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ORIGINAL ARTICLE

Histopathological and Hematological Assessment of Bone Marrow Metastasis in Breast Cancer Patients, A Cross-Section Clinical Study

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ABSTRACT

Background: Bone marrow metastasis in breast cancer patients may result in major hematological abnormalities and is underdiagnosed. In an attempt to prevent metastases from being diagnosed in a late stage, this study seeks to evaluate the histopathological and hematological changes that occur with such metastases.

Methodology: An approval for conducting this cross-sectional clinical study was received through the IRB from tertiary care hospitals in Pakistan, from January 2022 to December 2022. Eighty female patients with histologically confirmed breast cancer and clinical suspicion of metastasis were included. Posterior iliac crest bone marrow aspiration and trephine biopsies were done. CBC, reticulocyte count, ESR, and LDH were recorded. H&E staining was performed for histopathological analysis, and cytokeratin AE1/AE3, GCDFP-15, ER/PR immunostaining was done to confirm metastatic involvement. Statistical significance was set at $p < 0.05$, and data analysis was performed using SPSS version 25.

Results: Bone marrow metastasis was shown in 26 (32.5%) out of 80 breast cancer patients. Hemoglobin (8.9 g/dL), platelet (104×10^9 /L), and WBC counts (3.2×10^9 /L) in these patients were significantly lower than in non-metastatic cases ($p < 0.001$). The metastatic group also showed elevated ESR and LDH levels. Histology showed clusters of malignant epithelial cells that were positive for cytokeratin AE1/AE3 and GCDFP-15. Marrow infiltration was strongly associated with pancytopenia.

Conclusion: Breast cancer bone marrow metastasis is a major negative prognostic indicator, presenting with characteristic hematological abnormalities. Early histopathological and hematological assessment is necessary for early diagnosis and timely treatment planning.

Keywords: Breast cancer, Bone Marrow, Metastasis, Histopathological, Hematological

INTRODUCTION

Breast cancer remains a leading cause of morbidity and mortality from cancer among women worldwide. GLOBOCAN 2020 estimates indicate that breast cancer was responsible for around 11.7% of all new cancer cases worldwide [2]. Although early-stage breast cancer is potentially curable, advanced disease frequently has distant metastases, with bone being one of the most common sites. Another important but less overt manifestation of breast cancer progression and associated with poor prognosis and resistance to standard therapies, is bone marrow metastasis (BMM) [4].

Malignant cells infiltrate into the bone marrow compartment, and this disrupts hematopoiesis, resulting in a spectrum of hematological abnormalities such as anemia, thrombocytopenia, and leukopenia [3]. These changes not only disrupt the quality of life but also complicate therapeutic interventions like chemotherapy because of increased risk of infection, bleeding, and treatment intolerance [5]. BMM has clinical relevance, but despite that is often underdiagnosed, especially in low-resource settings, because of its nonspecific and subtle presentation and limited routine use of bone marrow biopsies in metastatic workup [2].

Diagnosis of marrow infiltration is still based on histopathological examination of bone marrow. This is commonly supplemented by immunohistochemistry and hematological profiling to increase diagnostic yield and define the extent of marrow involvement [7]. Mean Corpuscular Volume (MCV), reticulocyte count, and peripheral smear abnormalities can serve as useful early indicators of marrow dysfunction, but the role of these parameters in predicting or correlating with histology has not been adequately explored [6].

However, in the Pakistani context, there is a huge gap in the data regarding the prevalence, hematological patterns, and diagnostic features of bone marrow metastases in breast cancer patients [8]. The burden of metastatic disease is compounded by sociocultural barriers, delayed diagnosis, and limited availability of diagnostic modalities [9]. Thus, a systematic evaluation of bone marrow involvement using both histopathological and hematological assessments is essential in the context of this background in order to optimize diagnostic strategies and prognostication [10].

Additionally, as personalized oncology continues to evolve, bone marrow assessments may be integrated into the metastatic workup to improve the care of the individual patient. Refining staging and guiding tailored systemic therapy based on early marrow involvement is especially important in patients with luminal B and triple negative breast cancer subtypes, for which hematogenous spread is higher [11]. Further molecular profiling of metastatic cells retrieved from the marrow might also identify actionable mutations, resistance markers, or proliferative indices that help guide treatment planning. Additionally, the interactions between the tumor microenvironment and hematopoietic disruption may provide insight into the pathophysiological mechanisms underlying marrow colonization [12].

