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Immunohistochemical Evaluation of Androgen Receptor Expression on Triple Negative Breast Cancer in Sample of Iraqi Female Patients (Clinicopathologic study)

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Abstract*—Background:** *Triple-negative breast cancer (TNBC), characterized by the absence of estrogen receptor (ER), progesterone receptor (PR), and Human Epidermal Growth Factor Receptor 2 (HER2) expression, aggressive clinical behavior and lack of targeting hormonal therapy. Studies have emphasized the significance of the androgen receptor (AR) in breast cancer pathogenesis and clinical outcomes, indicating its potential role as a therapeutic target.* **Aim of the study:** *Evaluation of androgen receptor expression in TNBC in a sample of Iraqi patients and its association with some clinicopathological parameters including age, tumor stage and grade.* **Patients and methods:** *A cross sectional study conducted at Al Yarmouk Teaching Hospital and private labs included 60 patients diagnosed with TNBC from mastectomy samples from May 2023 to January 2024. Data were collected focusing on patients with negative ER, PR, and HER-2/neu expression determined by immunohistochemistry.* **Results:** *In this study, 78.3% of patients with TNBC were positive for androgen receptor (AR). These patients had a higher mean age compared to those with AR-negative TNBC. TNBC patients with AR positivity tended to have lower T stage tumors and were more likely to have N0 nodal stage compared to AR-negative TNBC patients. Furthermore, AR-positive TNBC patients showed a trend towards lower TNM stage and lower histological grade compared to AR-negative TNBC patients. However, there was no statistically significant association between AR status with age and T stage with p-value >0.05.* **Conclusion: Higher AR expression is associated with less aggressive clinicopathological features, including lower tumor stage and grade, suggesting a potential role for AR in targeted therapies in managing TNBC.*

***Keywords*—** *Androgen receptor expression, Triple-negative breast cancer*.

I. INTRODUCTION

Breast cancer stands as the most prevalent malignancy affecting women globally. The disease manifests through various biological pathways, each associated with specific subtypes of human breast cancer (1).

The subdivision of human breast cancer using microarray-based gene expression analysis classified breast cancer into up to four distinct subtypes including luminal A, luminal B, HER2-enriched and TNBC (2). Over the past decade to fifteen years, significant advancements in therapeutic agents have notably enhanced clinical outcomes for patients that have steroid receptors expression and/or HER2 expression. However, for a subset of cancers that lack expression for all three markers (referred to as TNBC), clinical outcomes persistently poor and behind those of other subtypes (3).

TNBC, or triple-negative breast cancer, represents a subtype of breast cancer characterized by the absence of estrogen receptor (ER), progesterone receptor (PR), and human epidermal growth factor receptor 2 (HER2) expression. Its prevalence varies considerably, ranging from 6% to 60% of all breast cancer cases depending on the specific population under study (4-7)

The androgen receptor (AR) belongs to the steroid hormone receptor family and is composed of a single polypeptide with various domains (8, 9). The role of AR in TNBC carcinogenesis is significant but remains debated regarding its impact on patient prognosis and its predictive value in TNBC. TNBC lacks expression of estrogen receptor (ER), progesterone receptor (PR), and does not overexpress HER2/neu, rendering it typically aggressive with a poor prognosis despite treatment (10).

Several meta-analyses, including those led by Qu and Wang, covering over 4,000 TNBC cases, demonstrated that AR-positive status is associated with better disease-free state and overall survival (11, 12). However, a meta-analysis by Gonzalez-Angulo et al. noted only a non-significant trend towards improved disease-free state and overall survival. Numerous other studies have reported no significant difference or even a negative impact of AR status on outcomes. The diverse findings highlight the complexity of AR's role in TNBC prognosis and the need for further research to understand these relationships comprehensively(13-15).

When comparing the results of different studies, significant challenges arise due to inherent variations in the definition of androgen receptor (AR) positivity in carcinoma cells. This inconsistency presents a potential confounding factor when analyzing and comparing data reported in the literature. The frequency of AR expression in carcinoma cells varies considerably among studies, ranging from 0% in some studies and reaching 75% in others in TNBC patients. Even

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when studies utilize a uniform 10% cut-off point for defining AR positivity, heterogeneity persists (5-7). Several factors contribute to this heterogeneity, including disease subtype, disease progression, study cohort characteristics, and methodological issues such as tissue preparation, antigen retrieval methods, antibody variations, and staining techniques (16).

Aim of the study

1. To study the Immunohistochemical expression of androgen receptor in triple negative breast cancer as a predictive value for targeted therapy in sample of Iraqi patients.
2. To correlate the expression of androgen receptor in triple negative breast cancer with some clinicopathological parameters (age, tumor stage and grade).

II. PATIENTS AND METHODS

A single center cross-sectional study carried out in Al Yarmouk teaching hospital during the period from the 1st of May 2023 to the 1st of January 2024.

Study population and sampling technique

The study population included 60 patients all were previously diagnosed with invasive breast cancer by consultant pathologist.

Inclusion criteria: Female patients, Triple Negative Breast Cancer (TNBC) diagnosed using, immunohistochemistry, Negative ER, Negative PR, Negative HER-2/neu, IDC NST was the only type included in the study.

Exclusion criteria: Male patients; Patients on neoadjuvant therapy; If the primary tumor showed ER, PR and/or Her2/new positive on confirmatory staining; and Special subtypes other than IDC NST were not included in the study.

Data Collection: The data was collected by the researcher from the archives of teaching lab of Al Yarmouk teaching hospital and private lab in Baghdad for a period from 2022 to 2023.

Microscopic study: A digital light microscope (Micros Austria) was used in the examination of slides, each field was obtained from the region of 5 zones of the slide (corners and the center) which were randomly selected, then the image captured in high definition (HD) using the same device built in camera that displays the image on the LCD screen.

