% of cases, the second prevalence one was TNBC which represent 21.3% of cases, while the luminal B represent 16% of case and HER2 positive subtype represent 10% of cases. This findings was noticed to be in concordance with findings that revealed from other studies done in different countries in Asian and Western countries like in Saudi Arabia where prevalence of subtype was luminal A 58.5%, TNBC 16% ,luminal B 14% and HER2 positive11.5% ,16 in china the prevalence of subtype was luminal A 46.5%, TNBC 21.5% , luminal B 17% and HER2 positive 15%, 17 in Peru the prevalence of subtype was luminal A 49.3%, TNBC 21.3% , luminal B 13.2% and HER2 positive16.2% ,18 even there were some minor variations in prevalence of molecular subtypes that may be related to social, environmental, and/or genetic factors, racial and/or technological disparity.

In this study the 64% of patients with negative recurrence history had luminal A subtype while 47% of patients with recurrence history had luminal A subtype, the study finding was concordance to other studies, where The favorable prognosis for Luminal A subtype.19-21 In this study the TNBC subtype was in 8% of cases with negative recurrence history while in 28% of cases with recurrence history, this was concordance with previous studies that have revealed triple-negative receptor status is significantly associated with recurrence and bed prognosis.9, 22, 23 Our results revealed a significant association between molecular subtype and disease free survival with the TNBC subtype being the lowest mean of DFS and this in line of studies. 24, 25 Our study had several limitations. It was a single-center retrospective rather than a prospective assessment of consecutive breast cancer patients. Therefore, we cannot exclude selection biases, and caution must be exercised in applying the results to the general population. Other limitation are due to the lack of Ki67, which is the cellular marker that distinguished between luminal A and non-HER2 expressing luminal B tumors.4 Our research sample was relatively small and there was no long-term follow-up, which limits late recurrence of our findings. Finally , the research was carried out on the local population of a certain cultural , ethnic and social history and care must be taken in extending the findings to the global population.

In summary, based on our data, there was an association between molecular subtypes and recurrence of breast cancer, however, the molecular subtypes should be integrated with the other significant typical prognostic variables like age, stage, grade, comorbidity, and use of adjuvant therapy, for each patient with breast cancer.

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