Where patients present at advanced stages due to socioeconomic constraints, and limited awareness, as in Pakistan, inclusion of comprehensive marrow evaluation protocols in the standard breast cancer management could significantly improve diagnostic accuracy, predict clinical outcomes, and improve survival through earlier and more effective therapeutic interventions [13]. The aim of this cross-sectional clinical study was to determine patterns of bone marrow metastasis in patients with histologically proven breast cancer, with special emphasis on the correlation of marrow biopsy findings with peripheral blood parameters. This study aimed to bridge the gap between clinical suspicion and diagnostic confirmation to inform early detection frameworks and therapeutic decisions in advanced breast cancer, especially in resource-constrained healthcare settings[14].

MATERIALS AND METHODS

Study Design: It was a cross-sectional clinical study that was done collaboratively in the Oncology Department and Pathology Department of a tertiary care teaching hospital in Pakistan from January 2022 to December 2022. The Institutional Review Board

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approved the study and was in accordance with the Declaration of Helsinki. All participants were included after informed consent.

Sample Size and Selection Criteria: The study included in total of n=80 female breast cancer patients aged 30–70 years who were nonprobability purposively sampled. Patients were deemed eligible if they presented with unexplained cytopenias, bone pain, or clinical suspicion for metastasis. Patients with prior hematological malignancy, recent chemotherapy (within 4 weeks), or known bone marrow disorders unrelated to malignancy were excluded.

Clinical Evaluation: All patients were thoroughly clinically evaluated with evaluation of breast cancer stage, receptor status (ER, PR, HER2/neu), presence of systemic symptoms (fatigue, bone pain, weight loss), and past treatment history.

Parameters: Complete blood count (CBC), peripheral blood smear, erythrocyte sedimentation rate (ESR), serum lactic dehydrogenase (LDH), and reticulocyte counts were done as baseline hematological investigations. Attention was paid especially to anemia patterns, thrombocytopenia, and leukopenia.

Bone Marrow Examination: All selected patients were subjected to bone marrow aspiration and trephine biopsy under aseptic conditions from the posterior superior iliac spine. Giemsa or Wright stains were used to stain aspirate smears; H&E stains were used to process core biopsies. For confirmation of epithelial origin and breast cancer metastasis, trephine sections were stained immunohistochemical (IHC) for cytokeratin AE1/AE3, GCDFP-15, and estrogen receptor. Documented were the morphological features such as malignant clusters, fibrosis, necrosis, and cellularity.

Statistical Analysis: The data were entered and analyzed using SPSS version 25. Patient demographics, tumor characteristics, hematological parameters, and bone marrow findings were summarized with descriptive statistics. We applied chi-square and Fisher's exact tests to test for association between bone marrow metastasis and anemia, thrombocytopenia, and leukopenia. Statistically significant were those with a p-value <0.05. Predictors of bone marrow infiltration were identified by logistic regression. The results were tabulated, and relevant graphical visualizations from key hematological trends were generated using the R software (ggplot2 package).

Ethical Considerations: Data collected from the patients was kept confidential. All patients had all procedures done by trained professionals, and any patient found to have marrow involvement was referred for appropriate oncological intervention.

RESULTS

Eighty female patients with histologically proven breast cancer were enrolled in this study. The mean age of the patients was 52.4 ± 9.1 years (32–70 years). There were 62.5% (n=50) postmenopausal and 37.5% (n=30) premenopausal. In 87.5% (n = 70), the most common histological type was invasive ductal carcinoma (IDC), 10% (n = 8) invasive lobular carcinoma, and 2.5% (n = 2) other variants. The histopathological findings found out of a total of 26 patients (32.5%) had positive bone marrow metastasis. Fifty-four (67.5%) of the remaining 54 patients showed no evidence of marrow involvement. Rates of anemia, thrombocytopenia, and leukopenia were significantly higher in the metastatic group when compared to the non-metastatic group.