Immunohistochemical Scoring: Immunohistochemical scoring was conducted by a specialized pathologist with expertise in mammary gland pathology. Following established methodologies, as documented in previous studies (17), the cut-off threshold for determining estrogen receptor (ER) and progesterone receptor (PR) positivity was set at ≥1%. This same threshold was applied for assessing androgen receptor (AR) positivity. Subsequently, AR expression was categorized as either negative or positive based on this threshold. Clinicopathological parameters were then analyzed in correlation with the androgen receptor status (17).

Following an extensive literature review, the expression of androgen receptors (AR) was further analyzed using s quantitative analysis. This involved evaluating the percentage of cells exhibiting nuclear positivity, this is in line previous studies that correlated AR expression levels with cancer severity (17). Prior to analysis, this approach was reviewed and refined by two consultant pathologists to ensure methodological accuracy.

The quantitative analysis scoring system was as follows: AR percentage of expression score on scale of 1 to 3

- Score (0): 0%;
- Score (1+): 1%–29%;
- Score (2+): 30%–69%;
- Score (3+): ≥70%.

Statistical analysis: Analysis of data was carried out using the available statistical package of SPSS-26 (Statistical Packages for Social Sciences- version 26). Data were presented as simple measures of frequency, percentage, mean, standard deviation and range (minimum-maximum values). The statistical significance difference for different percentages (qualitative data) was tested using Pearson Chi-square test with application of Yate's correction or Fisher Exact test whenever applicable. Statistical significance was considered whenever the P value was equal or less than 0.05.

III. RESULTS

Descriptive statistics: A total of 60 patients diagnosed with TNBC were involved in the current study. Patients with androgen receptor positive were 47 (78.3%) while those with androgen receptor negative were 13 (21.7%). The mean age for the patients of the study was (49.78±8.9). The mean age for patients with androgen receptor positive with mean age of (50.76±9.68) while the mean age for patients with androgen receptors negative was (46.23±3.51) as shown in table 1

Table 1: The distribution Different clinicopathological parameters for patients with TNBC of the study.

			Androgen receptor status		Total
			Positive	Negative	
	Mean (SD)		50.76 (±9.68)	46.23 (±3.51)	49.78 (±8.9)
Age groups	30-39 YO	Fr	4	0	4
		%	16.0%	0.0%	6.7%
	40-49 YO	Fr	9	10	19
		%	36.0%	28.6%	31.7%
	50-59 YO	Fr	8	17	25
		%	32.0%	48.6%	41.7%
	60-69 YO	Fr	0	7	7
		%	0.0%	20.0%	11.7%
	70-79 YO	Fr	4	1	5
		%	16.0%	2.9%	8.3%
T stage	T1	Fr	13	26	39
		%	52.0%	74.3%	65.0%
	T2	Fr	10	8	18
		%	40.0%	22.9%	30.0%
	T3	Fr	2	1	3
		%	8.0%	2.9%	5.0%
N stage	N0	Fr	14	25	39
		%	56.0%	71.4%	65.0%
	N1	Fr	10	6	16
		%	40.0%	17.1%	26.7%
	N2	Fr	1	2	3
		%	4.0%	5.7%	5.0%
	N3	Fr	0	2	2
		%	0.0%	5.7%	3.3%
TNM stage	I	Fr	13	23	36
		%	52.0%	65.7%	60.0%

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	II	Fr	8	9	17
		%	32.0%	25.7%	28.3%
	III	Fr	4	3	7
		%	16.0%	8.6%	11.7%
Tumor grade	Grade 1	Fr	6	6	12
		%	24.0%	17.1%	20.0%
	Grade 2	Fr	17	21	38
		%	68.0%	60.0%	63.3%
	Grade 3	Fr	2	8	10
		%	8.0%	22.9%	16.7%
Total		Fr	25	35	60
		%	100.0%	100.0%	100.0%

For the T stage of TNM staging, patients with androgen receptor positive TNBC who had T1 tumor size were 32 (53.3%) while those who had T2 tumor size were 13 (21.7%) and patients with T3 tumor size were 2 (3.3%). Meanwhile patients with androgen receptor negative TNBC who had T1 tumor size were 7 (11.7%) and patients with T2 tumor size were 5 (8.3%) while one patient (1.7%) had T3 tumor size as shown in figure 1 below.

Nodal stage of the tumor showed that majority of TNBC patients who had androgen positive receptors were staged with N0 nodal stage with 35 (58.3%), those with N1 nodal stage were 10 (16.7%) while only one patient (1.7%) was staged with N2 and N3 nodal stage. On the contrary, patients with androgen negative tumors were had more patients stage with N1 nodal stage TNBC with 6 (10%) of the patients and those who had N0 nodal stage were 4 (6.7%). Patients with N2 nodal stage were 2 (3.3%) and only one patient (1.7%) was staged with N3 nodal stage as shown in figure 2 below.

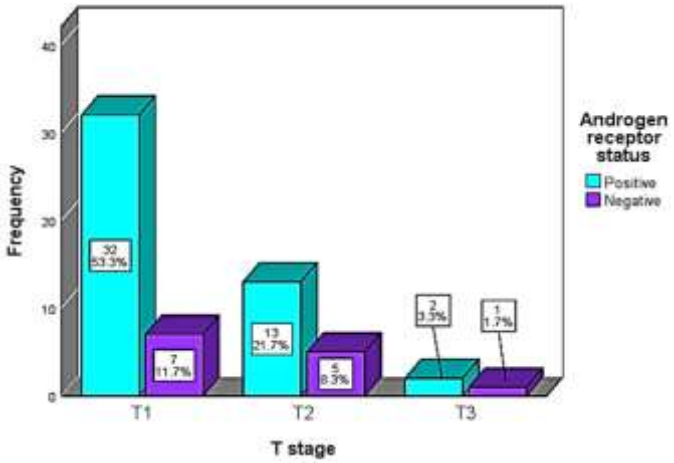

Figure 1: T stage of the TNM staging for both androgen positive and negative TNBC patients of the study.

The TNM staging of androgen positive TNBC shows a trend of declining number of patients with increasing TNM stage with 32 (53.3%) of patients with TNM stage I , 11 (18.3%) with TNM stage II and 4 (6.7%) with TNM stage III. While patients with androgen negative TNBC showed higher number of patients who had TNM stage II with 6 (10%), those with TNM stage I were 4 (6.7%) and patients with TNM stage III were 3 (5%) as shown in figure 3.

