

## **Analysis of Morphological Changes in Mast Cells within the Microenvironment of Breast Cancer**

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**Abstract:** This article examines the expression of mast cells in the tumor microenvironment of 50 patients diagnosed with breast cancer, using the immunohistochemical method, based on morphological luminal subtypes. The study focuses on Luminal A, Luminal B Her2/neu-negative, Luminal B Her2/neu-positive (non-luminal type), and Basal-like (triple-negative) subtypes, with 10 patients analyzed for each subtype to assess mast cell activity.

The study investigates the activity of mast cells around tumor blood vessels according to the indicators of luminal subtypes, confirming differences among the subtypes. The results show that the density of blood vessels and mast cells around tumor cells varies: in the Luminal A subtype, mast cells numbered 20–30 per field of view; in the Luminal B Her2-positive subtype, 60–80; in the Her2-positive (non-luminal) subtype, 100–150; and in the Basal-like (triple-negative) subtype, 200 or more.

These findings indicate that the activity level and aggressiveness of breast cancer are associated with the luminal subtype of the tumor and the identified density of mast cells. Mast cells play a role in the microenvironment of breast cancer and have prognostic significance. This highlights their importance as a factor in determining treatment strategies and studying prognosis.

**Keywords:** breast cancer, Luminal A, Luminal B Her2-negative, Luminal B Her2-positive, Her2-positive (non-luminal type), Basal-like, triple-negative subtype, blood vessel density, mast cells.

**Introduction. Mast cells** (also called mastocytes or labrocytes) are tissue basophils that belong to the myeloid lineage, containing basophilic granules in their cytoplasm that store histamine and heparin. Unlike basophils, which also contain basophilic granules, mast cells typically do not enter the bloodstream. Mast cells play a key role in inflammation, immediate (Type I) hypersensitivity reactions, defense against multicellular parasites and other pathogens, the formation of the blood-brain barrier, and other physiological processes.

Mast cells are actively involved in the development of allergies and anaphylactic shock. These cells are present in most tissues, usually located near blood vessels and nerves. Additionally, due to specific receptors on their surface, mast cells can be activated by certain components of the complement system. Histamine, a significant component of mast cell granules, causes post-capillary venules to dilate, activates the endothelium, and increases vascular permeability.

In the human brain, mast cells are present and interact with the neuroimmune system. They are found in structures involved in transmitting visceral sensory signals (e.g., pain), performing neuroendocrine functions, and in the blood-brain barrier. Mast cells are located in regions such as the pituitary gland, pineal gland, thalamus, hypothalamus, brainstem, choroid plexus, and areas of the cerebral cortex.

According to A.F. Lazarev et al. (2011), mast cells are closely associated with tumor neoangiogenesis. These cellular elements influence angiogenesis through various mechanisms, including factors such as VEGF, bFGF, TGF- $\beta$ , TNF- $\alpha$ , IL-1, IL-6, and IL-8.

The number and phenotype of mast cells within a tumor correlate with the degree of tumor malignancy. For instance, in gastric cancer, the level of angiogenesis increases in parallel with tumor malignancy, reaching its peak at stage IV. However, data on the relationship between mast cells and tumor metastasis remain inconsistent. Such correlations were not found in breast cancer[14] or squamous cell carcinoma of the oral cavity[18], but were observed in gastric cancer[20].

Increased mast cell activity is associated with tumor cell invasion into lymphatic and blood vessels, significantly enhancing tumor progression. In renal cancer, the microvascular density (MVD) and the number of mast cells within the tumor showed a correlation ( $r = 0.30$ ). Additionally, both MVD and mast cell density were higher in the peritumoral zone compared to the tumor center.

Understanding the mechanisms of angiogenesis can provide a rational basis for anti-angiogenic therapy for malignant diseases. Based on the role of mast cells in tumor neoangiogenesis, ongoing research is exploring their potential as therapeutic targets in cancer treatment[13,15,19].

The study of mast cells is particularly relevant for precancerous conditions. Their density increases with tumor invasion and metastasis. Furthermore, mast cell distribution is significantly associated with critical clinical and morphological tumor parameters, as well as long-term survival rates. Thus, mast cell activity can serve as an additional prognostic factor for malignancy. The morphological and phenotypic characteristics of mast cells in the peritumoral zone are equally important for analysis.

