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Assessing Clinicopathological Features and Prognosis of Triple-Negative Breast Cancer Patients: A Single-Center Study in Turkey

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ABSTRACT

Background: Triple-negative breast cancer (TNBC) is defined as tumors without estrogen receptor (ER), progesterone receptor (PR), and human epidermal growth factor receptor 2 (HER2) expression. This cancer is associated with higher rates of recurrence risk when compared to other subtypes of breast cancers. In this study, we aimed to explore the basic clinicopathological characteristics, prognosis, and recurrence patterns of TNBC patients.

Methods: In the current study, forty-five TNBC female patients operated on for breast cancer in the General Surgery Clinic of Kayseri City Training and Research Hospital between 2016 and 2021 were included and retrospectively evaluated.

Results: The percentage of TNBC was 12% of the 502 breast cancer patients who could access all three pieces of receptor information. The mean age of the patients was 58.9±15.2 years (27-90), and the mean BMI was 30.4±5.17 (21.5-40.6). It was observed that the most common histological subtype was invasive ductal carcinoma, and at the time of diagnosis, 11 patients were stage 1 (24.4%), 31 patients were stage 2 (68.8%), 2 patients were stage 3 (4.4%), and 1 patient was stage 4 (2.2%). During the follow-up period, 11 patients (24.4%) developed metastasis and the most common sites were the brain and bones. The mean time from diagnosis to metastasis was 20.7±5.75 (12-29) months. The 3-year disease-free survival was 62%, and the 3-year overall survival (OS) was 70%.

Conclusion: TNBCs are cancers with varying prevalence, poor prognosis, and limited treatment alternatives. The prevalence of TNBC in our center was found to be lower than the literature rates and consistent with the literature, the lymph node stage was related to poor OS and disease free survival (DFS).

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INTRODUCTION

Breast cancer has molecular subtypes based on the expression of hormone receptors and human epidermal growth factor receptor 2 (HER2) and has been shown

to have different clinicopathological features and prognoses.¹ Triple-negative breast cancer (TNBC) is defined as tumors without estrogen receptor (ER), progesterone receptor (PR), and HER2 expression.² TNBC is seen at a frequency of 15-20% of all breast cancers and is associated with increased local recurrence, distant metastasis, and poor prognosis in the first 3 to 5 years after diagnosis.^{3, 4} TNBC-related risk factors have been identified, such as BRCA mutation, ethnic differences, young age, and body

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mass index.^{5, 6} In a large series conducted by the National Breast Cancer Registry Program of the Turkish Breast Diseases Associations, the incidence of TNBC in this country was found to be 8.1%, but studies from different regions reported different incidences such as 12% and 27% and different survival rates.⁷⁻⁹ Today, breast cancer is a systemic disease, and individual treatment plans for the tumor are recommended. TNBC has no targeted treatment alternative; it can be seen in different incidences in different societies, treatment responses are also different, and it is said to be a heterogeneous disease in itself. This study aims to determine the prevalence of TNBC in our regional hospital where breast cancer patients are treated and to assess the associated risk factors and prognosis in our regional population.

METHODS

The study was conducted with the approval of the Non-interventional Clinical Research Ethics Committee of Kayseri City Training and Research Hospital (Protocol No: 2021/391). Forty-five of 523 patients diagnosed and operated on for breast cancer in the General Surgery Clinic of Kayseri City Training and Research Hospital between January 2016 and January 2021 and whose data could be accessed were included in the study. All patients had a histologically confirmed diagnosis of invasive breast cancer; initial breast cancer staging was determined according to the American Joint Committee on Cancer (AJCC). Primary tumor grade was evaluated depending on the Nottingham modification of Bloom-Richardson criteria. Baseline ER and PR status was determined by IHC staining, and if the percentage of positively stained cells was less than 1%, they were considered negative. Patients diagnosed with breast cancer in our hospital but whose treatment was continuing in another center were excluded from the study. Locoregional recurrence was defined as the involvement of ipsilateral axillary, internal mammary, or supraclavicular lymph nodes and/or skin or subcutaneous tissue with or without ipsilateral breast parenchyma involvement. Disease-free survival (DFS) was defined as the time from the moment of diagnosis to the moment of detecting a local recurrence or metastasis. Overall survival (OS) was defined as the time elapsed from the time the patient was diagnosed to the last visit or death.