The histological grade for TNBC patients with androgen receptor positive showed that the majority of these patients had grade II tumors with 31 (51.7%) of these patients. While patients with grade I tumors were 11 (18.3%) and those with grade III tumors were 5 (8.3%). The histological grade for androgen negative TNBC tumors showed that 1 (1.7%) patient had grade I tumor, 7 (11.7%) had grade II tumors (figure 7 and 8) and 5 (8.3%) had grade III tumors as shown in figure 4.

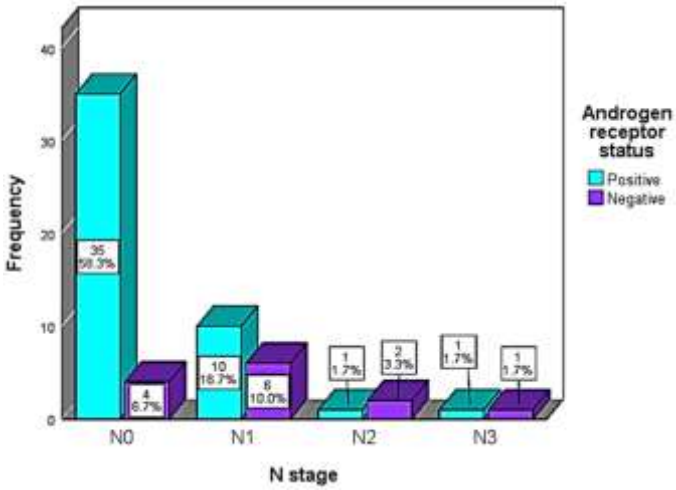

Figure 2: N stage of the TNM staging for both androgen positive and negative TNBC patients of the study.

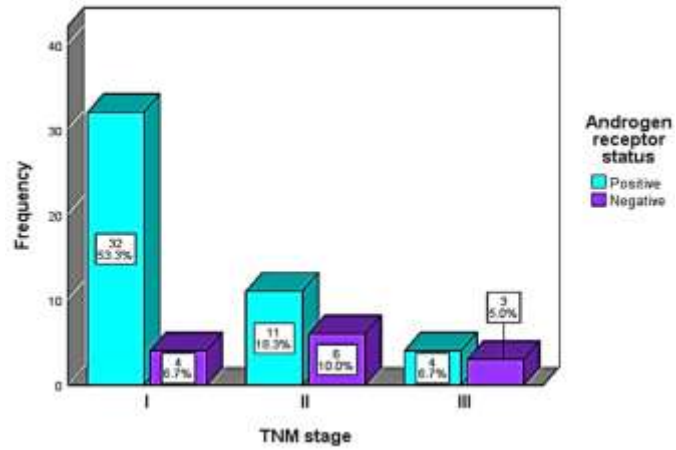

Figure 3: TNM stage for both androgen positive and negative TNBC patients of the current study.

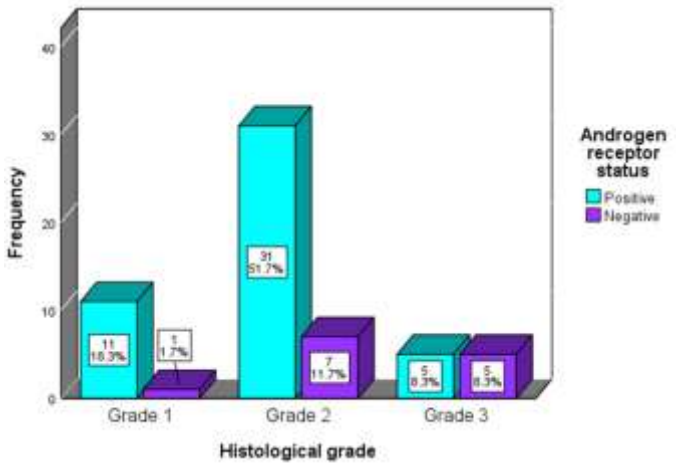

Figure 4: The histological grade for both androgen positive and negative TNBC patients of the current study.

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Figure 5 below shows the score for androgen receptor staining in TNBC patients which revealed a score of (0) in 13 (21.67%) of the patients (figure 8), score of (+1) in 21 (35%) of the patients (figure 10 and 11). while 10 (16.67%) had a tumor score of (+2) (figure 12 and 13). Patients who had a tumor score of (+3) were 16 (26.67%) (figure 14 and 15).

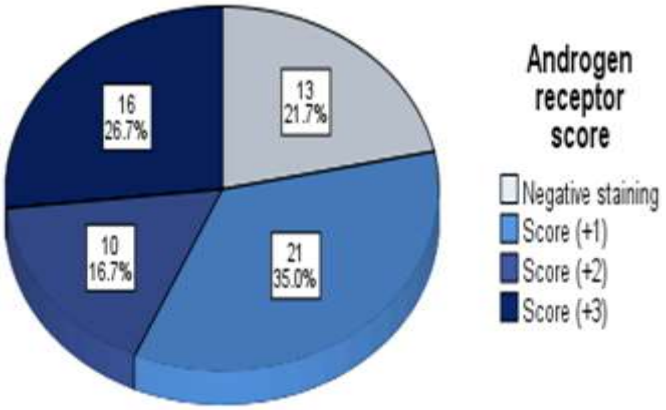

Figure 5: The score of androgen receptor staining for TNBC patients of the current study.

Androgen receptor (AR) status: Table 2 below shows the association for age of androgen receptor status, there was no statistically significant association with p-value of 0.24.

Table 2: The association of androgen receptor status with age in patients with TNBC of the study.

