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RESEARCH ARTICLE

Histopathological Patterns and Luminal Subtypes among Breast Cancer Patients in the Middle Euphrates Region of Iraq

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Abstract

Objectives: To evaluate the pathological patterns & luminal subtypes of breast cancer patients in the Middle Euphrates region of Iraq. **Methods:** A retrospective descriptive study was conducted at Al-Hussein Cancer Center in Karbala province of Iraq between January 2012 and August 2020. **Results:** There were 920 female patients with breast cancer. At the time of diagnosis, the mean age was 50.20 ±11.74 years. The most frequent pathological subtype was invasive ductal carcinoma in 91.85%, and 50.32% of tumors were grade II. Of luminal subtypes, Luminal A was the most prevalent subtype in 53.69%. **Conclusion:** Our results were similar to middle eastern patterns but significantly differed from western patterns.

Keywords: Breast Cancer- Histopathological Patterns- Luminal Subtypes- Iraq

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Introduction

Worldwide, breast cancer is the most frequently diagnosed tumor and the leading cause of cancer death [1]. In Iraq, it is the most common cancer in females presenting more than 40% of newly diagnosed cancer cases, making breast cancer management a great challenge to health workers in our country [2]. The main risks for developing breast cancer are: age, family history, sedentary lifestyle, early menstruation, late menopause, exposure to radiation, alcoholism, and hormonal replacement therapy [3].

In fact, breast cancer is a heterogeneous complex of diseases, a spectrum of many subtypes with distinct biological features that lead to differences in response patterns to various treatment modalities and clinical outcomes [4]. ER (Estrogen receptors), PR (progesterone receptors) and HER2 (human epidermal growth factor receptors 2) are recognized as major clinical and histopathological prognostic parameters. Based on their express, four major breast cancer luminal subtypes have been identified [5]. These include: Luminal A (ER/PR positive/HER2 negative), Luminal B or Triple Positive (ER/PR positive/HER2 positive), Basal-like or Triple Negative (ER/PR negative/HER2 negative) and HER2-enriched (ER/PR negative/HER2 positive) [4].

Significant geographic variation in luminal subtypes & histopathological pattern has been proved in several studies [6-8]. This study was designed to identify luminal subtypes of breast cancer patients in our region. Then try to compare our results with other researches from nearby middle eastern countries in addition to other developed and developing countries around the world. Our findings will help provide essential knowledge to understand the pathological distribution and establish potential plans for breast cancer management in Iraq.

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Materials and Methods

Study design and participants

This retrospective descriptive research was carried out in the Al-Hussein Cancer Center in Karbala province of Iraq on female breast cancer patients diagnosed between January 2012 and August 2020. Our center covers not only Karbala peoples but other patients from the Middle Euphrates area in Iraq who are referred to this center for solid and hematological malignancy management [9]. All patients were diagnosed by tru-cut or excisional biopsy and confirmed by immunohistochemistry markers. Complete information about age, histological subtypes and tumor grades were obtained from the pathological department.

Pathological & immunohistochemical assessment

The studied tissue biopsy specimens of the affected patients were embedded in paraffin and fixed in 10% formaldehyde, then cut and stained with Hematoxylin & Eosin for histopathological examination (Figure 1). Following the diagnoses of breast carcinoma, all blocks were assessed regarding the expressions of (ER, PR) and HER2 receptors by semi-quantitative immunohistochemical (IHC) staining technique.

ER and PR contents of the primary tumors were evaluated according to the Allred scoring system, which depends on the percentages of the positive and the staining intensity of tumor cells by using Dako kits ™ (Dako, Denmark), where higher scores indicated the presence and abundance of these receptors. The staining reactions to be considered positive must be reflected in at least 10% of the tumor cells, and the staining intensity was classified as strongly positive (+3), moderately positive (+2) and weak positive (+1) (Figure 2) [10]. Labeled streptavidin-biotin (LSAB) method was employed for immunohistochemical detection of HER-2/neu using Hercep Test Kit (K5204 Dako Co.). The intensity of HER-2/neu cell membrane stain was classified into score 0 (negative; no stain is observed or faint membrane staining presents in less than 10% of tumor cells), score 1+ (negative; a faint/barely perceptible membrane staining was detected in more than 10% of tumor cells; these cells exhibit incomplete membrane staining), score 2+ (weakly positive; a weak to moderate complete membrane staining was observed in more than 10% of tumor cells) and score 3+ (strongly positive; a strong complete membrane staining was observed in more than 10% of tumor cells) (Figure 2). Regarding score 2+ (equivocal), the samples were referred to tertiary center in Baghdad city (the capital of Iraq) where CISH (Chromogenic in situ hybridization) analysis was done, which was carried out later on the tissue sections of the same primary tumor. In Iraq, the ZytoDot ® 2C SPEC ERBB2/CEN 17 probe was designed for the simultaneous detection of ERBB2 and centromere 17 in formalin-fixed, paraffin-embedded tissue sections or cell samples [11].