In breast cancer, the prognostic value of tumor microenvironment parameters varies depending on the tumor's molecular genetic subtype. For example, high levels of lymphocytic infiltration in tumors are an integral marker of the tumor microenvironment and are considered unfavorable in triple-negative breast cancer[11].

The phenomenon of morphological heterogeneity, first described by V.M. Perelmuter and M.V. Zavyalova in 2006, has long served as a model for studying various tumor traits such as chemoresistance and invasiveness, as well as for predicting breast cancer progression and outcomes[12]. However, the significance of the tumor microenvironment in the formation and characteristics of various morphological structures remains insufficiently explored.

In conclusion, examining the tumor stroma's ability to modify itself in response to tumor cell interactions is of critical importance during tumor progression. This approach provides valuable insights into tumor cell activity and its influence on disease prognosis.

## **Research methods and materials**

An immunohistochemical study was conducted to analyze various processes. Based on immunohistochemical examination criteria, women with breast cancer were classified into Luminal A type, Luminal B type (Her2-negative), Luminal B type (Her2-positive), Her2-positive (non-luminal type), and Basal-like (triple-negative) types. Ten patients were selected from each group, and the level of perivascular proliferative activity of whole cells in breast cancer was studied using a "Leica" microscope (manufactured in Australia) in the Pathomorphology Department of the Republican Specialized Scientific and Practical Medical Center of Oncology and Radiology.

Research methods and objects: 50 patients with breast cancer treated at the Tashkent Regional Branch of the Republican Specialized Scientific and Practical Medical Center of Oncology and Radiology.

The main goal of the study was to study the histological preparations of patients examined and treated at different stages of breast cancer, the expression of Mast cells in microscopic changes around the cell, and the extent of its influence on tumor progression. The age of the patients with breast cancer studied in the study ranged from 35 to 60 years, with an average of 45 years. Of the 50 patients, 20 were examined during menstruation and 30 during menopause. The disease development period ranged from 1 to 24 months. The initial symptoms of the disease in the examined patients were as follows: 50 (100%) patients had a tumor in the breast, 50 (100%) had a tumor and pain in the breast, 16 (30%) had a deformity in the breast, 17 (36%) had a pulsating pulp in the breast, 17 (36%) had a pulsating pulp, and 16 (35%) of 50 patients had clinically enlarged subcutaneous lymph nodes. Breast cancer was localized in the right breast in 26 (60%) and in the left breast in 24 (40%) of 50 patients. When compared to the breast quadrant, 23 (46%) are located in the upper pelvic quadrant 10 (20%), the lower pelvic quadrant 5 (10%), the upper inner quadrant 5 (10%), the lower inner quadrant 3 (6%), and the central quadrant 4 (8%).

When breast cancer was palpated, the tumor size ranged from 2.5 cm to 6.5 cm. Analysis of the clinical characteristics of the disease revealed that among 50 patients, T1N0M0-10 (20%), T2N0M0-10 (20%), T1N1M0-10 (20%), T2N1M0-5 (10%), T3N0M0-5 (10%), T3N1-2M0-5 (10%), T4N0M0-3 (6%), and T4N1-2M0-2 (4%).

All patients were diagnosed clinically and morphologically after conducting clinical, ultrasound, mammographic and histological, and immunohistochemical studies.

According to the results of the histological examination, in 50 patients, 30 (60%) were found to have infiltrative intra-breast cancer, 15 (30%) - infiltrative intra-bundle cancer, and 5 (10%) - nonspecific cancer.