			Androgen receptor status		Total	Pearson chi-square	P-value
			Positive	Negative			
Age groups	30-39 YO	Fr	4	0	4	5.443	0.24
		%	6.7%	0.0%	6.7%		
	40-49 YO	Fr	13	6	19		
		%	21.7%	10.0%	31.7%		
	50-59 YO	Fr	22	3	25		
		%	36.7%	5.0%	41.7%		
	60-69 YO	Fr	4	3	7		
		%	6.7%	5.0%	11.7%		
	70-79 YO	Fr	4	1	5		
		%	6.7%	1.7%	8.3%		
Total		Fr	47	13	60		
		%	78.3%	21.7%	100.0%		

The association for androgen receptor status and T-stage of the tumor showed no statistically significant difference with p-value of 0.462 as shown in table 3.

Meanwhile, the statistical significance of the association for androgen receptor status with N-stage of the tumor, there was a statistically significant difference with p-value 0.011 as shown in table 4.

For the association of TNM staging of the tumors with androgen receptor status for patients with TNBC of the study, there was a statistically significant difference with p-value of 0.037 as shown in table 5.

Table 3: The association for androgen receptor status and T-stage of the tumors in patients with TNBC of the study

			Androgen receptor status		Total	Pearson chi-square	P-value
			Positive	Negative			
T stage	T1	Fr	32	7	39	1.454	0.462
		%	53.3%	11.7%	65.0%		
	T2	Fr	13	5	18		
		%	21.7%	8.3%	30.0%		
	T3	Fr	2	1	3		
		%	3.3%	1.7%	5.0%		
Total		Fr	47	13	60		
		%	78.3%	21.7%	100.0%		

Table 4: The association for androgen receptor status and N-stage of the tumors in patients with TNBC of the study

			Androgen receptor status		Total	Pearson chi-square	P-value
			Positive	Negative			
N stage	N0	Fr	35	4	39	10.025	0.011
		%	58.3%	6.7%	65.0%		
	N1	Fr	10	6	16		
		%	16.7%	10.0%	26.7%		
	N2	Fr	1	2	3		
		%	1.7%	3.3%	5.0%		
	N3	Fr	1	1	2		
		%	1.7%	1.7%	3.3%		
Total		Fr	47	13	60		
		%	78.3%	21.7%	100.0%		

Table 5: The association for androgen receptor status and TNM staging of the tumors in patients with TNBC of the study

			Androgen receptor status		Total	Pearson chi-square	P-value
			Positive	Negative			
TNM staging	I	Fr	32	4	36	6.225	0.037
		%	53.3%	6.7%	60.0%		
	II	Fr	11	6	17		
		%	18.3%	10.0%	28.3%		
	III	Fr	4	3	7		
		%	6.7%	5.0%	11.7%		
Total		Fr	47	13	60		
		%	78.3%	21.7%	100.0%		

Likewise, tumor grade also showed a statistically significant association with p-value of 0.045 as shown in table 6 below.

Androgen receptor (AR) percentage

The percentage of AR expression in association with T stage showed no statistically significant difference with p-value of 0.185. Meanwhile, there was a statistically significant difference with N stage with p-value of 0.019 as shown in table 7 below.

Analysis of data to assess the association of both TNM stage and tumor grade showed a statistically significant association with the percentage for AR expression with p-value<0.05 as shown in table 8.

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Table 6: The association for androgen receptor status and grade of the tumors in patients with TNBC of the study.

			Androgen receptor status		Total	Pearson chi-square	P-value
			Positive	Negative			
Tumor grade	Grade 1	Fr	11	1	12	6.223	0.045
		%	18.3%	1.7%	20.0%		
	Grade 2	Fr	31	7	38		
		%	51.7%	11.7%	63.3%		
	Grade 3	Fr	5	5	10		
		%	8.3%	8.3%	16.7%		
Total		Fr	47	13	60		
		%	78.3%	21.7%	100.0%		

Table 7: The association of the Immuno-histochemical score for AR expression with T and N stage of the tumors in patients with TNBC of the study.

			Percentage of AR expression				Total	Pearson chi-square	P-value
			Negative staining	Score (+1)	Score (+2)	Score (+3)			
T stage	T1	Fr	7	13	10	9	39	7.63	0.185
		%	11.7%	21.7%	16.7%	15.0%	65.0%		
	T2	Fr	5	7	0	6	18		
		%	8.3%	11.7%	0.0%	10.0%	30.0%		
	T3	Fr	1	1	0	1	3		
		%	1.7%	1.7%	0.0%	1.7%	5.0%		
N stage	N0	Fr	4	12	10	13	39	15.22	0.019
		%	6.7%	20.0%	16.7%	21.7%	65.0%		
	N1	Fr	6	7	0	3	16		
		%	10.0%	11.7%	0.0%	5.0%	26.7%		
	N2	Fr	2	1	0	0	3		
		%	3.3%	1.7%	0.0%	0.0%	5.0%		
	N3	Fr	1	1	0	0	2		
		%	1.7%	1.7%	0.0%	0.0%	3.3%		
Total		Fr	13	21	10	16	60		
		%	21.7%	35.0%	16.7%	26.7%	100.0%		

Table 8: The association of the Immuno-histochemical score for AR expression with TNM stage of the tumors and tumor grade in patients with TNBC of the study.

			Percentage of AR expression				Total	Pearson chi-square	P-value
			Negative staining	Score (+1)	Score (+2)	Score (+3)			
TNM stage	I	Fr	4	13	10	9	36	12.22	0.034
		%	6.7%	21.7%	16.7%	15.0%	60.0%		
	II	Fr	6	5	0	6	17		
		%	10.0%	8.3%	0.0%	10.0%	28.3%		
	III	Fr	3	3	0	1	7		
		%	5.0%	5.0%	0.0%	1.7%	11.7%		
Tumor grade	G1	Fr	1	3	6	2	12	14.03	0.015
		%	1.7%	5.0%	10.0%	3.3%	20.0%		
	G2	Fr	7	14	4	13	38		
		%	11.7%	23.3%	6.7%	21.7%	63.3%		
	G3	Fr	5	4	0	1	10		
		%	8.3%	6.7%	0.0%	1.7%	16.7%		
Total		Fr	13	21	10	16	60		
		%	21.7%	35.0%	16.7%	26.7%	100.0%		

