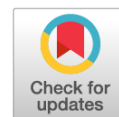


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# Detection of Tumor-Associated Tryptase-Positive Mast Cells in Sporadic Medullary Thyroid Carcinoma

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## ABSTRACT

**BACKGROUND:** There are currently no published studies on the role of mast cells in the pathogenesis of sporadic medullary thyroid carcinoma. However, their involvement in the progression of a variety of epithelial malignant neoplasms has been demonstrated. Furthermore, mast cell count may be an independent predictor of long-term progression-free survival in patients with pancreatic neuroendocrine tumors.

**AIM:** The work aimed to evaluate the potential of immunohistochemical detection of mast cells in the tumor microenvironment in sporadic medullary thyroid carcinoma.

**METHODS:** Histological specimens of sporadic medullary thyroid carcinoma were assessed using immunohistochemical detection of tryptase in mast cells. A convolutional neural network (CNN) model was then trained to segment positively stained cells, followed by quantitative analysis of the results.

**RESULTS:** Several potentially clinically significant parameters were identified, including correlations between mast cell count in the thyroid stroma and age; correlations between intratumoral mast cell count and T stage according to the TNM (8th edition) classification; and patterns of mast cell colocalization with other cells of the tumor microenvironment.

**CONCLUSION:** The study confirmed the presence of mast cells in the stroma of medullary thyroid carcinoma and revealed quantitative differences depending on tumor size. The observed active interactions of mast cells with atypical cells of sporadic medullary thyroid carcinoma and other components of the tumor microenvironment are a significant criterion for interpreting the biological effects of mast cells in this tumor type. These findings warrant further analysis to develop diagnostic algorithms and improve prognostic accuracy.

**Keywords:** medullary carcinoma; mast cells; immune microenvironment; immunohistochemical detection.

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# Возможность выявления триптаза-позитивных тучных клеток, ассоциированных с опухолью при спорадической медуллярной карциноме щитовидной железы

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## АННОТАЦИЯ

**Обоснование.** На сегодняшний день в литературе не описаны исследования, направленные на изучение значения тучных клеток в развитии спорадической медуллярной карциномы щитовидной железы. В то же время доказано их участие в прогрессировании эпителиальных злокачественных новообразований различных локализаций. Кроме того, установлено, что количество тучных клеток может служить независимым предиктором длительного выживания без прогрессирования у пациентов с нейроэндокринными опухолями поджелудочной железы.

**Цель** — оценить возможности иммуногистохимической детекции тучных клеток в опухолевом микроокружении спорадической медуллярной карциномы щитовидной железы.

**Методы.** Исследование проведено на гистологических срезах образцов спорадической медуллярной карциномы щитовидной железы с применением иммуногистохимической детекции триптазы в тучных клетках. Далее проведено обучение модели свёрточной нейронной сети (CNN, convolutional neural network) для сегментации положительно окрашенных клеток с последующим расчётом результатов.

**Результаты.** Выявлен ряд потенциально клинически значимых показателей, таких как: взаимосвязь количества тучных клеток в строме щитовидной железы с возрастом пациентов; корреляция количества тучных клеток в опухоли со стадией Т согласно классификации TNM (8-е издание); особенности локализации тучных клеток с иными клетками опухолевого микроокружения.

**Заключение.** Проведённое исследование подтвердило наличие тучных клеток в строме медуллярной карциномы щитовидной железы и выявило их количественные различия в зависимости от размера узла. Обнаруженное активное воздействие тучных клеток на атипичные клетки спорадической медуллярной карциномы щитовидной железы и другие компоненты опухолевого микроокружения является важным критерием для интерпретации биологических эффектов тучных клеток в отношении данного типа опухоли и заслуживает дополнительного анализа с целью разработки диагностических алгоритмов и повышения объективности прогноза.