Histologic grading was classified according the Nottingham Grading System which is based on a microscopic evaluation of morphologic and cytologic features of tumor cells, including degree of tubule formation, nuclear pleomorphism, and mitotic count. The sum of these scores stratifies breast tumors into grade 1 (G1; well-differentiated, slow-growing), grade 2 (G2; moderately differentiated), and grade 3 (G3; poorly differentiated, highly proliferative) malignancies [12]. All the results were assessed by two expert pathologists before the final diagnosis.

Analysis of luminal subtypes

According to these parameters four major molecular subtypes are recognized: The luminal A subtype was defined as ER+/HER2- with a KI67 index ≤20% and PR+ with an Allred PR score ≥ 4; the luminal B subtype was defined as an ER+ and/or PR+ tumor, ± HER2+, and with a Ki67 index > 20%, HER2-positive is ER & PR - / HER2 + and Basal-type (triple-negative) is ER, PR and HER2 (negative) [4].

Inclusion / Exclusion criteria

Included in this study were all breast cancer patients diagnosed between January 2012 and August 2020. In addition, any patient with inconclusive results was excluded from the study.

Ethical considerations

Institutional review board (IRB) approval was obtained from the Ethics Committee at Al-Hussein Medical City, Karbala, Iraq.

Statistical analysis

Data of all patients were entered and managed using the Statistical Package for Social Sciences (SPSS) software (version 25). Descriptive statistics of the variables were expressed as number, percentage, median, and range.

Results

There were 920 patients with female breast cancer enrolled in this study. At the time of diagnosis, the mean age was 50.20 ±11.74 years, and the range was (16-97) years. The most frequent histological subtype was invasive ductal carcinoma in 845 patients (91.85%), followed by invasive lobular carcinoma in 39 patients (4.24%), mucinous carcinoma in 25 patients (2.72%), papillary carcinoma in 6 patients (0.65%) and medullary carcinoma in 5 patients (0.54%) as shown in (Figure 3).

Out of the 920 patients, grade II was the most common grade in 463 patients (50.32%), followed by grade III in 286 patients (31.09%) and grade I in 171 patients (18.59%) as shown in (Figure 4).

Luminal A is classified as the significant subtype among our patients in 494 patients (53.69%), followed by luminal B in 146 patients (15.87%), Her-2 positive in 141 patients (15.33%) and basal type in 139 patients (15.11%) as shown in (Figure 5).

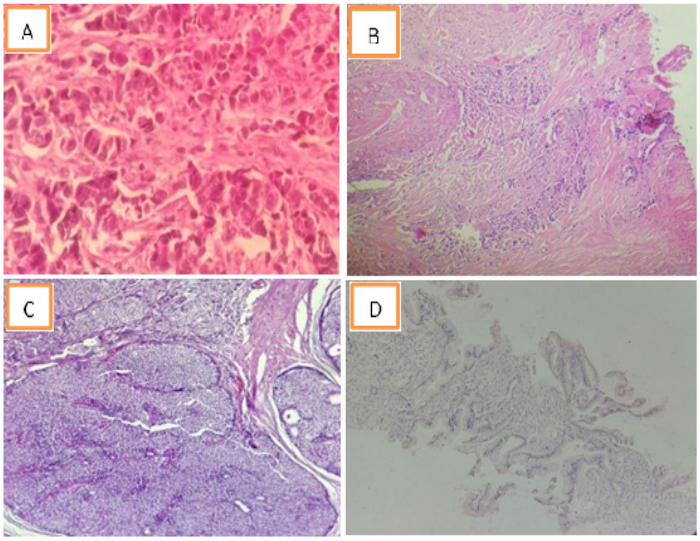

Figure 1. A. Invasive Ductal Carcinoma (400X) B. Invasive Lobular Carcinoma (40X) C. Solid Papillary Carcinoma (100X) D. Invasive Ductal Carcinoma with Mucinous Component (100X).

Discussion

Breast cancer is a complex disease with various subtypes that have different cellular structures, molecular changes, and clinical manifestations. In addition, the prognosis and response to breast cancer treatment depend on multiple variables, including tumor grade & size, lymph node infiltration, ER receptors, PR receptors, and HER-2/neu receptors [13]. In this study, histological pattern and molecular subtype distribution were evaluated in the Middle Euphrates region of Iraq. This is the first large study on breast cancer patients in Karbala province to the best of our knowledge.