Histological assessment of specimens

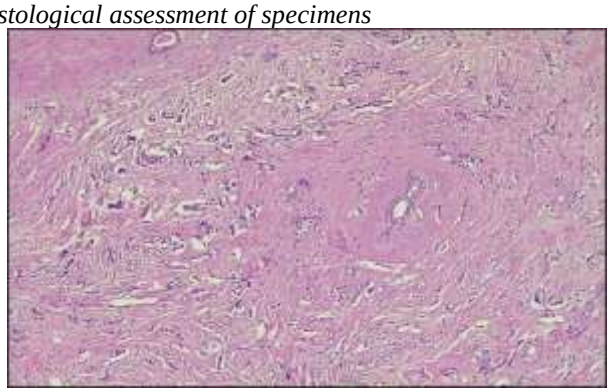

Figure 6: Breast tissue showing Invasive ductal carcinoma (NST) grade II according to Nottingham histological grading score (H&E 10x).

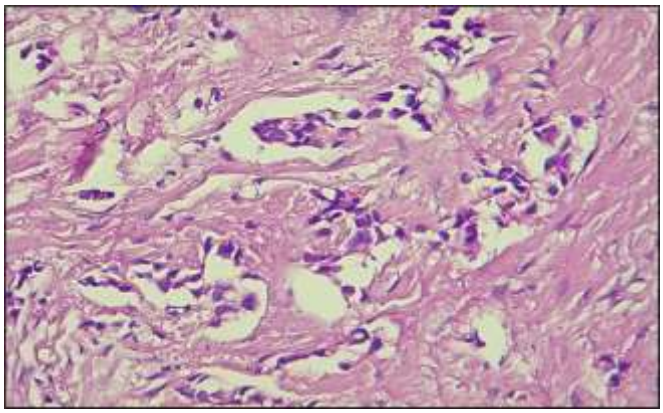

Figure 7: Breast tissue showing Invasive ductal carcinoma (NST) grade II according to Nottingham histological grading score (H&E 40x).

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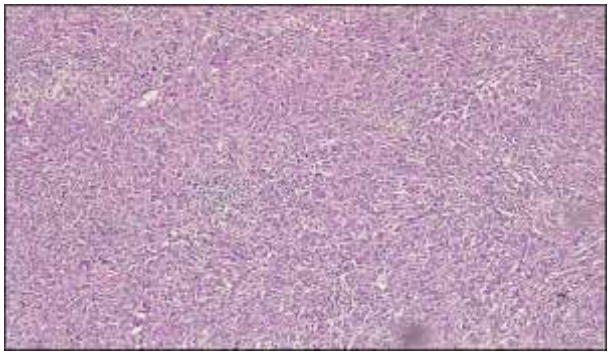

Figure 8: Breast tissue showing invasive ductal carcinoma (NST) grade III according to Nottingham histological grading score (H&E 10x)

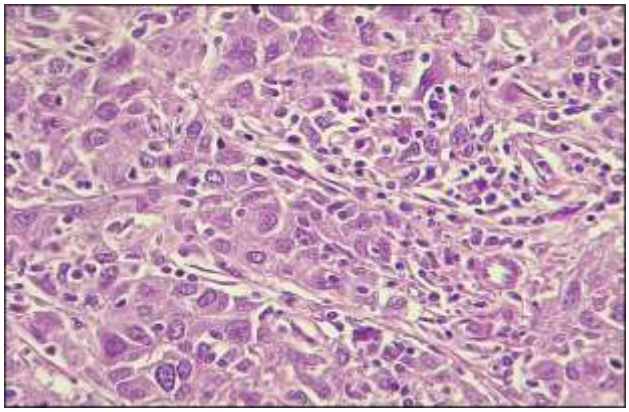

Figure 9: Breast tissue showing invasive ductal carcinoma (NST) grade III according to Nottingham histological grading score (H&E 40x).

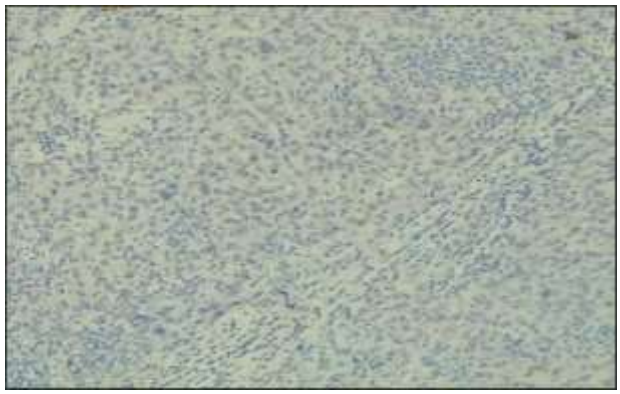

Figure 10: Breast tissue showing negative nuclear staining with score zero of AR protein (10x)

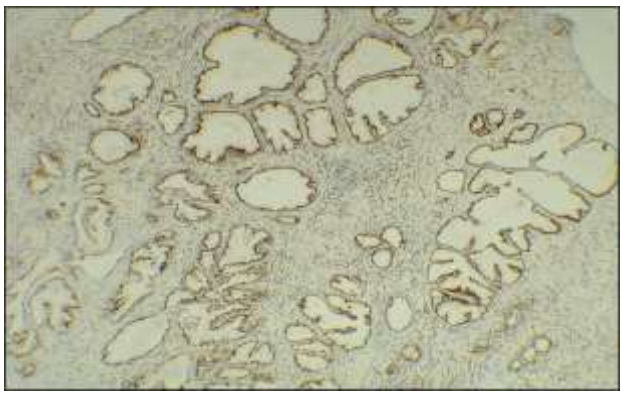

Figure 11: Breast tissue showing positive internal control showing strong positive nuclear staining in prostatic stromal and epithelial cells (10x).