**Ключевые слова:** медуллярная карцинома; тучные клетки; иммунное окружение; иммуногистохимическая детекция.

## Как цитировать:

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# 检测与散发性甲状腺髓样癌相关的胰蛋白酶阳性肥大细胞的可能性

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## 摘要

**论证。**目前尚无文献报道针对肥大细胞在散发性甲状腺髓样癌发生与发展中的作用的研究。然而，大量证据表明，肥大细胞参与多种上皮性恶性肿瘤的进展过程。此外，已有研究显示，肥大细胞数量可作为胰腺神经内分泌肿瘤患者无进展生存期的独立预测指标。

**目的：**采用免疫组化方法展示在散发性甲状腺髓样癌肿瘤微环境中检测肥大细胞的可行性。

**方法。**在散发性甲状腺髓样癌的组织石蜡切片上，采用免疫组化方法检测肥大细胞中胰蛋白酶（tryptase）的表达。随后基于获得的染色图像训练卷积神经网络（CNN, convolutional neural network）模型，对阳性染色细胞进行分割与定量分析。

**结果。**研究发现若干具有潜在临床意义的指标，包括：甲状腺间质中肥大细胞数量与患者年龄的相关性；肿瘤组织内肥大细胞数量与TNM分期（第8版）中T分期的关系；以及肥大细胞与肿瘤微环境中其他细胞的共定位特征。

**结论。**本研究证实肥大细胞存在于甲状腺髓样癌的间质中，并显示其数量随肿瘤结节大小而变化。肥大细胞对散发性甲状腺髓样癌异型细胞及肿瘤微环境中其他成分的活跃作用，是解释该肿瘤类型中肥大细胞生物学效应的重要判定标准，值得进一步深入研究，以建立更客观的诊断与预后评估算法。

**关键词：**髓样癌；肥大细胞；免疫微环境；免疫组化检测。

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## BACKGROUND

Medullary thyroid carcinoma (MTC) is a malignant tumor arising from calcitonin-producing parafollicular C cells of the thyroid gland [1]. MTC accounts for approximately 2% of all thyroid malignant neoplasms and is hereditary in about 25% of cases, resulting from autosomal dominant germline gain-of-function mutations in the *RET* proto-oncogene, indicating multiple endocrine neoplasia type 2A or 2B (MEN2A or MEN2B). The etiology of sporadic forms of MTC remains unknown [2].

The clinical course of MTC is currently unpredictable, with phenotypes ranging from indolent to fulminant. The development of an MTC classification incorporating molecular characteristics and progression risk stratification is of critical importance for optimizing treatment algorithms in patients with this carcinoma. According to most international clinical guidelines, surgery is the primary treatment modality, involving total thyroidectomy with central lymph node dissection, which is associated with a relatively high risk of complications [3–8]. Risk stratification of MTC would enable identification of patient groups requiring more aggressive therapy as well as those with more slowly progressing disease who may require only active monitoring. The implementation of molecular stratification would improve prediction of treatment response, minimize treatment-related adverse effects, and enhance overall survival outcomes. Moreover, such a classification would provide a foundation for the development of new targeted therapies directed at specific molecular targets, thereby increasing therapeutic efficacy and improving patient's quality of life. At present, risk stratification is based solely on the proliferative activity index and the presence of tumor necrosis; however, the search for additional biomarkers and disease predictors is ongoing [9].

The role of mast cells (MCs) in thyroid condition has been insufficiently investigated. It is well established that MCs are innate immune cells, and their involvement in the pathogenesis of allergic diseases has been extensively studied [10]. Despite the limited number of investigations, MCs have also been shown to play a remarkable role in thyroid pathological processes. In the study by Zdor et al. [11], a relationship between MC density and various non-neoplastic thyroid lesions was demonstrated. Excessive thyroid hormone exposure induces inflammatory responses within the thyroid gland. Thyroid MCs overexpress the costimulatory molecule CD86, which confirms their involvement in autoantigen presentation [11]. MCs also participate in the regulation of microcirculation and angiogenesis, thereby influencing thyrocyte functional activity. Data reported by several authors indicate that in subacute thyroiditis, MCs expressing growth factors contribute to thyroid tissue repair through modulation of folliculogenesis and angiogenesis [12].