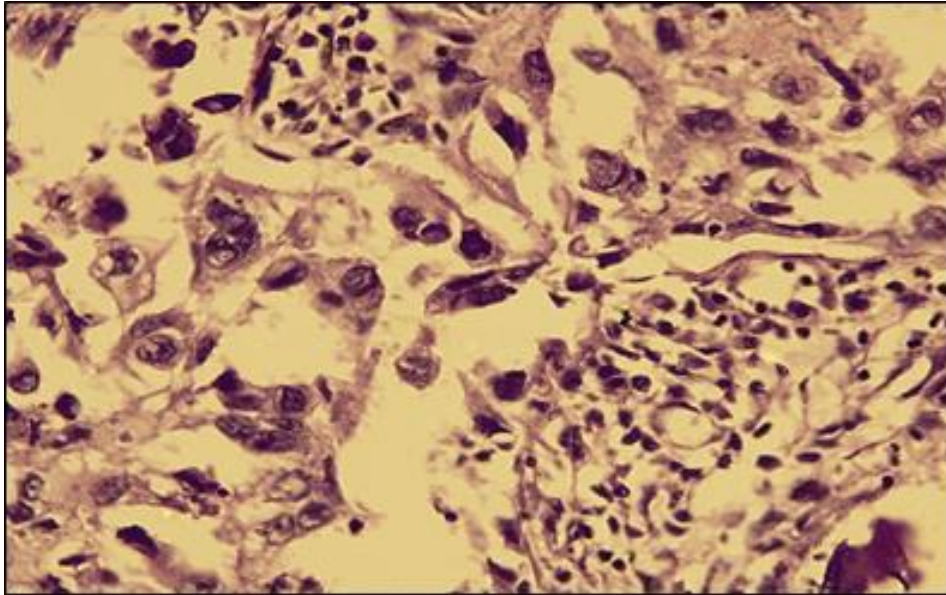
### The results of the study.

Our study investigated the proliferative activity of Mast cells in the vascular wall, the density of their location around the vessels under a microscope in a single field of view of 40°C x 0.65°C. Ten patients were selected from each group based on the criteria.

### Patients selected according to groups based on immunohistochemical criteria

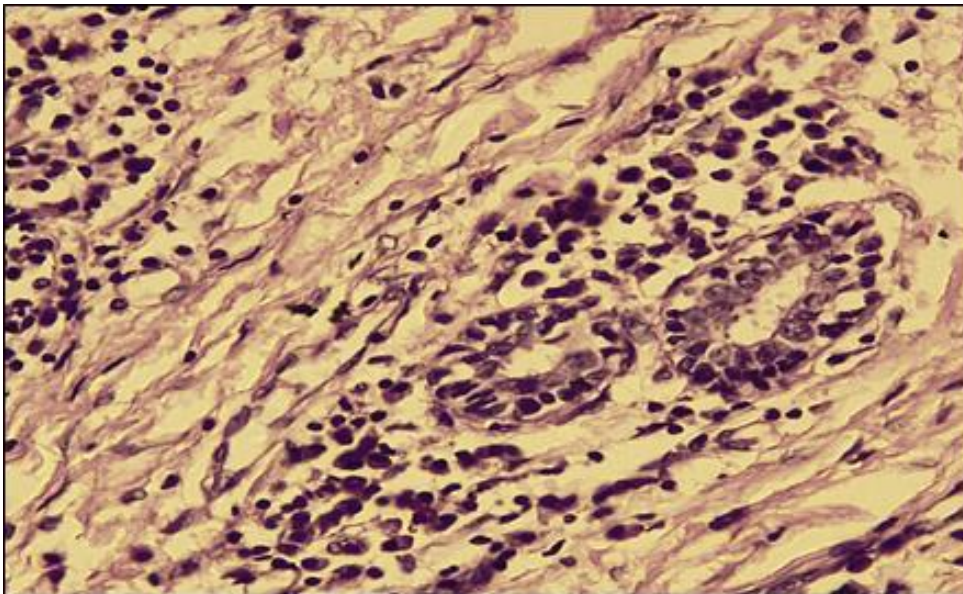
No	Criteria	Patients number
1	Luminal Type A	10
2	Luminal B Type (Her 2-negative)	10
3	Luminal B Type (Her 2-positive)	10
4	Her 2-positive (non-luminal type)	10
5	Basal like (Three times negative)	10

Luminal type A. When we observed the proliferative activity of the Mast cells under a microscope in all 10 selected patients, malignant tumor cells consisting of multiple pathological mitoses with hyperchromic nuclei, polymorphized in the intestines of the mammary gland and internal parts of the epithelium of the channel, were detected in the area of one view at 40°C x 0.65° in all patients. (Figure 1).