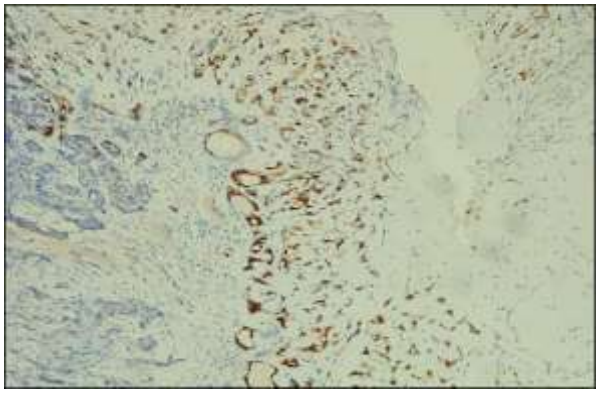

Figure 12: Breast tissue showing invasive ductal carcinoma showing positive nuclear staining of androgen receptors (AR) protein score +1 (10x)

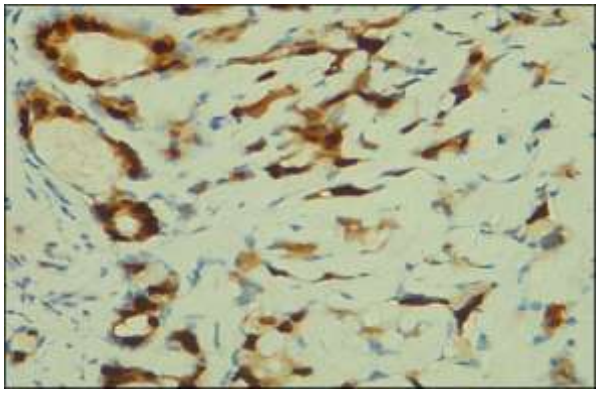

Figure 13: Breast tissue showing invasive ductal carcinoma showing positive nuclear staining of androgen receptors (AR) protein score +1 (40x)

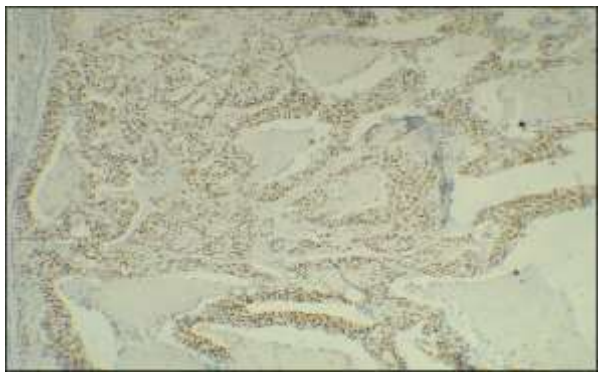

Figure 14: Breast tissue showing invasive ductal carcinoma showing positive nuclear staining of androgen receptor (AR) protein score +2 (10x)

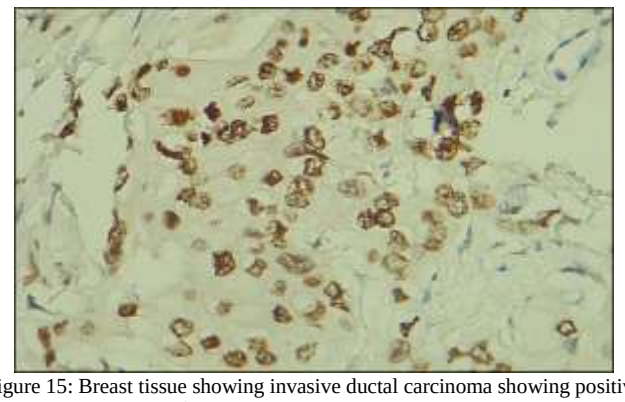

Figure 15: Breast tissue showing invasive ductal carcinoma showing positive nuclear staining of androgen receptors (AR) protein score +2 (40x)

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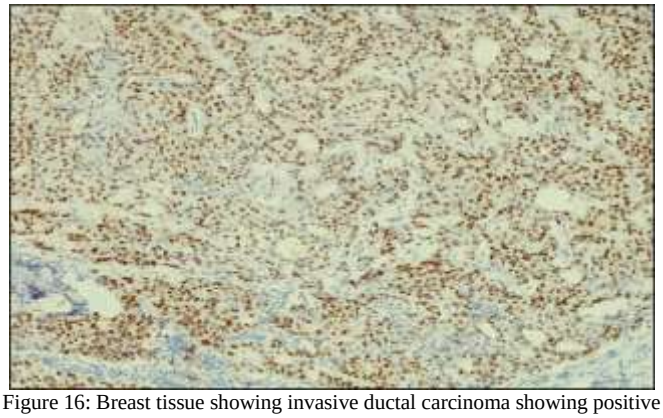

Figure 16: Breast tissue showing invasive ductal carcinoma showing positive nuclear staining of androgen receptors (AR) protein score +3 (10x)

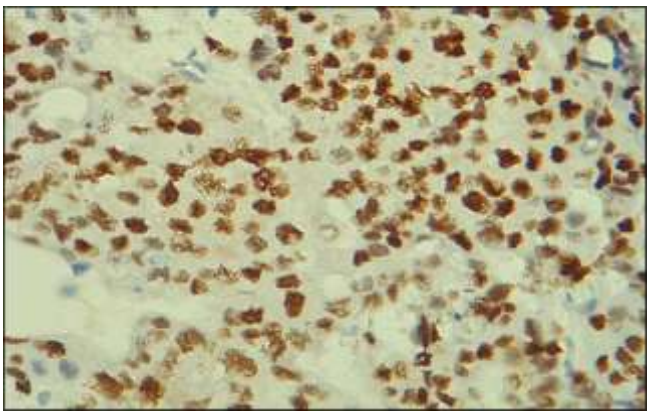

Figure 17: Breast tissue showing invasive ductal carcinoma showing positive nuclear staining of androgen receptors (AR) protein score +3 (40x)

IV. Discussion

Triple-negative breast cancer (TNBC) is well known for its aggressive clinical behavior (18). ER and PR are well known as promoters for the pathogenesis and growth of breast cancer and clinically, they have been widely recognized to guide endocrine therapy of breast cancer. The absence of ER, PR and HER2 receptors in TNBC limits the effectiveness of targeted therapies commonly used for other types of breast cancer (19).