Statistical analysis

Statistical evaluations were performed on computers using the SPSS 24 statistics software. Descriptive statistics were given as mean±standard deviation or median with the interquartile range (IQR), minimum maximum [min-max] depending on the distribution of the continuous

variables, while categorical variables were summarized as numbers and percentages. The normality test of the numerical variables was controlled by the Kolmogorov-Smirnov test. Wilcoxon Signed Ranks test was used to compare dependent continuous variables. Relationships between the variables to DFS and OS were assessed by the Kaplan-Meier survival analysis. The level of significance was established at $P<0.05$.

RESULTS

All three pieces of receptor information could be accessed in 502 out of the 523 patients who were operated on between 2016 and 2021, 45 (8.6%) of these patients were triple negative. The mean age of the patients was 58.9±15.2 years (27-90), and the mean BMI was 30.4±5.17 (21.5-40.6). The median follow-up time was 30.1 (21.5) months (6-60).

It was observed that the most common histological subtype was invasive ductal carcinoma (IDC), where two patients had medullary carcinoma, and 3 patients with IDC had medullary features. In one patient, an osteoclast-like giant cell carcinoma and an invasive ductal component were observed. At the time of diagnosis, 11 patients were stage 1 (24.4%), 31 patients were stage 2 (68.8%), 2 patients were stage 3 (4.4%), and 1 patient was stage 4 (2.2%). Mastectomy was the most common surgical procedure. Only 2 (4.4%) of the patients had grade 1 tumors. Ki 67 value was reached in 39 patients, and in 29 (74.4%) of these patients, Ki-67 was observed to be 30 and above. The tumors were 25 (55.6%) upper outer quadrant, 9 (20%) upper inner quadrant, 4 (8.9%) lower outer quadrant, and 3 (2.2%) lower inner quadrant. Tumor localization in the breast was 53.3% on the right side. The demographic data and tumor characteristics of the patients are summarized in table 1.

In total, two patients were not given adjuvant therapy due to advanced age and comorbidities, 10 patients (22.2%) received neoadjuvant chemotherapy (NACT), and all other patients received adjuvant chemotherapy. It was observed that 8 (80%) of the patients who received NACT were clinically positive for lymph nodes, and two patients with N0 had T2 tumors. One patient with N0 underwent breast-conserving surgery (BCS) after treatment, and the other patient underwent mastectomy and reconstruction with implants due to lack of response after treatment. In three patients (30%), axillary lymph node dissection (ALND) was performed without sentinel lymph node biopsy (SLNB) after treatment. It was observed that SLNB was applied to the remaining seven (70%) patients and in 3 of them, ALND was performed due to the detection of one metastatic lymph node. It was observed that 4 (50%) of the patients with clinically



positive lymph nodes had a complete axillary response, 3 (37.5%) had 1-3 LN positivity, and one (12.5%) had 4 LN positivity despite the treatment. It was observed that the median tumor diameter before treatment was 30 mm (3.25) [23-50] and after treatment 14.5±16.0 mm (0-50) and was statistically significantly regressed

($P=0.008$). Overall, 4 (40%) patients who received NACT had a complete pathological response (grade 5 according to the Miller-Payne rating), 2 (20%) patients had no change in tumor size, and 4 (40%) patients had a partial response.

Table 1. Patients and tumours characteristics.