MCs have been shown to participate in tumor progression across multiple anatomical sites. Among the cellular populations infiltrating the tumor stroma, MCs can influence several aspects of tumor biology, including tumor development and progression, angiogenesis, lymphangiogenesis, and tissue remodeling [12–16]. During tumor growth, targeted MC degranulation occurs, accompanied by the release of factors that stimulate angiogenesis and metastasis [17, 18]. In malignant neoplasms of the thyroid gland (papillary, follicular, highly aggressive differentiated, and poorly differentiated carcinomas), enhanced MC infiltration is observed compared with normal thyroid tissue, and increased MC density correlates with an unfavorable prognosis [19].

The histogenesis of MTC warrants special consideration. The etiology of this carcinoma differs fundamentally from that of other thyroid neoplasms. MTC is traditionally regarded as a neuroendocrine tumor [20]. According to studies by Mo et al. and Meng et al., a high MC density is an independent predictor of prolonged progression-free survival in pancreatic neuroendocrine tumors of varying malignancy grades (grades 1, 2, and 3) [21, 22].

To date, no studies have specifically addressed the role of MCs in the course of sporadic medullary thyroid carcinoma, making this issue particularly relevant considering the divergent concepts proposed by different investigators.

**This study aimed** to demonstrate the feasibility of immunohistochemical detection of mast cells in the tumor microenvironment of sporadic medullary thyroid carcinoma.

## METHODS

### Study Design

This was a cross-sectional multicenter study of paraffin-embedded histological sections of specimens from sporadic medullary thyroid carcinoma.

### Study Setting

The study was conducted at the National Medical Research Center of Endocrinology named after Academician I.I. Dedov and at Patrice Lumumba Peoples' Friendship University of Russia.

The study was conducted in the period: April 2024 - February 2025.

### Eligibility Criteria

A total of 43 patients participated in the study: 12 men and 31 women aged 40–65 years. At the preoperative stage, all patients underwent molecular genetic analysis of peripheral blood using next-generation sequencing (NGS). No germline pathogenic or likely pathogenic single nucleotide variants (SNVs) in the *RET* gene were detected.

### Intervention

All patients underwent surgical treatment consisting of total thyroidectomy (76.5% of patients) and hemithyroidectomy

(23.5%). Each thyroid tissue specimen was assigned an individual identification number, which allowed anonymization of the material for subsequent analysis.

## Study Outcome

### Main Study Outcome

Quantitative assessment of tryptase-positive mast cells in the tumor microenvironment of sporadic medullary thyroid carcinoma.

### Outcomes Registration

#### Immunohistochemical Analysis

For immunohistochemical analysis, tissue sections (3  $\mu\text{m}$ ) were deparaffinized. Antigen retrieval was then performed by heating the sections in R-UNIVERSAL epitope retrieval buffer (Aptum Biologics Ltd., United Kingdom) using a steam generator at 95 °C for 30 minutes. The step of blocking endogenous Fc receptors before incubation with primary antibodies was omitted in accordance with published guidelines [23]. After antigen retrieval and neutralization of endogenous peroxidase activity, the sections were immunostained using primary antibodies against tryptase (dilution 1:2000, mouse monoclonal antibodies, Cat. No. ab2378; Abcam, United Kingdom). Antigen visualization was performed using secondary antibodies (dilution 1:2000, goat polyclonal antibodies, Cat. No. ab6789; Abcam, United Kingdom) and the chromogen DAB (3,3'-diaminobenzidine).