Despite accounting for only 15–25% of all breast cancers, TNBC exhibits consistent proportions across various age groups, with younger and older women showing increased rates of specific subtypes such as BRCA-associated and basal-like TNBC, as well as apocrine and neuroendocrine TNBC variants (20). Recent trends indicate a steady rise in the incidence of TNBC, with reports suggesting that TNBCs accounted for 24% of newly diagnosed breast cancers. In 2018 alone, approximately 2,088,849 cases of TNBC were reported, making it one of the most prevalent cancers in women worldwide (21).

TNBC remains challenging, with an average survival rate of approximately 10.2 months based on current therapeutic options. Despite advancements in treatment, the 5-year survival rate for TNBC remains relatively low, particularly for cases with distant metastases. Regional tumors exhibit a higher 5-year survival rate of 65% (22).

The reported prevalence of triple-negative breast cancer (TNBC) in Iraqi women stands at 10.4% (23), adding a significant dimension to the global epidemiology of this aggressive breast cancer subtype. While the Iraqi prevalence aligns with the general trend observed worldwide, it also reflects the burden of TNBC within a specific population.

In the last two decades, the role of androgen receptors (AR) has gained attention in research due to its potential role in TNBC. The investigation into the androgen receptor has provided new insights into potential therapeutic avenues. Studies suggest that a subset of TNBC tumors may express androgen receptors. Unlike ER, PR, or HER2, the presence of androgen receptors in TNBC opens up the possibility of using hormonal therapies to target this specific receptor. It has been observed that TNBC tumors with androgen receptor expression may have distinct molecular characteristics and clinical behaviors compared to those without (5, 18, 24).

The role of androgen receptors in TNBC is complex and has been a subject of debate. On one hand, the androgen receptor may have a tumor-suppressive effect, acting as a negative regulator of cell growth(25). On the other hand, in some cases, androgen receptor signaling may promote tumor progression(25, 26). The exact impact of androgen receptors in TNBC likely depends on the specific molecular context of the tumor (27).

In this study, the incidence of androgen receptor expression among patients with TNBC of the study reached 78%. Previous studies showed high variability for androgen receptor expression in TNBC with reported androgen receptor expression ranging 7-75%. This wide range in the difference for the incidence of AR expression can be because of the differences in the methodology used. The positivity for androgen receptor immunohistochemistry threshold in tissue fixation is different from one study to another, currently there is no standards or consensus for guidelines on the acceptable threshold and scoring for reactivity of androgen receptor positive tumors. In this study, a cut-off point of 1% was used to determine androgen receptor positivity insight of ER and PR positivity cut-off point recommendation by the ASCO/CAP guidelines (28). The smaller sample size of the study could be a reason as well for the large difference between the studies.

Analysis of the TNM staging of tumors in association with androgen receptor status showed that there was no statistically significant difference observed in association with the T stage with androgen receptor status. However, notable findings emerged in relation to the N stage and overall TNM stage, both of which demonstrated a statistically significant association with androgen receptor status. These findings emphasize the interplay between tumor staging and androgen receptor expression, and were consistent with the findings of Luo X et al and Mrklic I et al who showed a statistically significant association of tumor stage with androgen receptor status (7, 27). Luo X et al showed similar findings with a statistically significant association shown only with nodal stage in association with androgen receptor status (27).

Likewise, for the association of the histological grade of the tumor with androgen receptor status, there was statistically

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significant and consistent with the findings of Luo X et al, Mrklic I et al, tang D et al and Gasparini P et al (7, 26, 29, 30).

The Clinicopathological parameters previously mentioned when further studied showed inverse relationship between androgen positivity and tumor stage and grade which is also in accordance with the findings of the previously mentioned studies and so, it can be inferred that the presence of positive androgen receptor (AR) immunostaining is related with less aggressive tumors, this is supported by the findings of this study in which there was a statistically significant association between percentage of androgen receptors (AR) expression with N stage, TNM stage and the grade of the tumor. Further inspection of data showed high levels of AR expression including score (+2) and (+3) showed low number of patients with high TNM stage and tumor grade, in contrast, patients with negative or low AR expression showed higher number of patients with high TNM stage and grade of tumor. Extensive literature review showed no previously reported similar type of analysis done between level of AR expression and tumor aggressiveness assessment using tumor staging and grading

Androgen receptor expression have a significant influence on breast cancer development; however, the precise mechanism through which they contribute to this process remains inadequately elucidated

The mean age for patients of the study was 49.7 years old, although this is lower than that reported in previous studies that reported mean age ranging between 54 to 61 years old, patients of this study still has a mean age for post-menopausal period comparable to that reported previously (18, 24, 27, 31). Differences could be due to differences in the demographics of the patients in different areas exposed to different risk factors, the small sample size could be another reason. Moreover, there was no statistically significant difference between AR positive and negative patients for age. This was consistent with the findings seen in previous studies (31, 32).

V. CONCLUSIONS

1. While AR expression varies widely among TNBC tumors, the expression in this study reached about 78%.
2. The association of AR association with less aggressive clinicopathological features as suggested by the lower tumor stage and grade in patients with higher AR expression in this study.