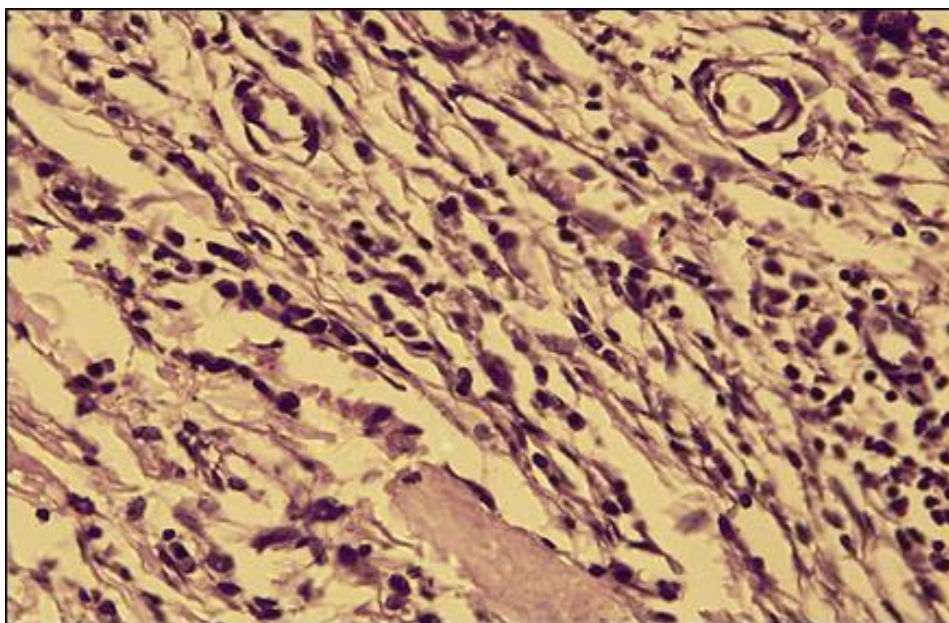
**Figure 1. Proliferation of Mast cells around the vessels in Luminal A type of breast cancer.**

When we observed the proliferative activity of Mast cells under a microscope in all 10 patients selected with Luminal B type (negative), hyperchromic nuclei with polymorphic nuclei in the intestines of the breast and internal parts of the epithelium of the canal were found, and in all patients, 30-40 Mast cells were detected in the area of vision of blood vessels around malignant tumor cells consisting of multiple pathological mitoses. (Figure 2)



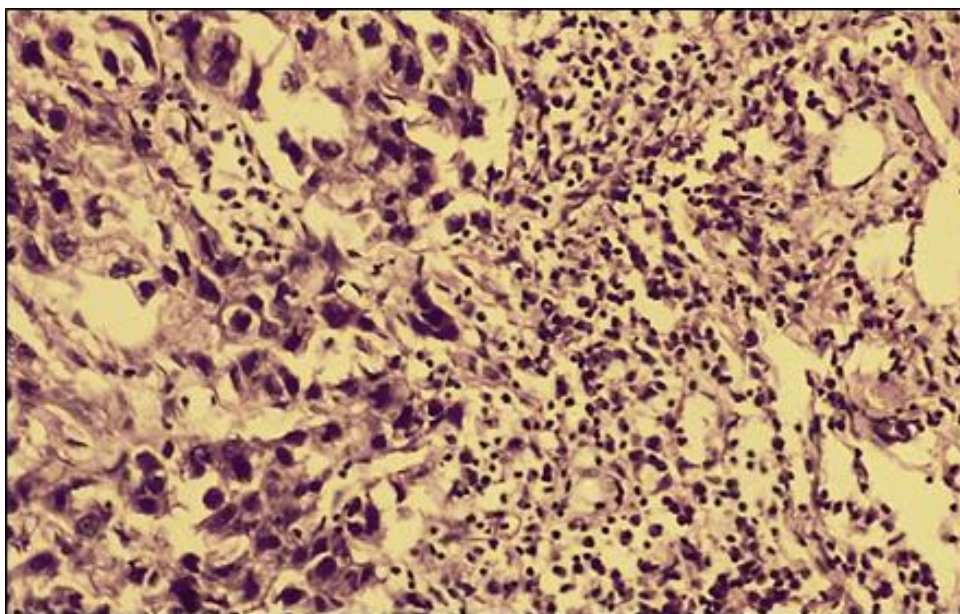
**Figure 2. Luminal B type of breast cancer (Neg2-negative) proliferation of Mast cells around blood vessels.**

Luminal type B (Her 2-positive), when we observed the proliferative activity of Mast cells under a microscope in all 10 selected patients, it was found that blood vessels around tumor cells consisting of multiple pathological mitoses with hyperchromic nuclei undergoing polymorphism in the intestinal sections of the epithelium and in the intestinal sections of the breast were detected in areas of 60-80 Mast cells in all patients. (Figure 3).