Variables	N(%) (Min-Max)
Age† (years)	
<50 years	14 (31.1)
≥50 years	31 (68.9)
Menopausal Status†	
Pre-menopause	28 (62.2)
Post-menopause	17 (37.8)
Breast Density†	
A	8 (17.8)
B	17 (37.8)
C	15 (33.3)
D	5 (11.1)
BMI†	
<30	24 (53.3)
≥30	21 (46.7)
Site†	
Right	24 (53.3)
Left	21 (46.7)
Surgery†	
Mastectomy	31 (54.5)
BCS	14 (45.5)
ALND†	
Yes	24 (53.3)
No	21 (46.7)
ALN number¶	24±15.9 (4-38)
Metastatic ALN number‡	1(1.25) (1-5)
Histological Type†	
IDC	42 (93.3)
Noroendokrin Carsinom	1 (2.2)
Meduller Carcinoma	2 (4.4)
PNI†*	
Yes	7 (17.5)
No	33 (82.5)
LVI†*	
Yes	14 (35.0)
No	26 (65.0)
Grade†	
I	2 (4.4)
II	20 (44.4)
III	23 (51.1)
Clinical T Stage†	
1	12 (26.7)



Variables	N(%) (Min-Max)
2	29 (64.4)
3	2 (4.4)
4	2 (4.4)
Clinical Stage†	
1	11 (24.4)
2	25 (55.6)
3	3 (17.8)
4	1 (4.4)
Ki-67‡*	50 (30) [10-90]

¶: mean±standard deviation [minimum-maximum], ‡:median (interquartile range) [minimum-maximum]. †: n (%), *: less than 45 patients, IDC: Invasive ductal carcinoma, PNI: Perineural invasion, LVI: Lymphovascular invasion, BCS: Breast-conserving surgery, ALND: Axillary lymph nodes dissection, ALN: Axillary lymph nodes, BMI: Body mass index

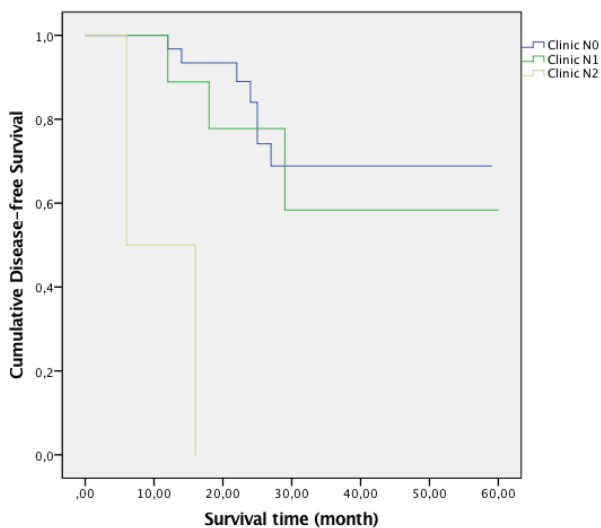


Figure 1. The association of cN stage with disease-free survival (Kaplan-Meier).

Metastasis developed in 11 of the patients (24.4%); it was observed that liver metastasis developed in the follow-up of a patient who had bone metastatic disease at the beginning and a locally advanced disease. It was observed that the most common metastatic sites were bone and brain metastasis, where three patients had brain and lung metastases, and one patient had bone and lung metastases. The mean time from diagnosis to metastasis was 20.7 ± 5.75 (12-29) months. It was observed that 7 (15.6%) patients who died were all metastatic, 5 had brain metastases, one had lung and bone metastasis, and the other had bone and liver metastases. Two patients with isolated bone metastasis, one patient with mediastinal lymph node metastasis, and one patient with liver metastasis, were also observed. A patient with liver metastasis was treated with Cyclophosphamide-anthracycline + taxane. Still, the tumor did not regress, the patient