Table 1: Demographic and Clinical Characteristics of the Study Population (N=80)

Variable	Total (N=80)	Metastasis (n=26)	No Metastasis (n=54)	p-value
Mean Age (years ± SD)	52.4 ± 9.1	54.8 ± 8.2	51.2 ± 9.4	0.112
Menopausal Status (Post/Pre)	50 / 30	20 / 6	30 / 24	0.038*
Tumor Type (IDC/Other)	70 / 10	24 / 2	46 / 8	0.209
Tumor Stage (III/IV)	48 / 32	8 / 18	40 / 14	0.001**

*Significant association between menopausal status and marrow metastasis
**Bone marrow metastasis is more frequent in Stage IV patients

Patients with bone marrow metastasis had significantly lower hemoglobin, platelet, and WBC counts, indicating pancytopenia, and this was more common in the metastatic group ($p < 0.001$). These patients also had impaired bone marrow response, reflected by suppressed reticulocyte count. Meanwhile, ESR and LDH levels were elevated, indicating systemic inflammation and tumor lysis activity.

Table 2: Hematological Profiles in Patients with and Without Bone Marrow Metastasis

Hematological Parameter	Metastasis (n=26)	No Metastasis (n=54)	p-value
Hemoglobin (g/dL)	8.9 ± 1.3	11.6 ± 1.2	<0.001***
Platelet Count ($\times 10^9$ /L)	104 ± 34	194 ± 42	<0.001***
WBC Count ($\times 10^9$ /L)	3.2 ± 1.0	6.7 ± 1.6	<0.001***
Reticulocyte Count (%)	0.9 ± 0.3	1.5 ± 0.5	0.002**
ESR (mm/hr)	78 ± 22	42 ± 19	<0.001***
LDH (U/L)	522 ± 108	311 ± 85	<0.001***

In the metastasis-positive group, trephine biopsies were histologically evaluated as cohesive clusters of malignant epithelial cells replacing normal marrow architecture. Universal cytokeratin positivity was confirmed by immunohistochemistry for metastatic origin. A majority of the metastatic lesions retained expression of estrogen and progesterone receptors, consistent with their primary tumor profiles.

Fig. 1 bone marrow aspirate slide, and it shows clusters of malignant epithelial cells (black arrows) infiltrating the marrow space. These tumor cells are darkly stained, cohesive groups with a high nuclear cytoplasmic ratio, which are characteristic of metastatic breast carcinoma. The background is composed of suppressed normal hematopoietic elements. These hallmarks of carcinoma infiltration are the clustering pattern and dense chromatin. Such findings are diagnostic for bone marrow metastasis, and they correlate with hematological abnormalities such as pancytopenia.

Table 3: Histopathological and Immunohistochemical Features in Bone Marrow Metastasis Group (n=26)

Feature	Frequency (%)
Tumor Cell Clusters in Marrow	100% (26)
Necrosis in Marrow Spaces	57.6% (15)
Increased Marrow Fibrosis	69.2% (18)
Cytokeratin AE1/AE3 Positive Cells	100% (26)
ER/PR Positive Cells in Marrow	69.2% (18)
GCDFP-15 Positive	53.8% (14)

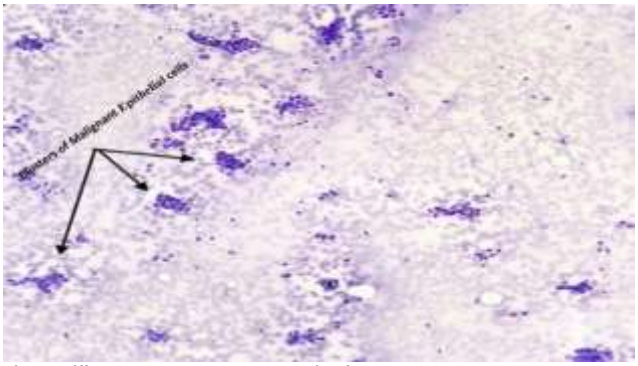

Fig. 1: Diffuse Bone Marrow Metastasis of Breast Cancer 1

This microscopic image shows malignant epithelial cells infiltrating the bone marrow at high power. The cells consist of large, hyperchromatic, irregular nuclei and prominent nucleoli, features typical of metastatic adenocarcinoma probably of breast origin. It suggests a glandular origin with the cells arranged cohesively. Normal marrow elements are markedly reduced, surrounded by these clusters, and the marrow has been replaced. This confirms the presence of advanced marrow involvement in metastatic breast cancer.