REFERENCES

1. Dunn B. Cancer: Solving an age-old problem. Nature. 2012;483(7387):S2-S6.
2. Lund MJ, Butler EN, Hair BY, Ward KC, Andrews JH, Oprea-Ilies G, et al. Age/race differences in HER2 testing and in incidence rates for breast cancer triple subtypes: a population-based study and first report. Cancer: Interdisciplinary International Journal of the American Cancer Society. 2010;116(11):2549-59.
3. Carey LA. Directed therapy of subtypes of triple-negative breast cancer. The oncologist. 2010;15(S5):49-56.
4. Meijnen P, Peterse JL, Antonini N, Rutgers EJ, van de Vijver MJ. Immunohistochemical categorisation of ductal carcinoma in situ of the breast. British journal of cancer. 2008;98(1):137-42.
5. Park S, Koo J, Park HS, Kim JH, Choi SY, Lee JH, et al. Expression of androgen receptors in primary breast cancer. Annals of oncology. 2010;21(3):488-92.
6. Niemeier LA, Dabbs DJ, Beriwal S, Striebel JM, Bhargava R. Androgen receptor in breast cancer: expression in estrogen receptor-positive tumors and in estrogen receptor-negative tumors with apocrine differentiation. Modern Pathology. 2010;23(2):205-12.
7. Luo X, Shi Y, Li Z, Jiang W. Expression and clinical significance of androgen receptor in triple negative breast cancer. Chinese journal of cancer. 2010;29(6):585-90.
8. Olefsky JM. Nuclear receptor minireview series. Journal of Biological Chemistry. 2001;276(40):36863-4.
9. Brinkmann AO. Molecular mechanisms of androgen action–a historical perspective. Androgen Action: Methods and Protocols. 2011:3-24.
10. Gerratana L, Basile D, Buono G, De Placido S, Giuliano M, Minichillo S, et al. Androgen receptor in triple negative breast cancer: A potential target for the targetless subtype. Cancer treatment reviews. 2018;68:102-10.
11. Qu Q, Mao Y, Fei X-c, Shen K-w. The impact of androgen receptor expression on breast cancer survival: a retrospective study and meta-analysis. PloS one. 2013;8(12):e82650.
12. Wang C, Pan B, Zhu H, Zhou Y, Mao F, Lin Y, et al. Prognostic value of androgen receptor in triple negative breast cancer: A meta-analysis. Oncotarget. 2016;7(29):46482.
13. Cochrane DR, Bernales S, Jacobsen BM, Cittelly DM, Howe EN, D'Amato NC, et al. Role of the androgen receptor in breast cancer and preclinical analysis of enzalutamide. Breast Cancer Research. 2014;16:1-19.
14. Aleskandarany MA, Abduljabbar R, Ashankyty I, Elmouna A, Jerjees D, Ali S, et al. Prognostic significance of androgen receptor expression in invasive breast cancer: transcriptomic and protein expression analysis. Breast cancer research and treatment. 2016;159:215-27.
15. Park S, Koo JS, Kim MS, Park HS, Lee JS, Kim SI, et al. Androgen receptor expression is significantly associated with better outcomes in estrogen receptor-positive breast cancers. Annals of Oncology. 2011;22(8):1755-62.
16. McNamara KM, Yoda T, Takagi K, Miki Y, Suzuki T, Sasano H. Androgen receptor in triple negative breast cancer. The Journal of steroid biochemistry and molecular biology. 2013;133:66-76.
17. Asano Y, Kashiwagi S, Goto W, Tanaka S, Morisaki T, Takashima T, et al. Expression and clinical significance of androgen receptor in triple-negative breast cancer. Cancers. 2017;9(1):4.
18. He J, Peng R, Yuan Z, Wang S, Peng J, Lin G, et al. Prognostic value of androgen receptor expression in operable triple-negative breast cancer: a retrospective analysis based on a tissue microarray. Medical oncology. 2012;29:406-10.
19. Lyons TG. Targeted therapies for triple-negative breast cancer. Current treatment options in oncology. 2019;20(11):82.
20. Yin L, Duan J-J, Bian X-W, Yu S-c. Triple-negative breast cancer molecular subtyping and treatment progress. Breast Cancer Research. 2020;22:1-13.
21. Singh S, Numan A, Maddiboyina B, Arora S, Riadi Y, Md S, et al. The emerging role of immune checkpoint inhibitors in the treatment of triple-negative breast cancer. Drug discovery today. 2021;26(7):1721-7.
22. Almansour NM. Triple-negative breast cancer: a brief review about epidemiology, risk factors, signaling pathways, treatment and role of artificial intelligence. Frontiers in Molecular Biosciences. 2022;9:836417.
23. Lattef FA, Ali NFM, Abdulmajeed BM, Mohammed AS. Triple Negative Breast Tumors In Iraqi Women. Indian Journal of Forensic Medicine & Toxicology. 2020;14(1):944-9.
24. Hu R, Dawood S, Holmes MD, Collins LC, Schnitt SJ, Cole K, et al. Androgen receptor expression and breast cancer survival in postmenopausal women. Clinical cancer research. 2011;17(7):1867-74.
25. Garay JP, Karakas B, Abukhdeir AM, Cosgrove DP, Gustin JP, Higgins MJ, et al. The growth response to androgen receptor signaling in ER α-negative human breast cells is dependent on p21 and mediated by MAPK activation. Breast Cancer Research. 2012;14:1-17.
26. Birrell SN, Hall RE, Tilley WD. Role of the androgen receptor in human breast cancer. Journal of mammary gland biology and neoplasia. 1998;3:95-103.
27. Mrklić I, Pogorelić Z, Ćapkun V, Tomić S. Expression of androgen receptors in triple negative breast carcinomas. Acta histochemica. 2013;115(4):344-8.
28. Hammond MEH, Hayes DF, Dowsett M, Allred DC, Hagerty KL, Badve S, et al. American Society of Clinical Oncology/College of American

Pathologists guideline recommendations for immunohistochemical testing of estrogen and progesterone receptors in breast cancer (unabridged version). Archives of pathology & laboratory medicine. 2010;134(7):e48-e72.
29. Tang D, Xu S, Zhang Q, Zhao W. The expression and clinical significance of the androgen receptor and E-cadherin in triple-negative breast cancer. Medical oncology. 2012;29:526-33.
30. Gasparini P, Fassan M, Cascione L, Guler G, Balci S, Irkkan C, et al. Androgen receptor status is a prognostic marker in non-basal triple negative breast cancers and determines novel therapeutic options. PLoS One. 2014;9(2):e88525.
31. McGhan LJ, McCullough AE, Protheroe CA, Dueck AC, Lee JJ, Nunez-Nateras R, et al. Androgen receptor-positive triple negative breast cancer: a unique breast cancer subtype. Annals of surgical oncology. 2014;21:361-7.