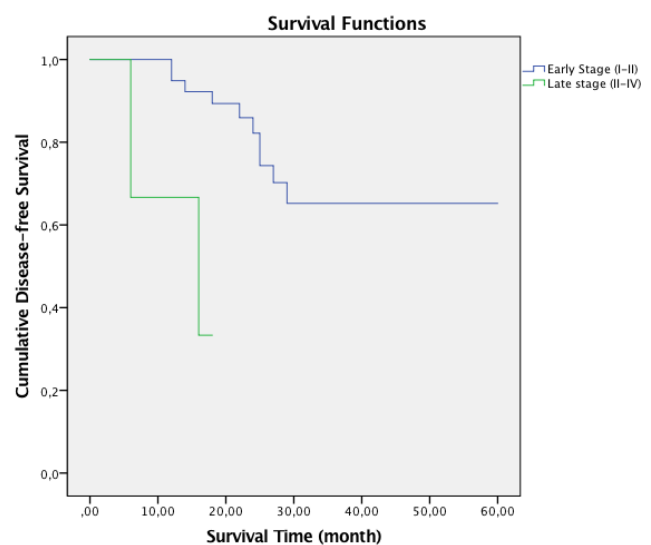


Figure 2. The association of the clinical-stage with disease-free survival (Kaplan-Meier).

developed extensive liver metastases in the 12th month, and the metastases disappeared under gemcitabine treatment and were disease-free in the 40th month of follow-up. It was observed that local recurrence was observed in only two patients. One of them was a patient with lung and bone metastases, and the other patient received NACT for clinical N1 disease. SLNB was applied to the second patient after the treatment, and 4 lymph nodes were sampled, and dissection was not performed because all four were negative. However, it was observed that 7 lymph nodes were dissected, and 2 metastatic lymph nodes were found in the patient who underwent axillary dissection due to axillary recurrence at the 12th month, and was disease-free in the 39th month of the follow-up. The treatment and metastasis regions of the patients are shown in table 2.

The 3-year DFS was 62% and the 3-year OS was 70%. The overall predicted DFS time is $[45.2 \pm 3.5$

**Table 2.** Treatment modalities and recurrence sites.

Variables	N (%)
	10 (22.2)
	33 (73.3)
Cyclophosphamide-anthracycline	8 (17.8)
Cyclophosphamide-anthracycline + taxane	35 (77.8)
Radiotherapy	37 (82.2)
Metastasis Sites	
Bone	3 (36.4)
Brain	5 (45.5)
Liver	2 (18.2)
Lung	4 (36.4)
Mediastinal Lymph Node	1 (9.1)
Multiple	5 (45.5)

months (38.4-52.0)], in the group with clinical N stage 2 [11.0±5.0 months (1.2-20.8)], the DFS time predicted from N stage 1 [44.0± 7.4 months (29.6-58.4)] months and N stage 0 [47.5± 3.7 months (40.4-54.7)] group was significantly shorter ($P=0.000$) (Figure 1). The predicted DFS time in the clinical early stage (I-II) group [46.8±3.5 months (40.0-53.6)], from the group with clinical late stage (III-IV) [13.3±3.0 months (38.4-52.0)] was significantly longer ($P=0.002$) (Figure 2).

The overall predicted OS time is [50.9± 3.0 months (45.1-56.7)], in the clinical early stage I-II group 52.7±2.9 months (47.1-58.4), from the group with clinical late stage III- IV [31.3±8.9 months (14.0-48.7)] was significantly longer ($P=0.008$) (Figure 3). In the group with clinical N stage 2 [27.5±11.5 months (5.0- 50.0)], the OS time predicted from N stage 1 [54.7±4.9 months (45.1-64.2)] months and N stage 0 [51.5±3.3 months (45.0-57.9)] group was significantly shorter ($P=0.021$) (Figure 4).

DISCUSSION

Breast cancer is the most common malignancy in women, and it is a highly heterogeneous disease. Epidemiological data show that TNBC occurs mostly in premenopausal young women below the age of 50, which accounts for approximately 10-20% of all breast cancer patients.^{10, 11} In studies conducted in Turkey, this rate has been reported to be between 12 and 27%.⁷⁻⁹ In our series, 62.2% of the patients were in the premenopausal period, but only 14% were 50 years and younger; TNBC prevalence was found to be lower, unlike the literature, standing at 8.6%.