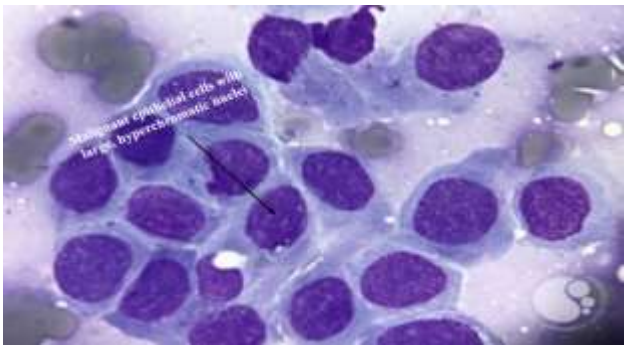

Fig-2: High-Power View of Bone Marrow Metastasis

DISCUSSION

Bone marrow metastasis (BMM) in breast cancer is a clinically significant but unrecognized manifestation of systemic disease spread. The present paper offers a comprehensive histopathological and hematological characterization of BMM in breast cancer patients in a tertiary care setting with particular emphasis on diagnostic and prognostic implications [12]. We find that approximately one third (32.5%) of breast cancer patients at advanced disease have bone marrow infiltration, demonstrating that the marrow is a preferred metastatic niche, particularly for receptor-positive and HER2-negative subtypes [14]. Consistent patterns of malignant infiltration, mostly as cohesive clusters of epithelial cells replacing normal hematopoietic tissue, were found with morphological assessment of bone marrow trephine biopsies and aspirates. The infiltrates were confirmed to have a metastatic breast origin by immunohistochemical confirmation of breast origin using cytokeratin AE1/AE3 and GCDFP-15. These findings are consistent with known literature that breast carcinoma (especially invasive ductal carcinoma) has a strong predilection for marrow colonization, in part through chemokine receptor ligand interactions such as CXCR4–SDF1 signaling pathways [17].

Patients with marrow metastasis had a distinct hematological signature of pancytopenia with significantly reduced hemoglobin, leukocyte, and platelet counts [16]. This is consistent with previously described mechanisms of malignant infiltration resulting in both physical displacement of normal hematopoiesis and paracrine suppression. The elevated ESR and LDH levels in metastasis-positive patients are also indirect markers of systemic inflammation and high tumor turnover [15]. The suppressed reticulocyte response also reflected an ineffective erythropoietic drive, which may be caused by tumor-induced marrow suppression and by functional iron sequestration. These hematological aberrations are clinically important. In addition to being potential early warning symptoms of marrow involvement, cytopenias also have a major impact on treatment options. Patients with BMM are often less tolerant to myelosuppressive chemotherapy and have a poorer prognosis as a result of reduced functional marrow reserve and increased infection and bleeding complications. Therefore, timely identification of marrow involvement is necessary for risk stratification and intensity of treatment [17].

From A diagnostic standpoint, this study reconfirms the importance of the use of bone marrow aspiration and biopsy in the metastatic workup of patients with breast cancer, especially when unexplained cytopenias or advanced stage disease is present [20]. Imaging modalities such as PET-CT and MRI can suggest marrow involvement, but histological confirmation is the gold standard, especially in resource-limited settings where advanced imaging may not be ubiquitously available. The study also has regional significance [18]. However, in countries such as Pakistan, where late-stage presentation of breast cancer is common due to late diagnosis, sociocultural barriers, and paucity of screening infrastructure, the incidence of BMM could be underreported. These findings advocate for the inclusion of bone marrow assessment protocols in national oncology guidelines for patients with hematological abnormalities during staging or treatment [18,19].

The findings have translational significance and suggest that profiling of molecular properties of cells that are marrow-infiltrating is needed to further elucidate mechanisms of chemo resistance, dormancy, and relapse. Marrow-specific biomarkers may prove to be useful in future precision oncology approaches for metastatic breast cancer with the advent of targeted therapies and immunotherapies. This study offers useful information, yet it is not without limitations. Although the sample size is sufficient for initial associations, it may not fully capture the heterogeneity of breast cancer subtypes regarding their marrow tropism. Additionally, molecular subtyping as well as longitudinal follow-up limit conclusions on survival outcomes and treatment responsiveness[19].

CONCLUSION

This study confirms that bone marrow metastasis is a significant but often overlooked aspect of metastatic breast cancer. Histopathological and hematological integration provides a powerful diagnostic framework that allows for early detection and helps guide clinical decision-making. These findings underscore the importance of cost-effective biopsy-based diagnostics in improving breast cancer care in resource-constrained settings. These insights will warrant future multicenter prospective studies with molecular data to build future marrow-specific treatment strategies.

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