Our results showed that the mean age is 50 years. These results are close to those reported in other parts of Iraq (49 years), Saudi Arabia (49 years) and Turkey (51 years), but it significantly younger than the US (59 years) [13-16]. This may be explained because > 58% of the Iraqi population are under 25 years old and only 3.4% are above 65 years [17].

Invasive ductal carcinoma was the most common histological subtype in (91.85%), which is close to prior studies in North Iraq (92.8%), West Iraq (88%), India (95.5%%), Saudi Arabia (88.8%). Still, it is higher than Turkey (78.7%) and the US (75%) [15,18-22].

The second most common histological type was invasive lobular carcinoma, accounting for 4.24% of all cases, which is similar to what has been previously reported in Iraq (3.9%), Saudi Arabia (3%), lower than that of the US (15%) and Turkey (7.8%). Still, it was higher than India, where this histological pattern represents only (1.7%) [15,19,21-23].

On the other hand, mucinous carcinoma represents only (2.72%) similar to the former studies in Iraq (2.1%) and Saudi Arabia (2%) [14,19]. While both papillary carcinoma & medullary carcinoma represent less than (1%) of cases, which is close to results in Saudi Arabia and India, where they represent only (1%) [14,21].

The finding of this study revealed that more than half of our patients' tumors were grade II (50.32%), same results in previous studies in Iraq (67.6%), India (59.5%) and Jordan (48%) [5,21,24].

In our study luminal A subtype was the most common subtype, same results in previous studies in Iraq and the Eastern Mediterranean Region [19,23,25,26]. At the same instant, our results were different than other previous results in Iraq, Indonesia, Vietnam, where luminal B tumors was the predominant subtype [27-29]. Genetics,

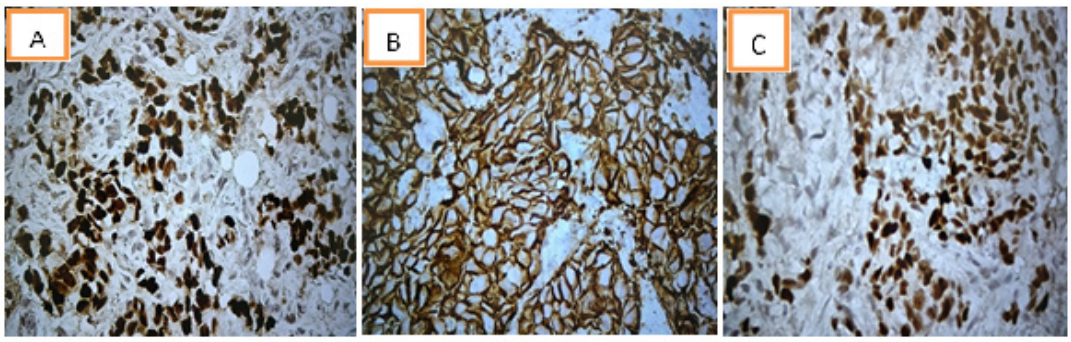

Figure 2. A. Diffuse Nuclear Expression of ER Receptors (400X) B. Diffuse Nuclear Expression of PR Receptors (400X) C. Complete Circumferential and Strong Membranous Stain of Her2/neu Receptors (400X).