In the literature, body weight and BMI have been defined as risks for TNBC.^{12,12} Young women in the premenopausal period showed a 5% increase in risk per 5 kg increase in body weight and a 16% increase in risk per 5 kg / m² increase in BMI and are associated with a worse prognosis. In our series, it was found that 84% of the patients had a BMI of 25 and above.^{12, 13} In the literature, the relationship between mammographic density and breast cancer has been investigated, showing that women with high mammographic density are more likely to develop breast cancer in their lifetime compared to women with low MD.¹⁴ However, the study by Mema *et al.*¹⁵ showed that women with entirely fatty breasts on mammography had increased odds of having TNBC compared to women with higher mammographic density. In another study, high mammographic density was associated with recurrence in patients treated for early-stage TNBC.¹⁶ Premenopausal higher MD is associated with higher subsequent risk of ER-negative than ER-positive cancer, whereas postmenopausal higher MD is associated with similar risk of both ER subtypes. In addition, the combination of obesity and higher breast density in premenopausal women is also associated with a higher risk of ER-negative cancer.^{17, 18} A limitation of our study includes the fact that we recruited a small sample size, and given the limited number of patients, we were not able to perform an analysis regarding these factors or to make any comparison with other subtypes.

In general, data from the literature show that 9-32% of patients with TNBC are germline BRCA (gBRCA) mutation carriers.¹⁹ In another study involving 802 TNBC patients with no family history of breast or ovarian cancer, the prevalence of gBRCA

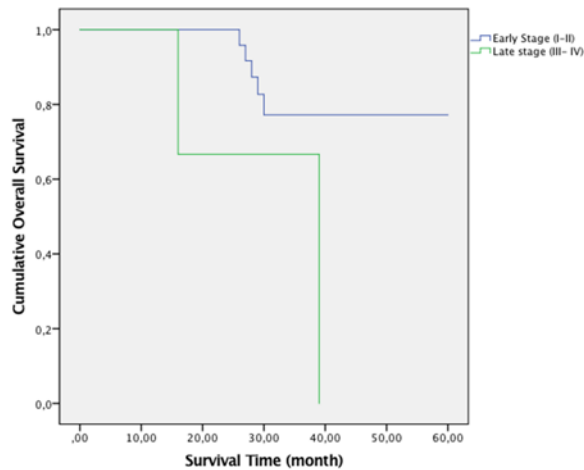


Figure 3. The association of the clinical-stage with overall survival (Kaplan-Meier).

mutations was 16 %.²⁰ In patients under the age of 40, this rate was reported to rise to 24 %.²¹ A meta-analysis involving 46870 TNBC patients concluded that patients with the gBRCA1 mutation were 3 and 9 times more likely to have TNBC compared to gBRCA2 carriers and non-carriers, respectively.²² Of the 30 patients whose family history could be accessed, only 3 (6.7%) patients had a family history of breast cancer, and only 8 patients had a germline mutation analysis. It was observed that only one patient had a BRCA2 mutation and had a family history of breast cancer. This patient underwent a skin-sparing mastectomy, and a prophylactic mastectomy for the contralateral breast. The National Comprehensive Cancer Network (NCCN) guidelines recommend the gBRCA test for all TNBC patients diagnosed at ≤ 60 years of age, regardless of their family history, and breast cancer patients diagnosed at any age with a strong family history.²³

Ki-67 guides the planning of treatment for luminal-like breast cancer, but its role for TNBC is unclear. In a study involving 800 early TNBCs, it was stated that it was an almost independent prognostic and predictive factor for both DFS and OS, and the cut-off value for Ki-67 level in prognosis was found to be 30 %.²⁴ In our study, the Ki-67 level of 86.7% of the cases was known, and the total of 64.4% was 30 and above. However, in our study, no difference was found in survival according to Ki-67 levels.