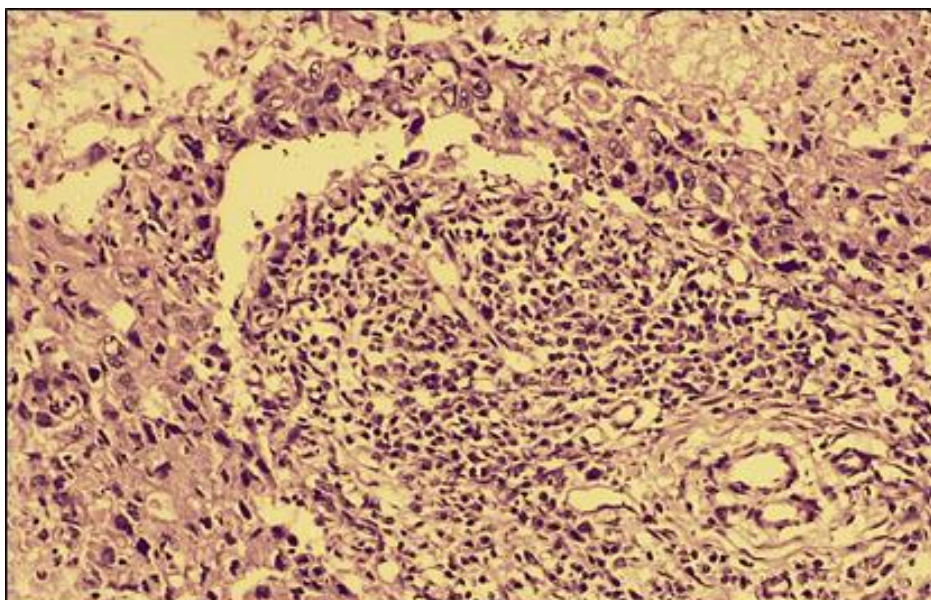
**Figure 3. Luminal B type (Her 2-positive) in breast cancer, proliferation of Mast cells around blood vessels.**

When we observed the proliferative activity of whole cells under a microscope in all 10 selected patients, malignant tumor cells consisting of multiple pathological mitoses with hyperchromic nuclei, polymorphized in the intestines of the breast and internal parts of the epithelium of the canal, were detected in 100-150 cases in the area around the convex vessels in all patients. (Fig. 4).



**Figure 4. Neur2-positive (non-luminal type) proliferation of Mast cells around blood vessels in breast cancer.**

In all 10 patients selected for the basal cell type (three times negative), when we observed the proliferative activity of the Mast cells under the microscope, it was found that in all patients around malignant tumor cells consisting of multiple pathological mitoses with hyperchromic ulcers with polymorphism in the intestines of the breast and internal parts of the epithelium of the canal, in the area around the blood vessels, there were 200 Mast cells and 0.65 Mast cells in one field of view in all patients. (Figure 5).



**Figure 5. Proliferation of Mast cells around blood vessels in the basal like (triple negative) type of breast cancer.**

### **Research Results and Discussion.**

The results obtained showed that according to vascular density, 60-80 Mast cells of the Luminal B type (Her 2-positive) are detected in one field of view, while 100-150 Mast cells of the Her 2-positive (non-luminal type) type are detected in the vicinity of blood vessels, and in the vicinity of the basal cell (three-negative type) tumor, the detection of more than 200 complete vascular cells in the vicinity of the tumor indicates a high level of breast cancer development, a high degree of Our study revealed that the activity of Mast cells in different immunohistochemical forms of breast cancer is different, and its activity is directly related to the luminal type of tumor cells. Mast cells actively participate in the microclimate of breast cancer and have prognostic significance, therefore, the study of Mast cells in breast cancer, its analysis depending on the type of luminal, is of great importance in determining treatment tactics and studying the prognosis.

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