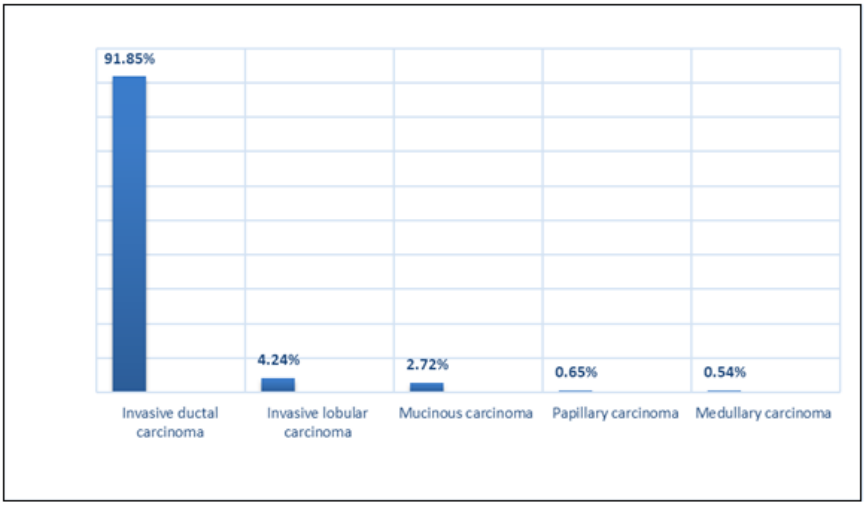

Figure 3. Histological Subtypes of 920 Breast Cancer Patients

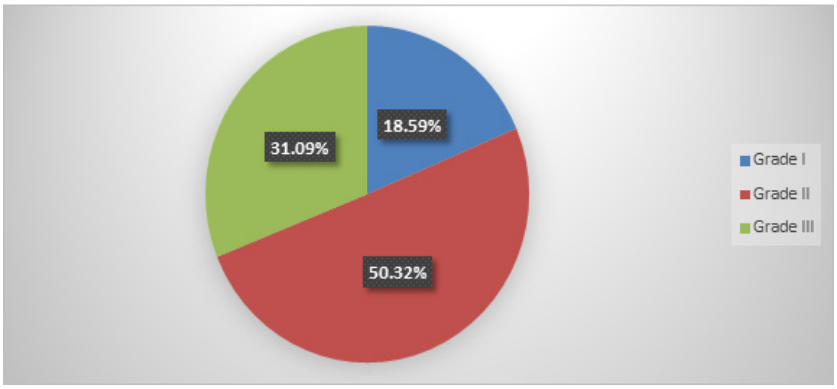

Figure 4. Histological Grades of Breast Cancer Patients

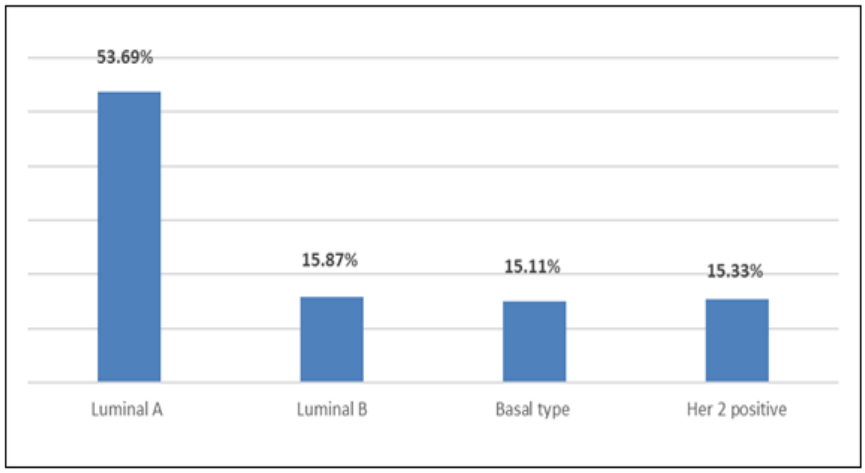

Figure 5. Luminal Subtypes of 920 Breast Cancer Patients

racial, and environmental factors may be responsible for the disparities & the variations in diagnostic facilities may have a role.

However, the proportion of luminal subtypes varies across the world, luminal A was the commonest molecular subtype in (53.70%) of patients, our results were lower than reported in US (73%) Jordan (60%), Turkey (62%), Saudi Arabia (58.5%) and slightly higher than China (48.6%) and Egypt 41.2% [5,8,15,30].

Luminal B represents (15.87%), that is close to findings in Jordan (13%), Turkey (15%), China (16.7%), Saudi Arabia (14.5%) and Egypt (13.9%) and slightly higher than US (11%) [5,8,15,22,30].

The HER-2/neu subtype was presented about (15.33%) of cases, close to Jordan (12%), Saudi Arabia (12.3%), and Egypt (19.4%) but higher than that of Turkey

(8.5%) and US (4%) [5,8,15,22,30,31].

The percentage of triple negative in this study was (15.11%) which is similar to Jordan (15%), Turkey (15%), Saudi Arabia (14.8%), US (12%) and lower than Egypt (28.5%) [5,15,22,30,31].

Our study can be evaluated in terms of its strengths and limitations. A strength of our study is that it can help to provide basic information about breast cancer in Iraq, comparing it to neighboring countries and the world. Moreover, to the best of our knowledge, this study is the first one to be carried out in the Middle Euphrates Region. A limitation of our study is that it covered only the Middle Euphrates Region and not other regions of Iraq.

In conclusion, this study showed that the median age in our patients was a decade younger than in western countries. The most prevalent pathological subtype was invasive ductal carcinoma, and the majority of cases were grade II. In our region, luminal A was the dominant luminal subtype. Future studies in other parts of Iraq with more patients are needed to understand breast cancer dissemination in this country.

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