TNBC is not sensitive to endocrine or molecular targeted therapy due to its specific molecular phenotype. Therefore, chemotherapy is the main systemic therapy, but the effectiveness of conventional postoperative adjuvant chemoradiotherapy is poor. The remaining metastatic lesions will eventually lead to tumor recurrence. Approximately 46% of TNBC patients are known to develop distant metastases, have been shown to have weaker DFS and OS than other

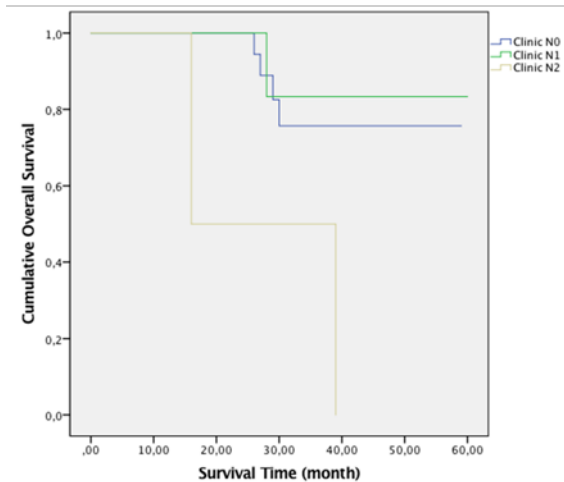


Figure 4. The association of cN stage with disease-free survival (Kaplan-Meier).

breast cancer subtypes, and the mortality rate within the first 5 years after diagnosis is 40 %.^{4, 25} Distant metastases usually occur 3 years after diagnosis, after which the median survival time is only 13.3 months, with a post-operative recurrence rate of up to 25 %.²⁶ Unlike bone and visceral metastases in luminal-like tumors, distant metastases usually involve the brain and lungs. In our series, the median survival of 11 metastatic cases after metastasis was 10 months (9.3-23), and the most common metastatic region was brain and bone metastasis. Patients with isolated bone metastases had a survival advantage.

Although NACT is the standard treatment for locally advanced breast cancer, pathological complete response (pCR) rates for TNBC are associated with better DFS and OS.²⁷ In our series, it was seen that 80% of the patients who received NACT had clinical N1 disease, and the other two patients had T2 tumors (tumor diameter was over 3 cm in both cases), and NACT was planned due to tumor size/breast ratio. It was observed that 4 patients with pathological complete response did not show recurrence. Unlike the survival advantage in those with pathological complete responses, unresponsive cases are associated with worse outcomes.^{28, 29} In our series, it was observed that one of the two patients who had no response after NACT had T2N0 disease, and metastases disappeared after the capecitabine treatment given due to liver metastases in the 12th month and was in the follow-up at the 5th month. It was observed that the other patient had T2N1 disease and was receiving capecitabine due to bone metastasis in the 29th month of the follow-up and still receiving treatment. CREATE-X has recently shown a survival benefit in using capecitabine as adjuvant therapy for TNBC patients unable to achieve pCR.³⁰ In its 2021 guideline, the American Society of Clinical Oncology recommends NACT with an anthracycline and taxane-containing regimen for



patients with TNBC with the clinically node-positive disease as well as at least T1c disease.³¹ One of the advantages of NACT treatment is to see the effectiveness of chemotherapy in breast cancer and to guide the use of adjuvant therapy. Despite the limited number of cases, NACT is beneficial in determining adjuvant therapy in metastatic cases. The limitations of this study are the limited number of patients and the short follow-up period. Another limitation is the low number of patients receiving NACT for this group, for which NACT is especially recommended due to its survival advantage. In our clinic, NACT is considered a priority in early-stage disease as recommended by current guidelines.

CONCLUSION

TNBCs are cancers with varying prevalence, poor prognosis, and limited treatment alternatives. The

TNBC rates of the patients in our study were lower than the data from our country and consistent with the literature, and the lymph node stage was related to poor OS and DFS.

FINANCIAL DISCLOSURE

The authors declared that this study has received no financial support.

CONFLICT OF INTEREST

The authors have no conflicts of interest to declare.

ETHICAL CONSIDERATION

Ethical committee approval was obtained for this study.

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