#### U-Net Segmentation of Tryptase-Positive Cells

To train the convolutional neural network (CNN) designed for segmentation of tryptase-positive cells, a dataset of 67 whole slide images (WSIs) of immunohistochemical specimens was generated. Subsequently, 694 regions of interest containing positively stained cells and regions without positive staining were identified. Each image (region of interest) had a size of 512  $\times$  512 pixels at a resolution of 0.5029  $\mu\text{m}/\text{pixel}$  and  $\times 20$  magnification. Expert annotation of the regions of interest was performed using QuPath software (version 0.5.1) [24]. The images were then binarized by assigning a value of 1 to immunopositive regions and 0 to immunonegative regions. The final dataset included 694 binary masks and the corresponding original immunohistochemical images. The total dataset was stratified into three subsets: 541 images for the training set (78.0%), 73 for the test set (10.5%), and 80 for the validation set (11.5%). Model training was performed using DeepMIB software (version 2.91 beta20; University of Helsinki, Finland) [25]. Segmentation was carried out using a U-Net convolutional neural network architecture with three depth levels and 32 filters. No downsampling was applied to preserve the original image resolution. After initial training, a human-in-the-loop approach was applied, consisting of verification of the results by a qualified pathologist, which expanded the annotated dataset to 808 images (655 images in the training set). The pathologist made corrections in

114 images in which segmentation was insufficient and/or erroneous. Subsequently, the model was retrained for 500 epochs with data augmentation applied during training.

## Subgroup Analysis

The distribution of patients according to the TNM classification (Tumor–Node–Metastasis, 8th edition) is presented in Table 1. The imbalance of the sample is attributable to the specific clinical focus of the National Medical Research Center of Endocrinology, which places particular emphasis on the early diagnosis, detection, and treatment of medullary carcinoma.

## Statistical Analysis

Model performance was evaluated using the following metrics: Intersection over Union (IoU), Boundary F1 Score (BF), and Accuracy (ACC). The trained model was applied to the test dataset using FastPathology software (version 1.1.3; Norwegian University of Science and Technology, Norway). The model was further verified on a dataset comprising all whole-slide images. The resulting segmentation of immunopositive cells was quantitatively analyzed using QuPath software.

Statistical analysis was performed in the R environment (version 4.4.0; R Foundation, Austria) using standard data processing methods. To assess model agreement, a correlation analysis was conducted using the Spearman rank correlation coefficient, comparing the density of tryptase-positive cells in tumor and non-tumor regions with the qualitative assessment provided by the pathologist. Quantitative data are presented as mean and standard deviation, median and first and third quartiles (25th, 75th percentiles), and minimum and maximum values. The IoU, F1 score, and ACC metrics were calculated for each image in the test dataset. Differences were considered statistically significant at  $p = 0.05$ .

**Table 1.** Distribution of patients according to the TNM classification (8th edition)

TNM classification	Number of cases (proportion in the sample, %)
<i>T (Tumor)</i>	
T1a	8 (18.6)
T1b	23 (53.5)
T2	8 (18.6)
T3a	4 (9.3)
<i>N (Nodes)</i>	
N0	24 (55.8)
N1a	5 (11.6)
N1b	9 (20.9)
Nx	5 (11.6)
<i>M (Metastasis)</i>	
Mx	43 (100)

**Table 2.** Confusion matrix and model accuracy by class

Reference labeling	Predicted class	
	Class 0	Class 1 (tryptase-positive cells)
Class 0	99.96%	0.04%
Class 1	25.08%	74.92%

## RESULTS

### Sample Characteristics

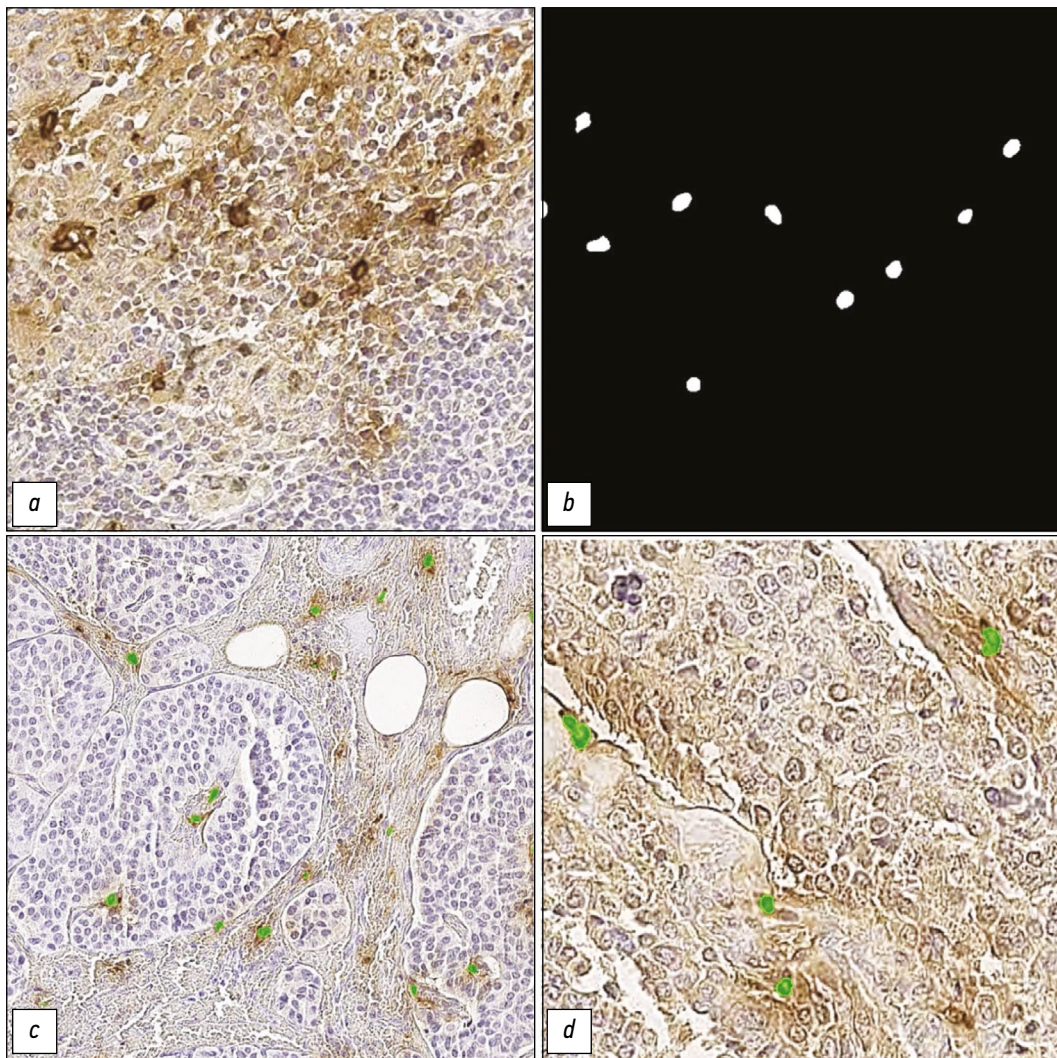
A total of 43 patients were included in the study. Their characteristics according to the TNM classification (8th edition) are presented in Table 1. Most patients were classified as T1b (53.5%), and 55.8% had no regional lymph node metastases (N0). The presence of distant metastases was not assessed at the time of the study (Mx, 100%).

### Primary Results

The trained model demonstrated the following performance metrics for class 1 (immunopositive tumor regions): accuracy

(ACC), 0.75; Intersection over Union (IoU), 0.57; and mean Boundary F1 Score (BF), 0.85 (Table 2). Despite the overall satisfactory model performance, a high proportion of false-negative segmentations indicates the need for expanding the training dataset. Spearman correlation coefficients for the quantification of tryptase-positive cells in WSI regions showed values of 0.70 for tumor regions and 0.42 for non-tumor regions, indicating a strong and moderate positive correlation, respectively, between the model output and expert assessment by a pathologist.

The study identified several potentially clinically relevant correlations. A relationship was observed between the number of mast cells per mm<sup>2</sup> in the thyroid stroma and



**Fig. 1.** Data annotation and CNN model output: *a*, original image; *b*, binarized mask of annotated regions; *c*, *d*, example of segmentation results produced by the CNN model.

patient age (Spearman coefficient  $r = 0.34$ ,  $p = 0.0310$ ), whereas no such association was found in the tumor region (Spearman coefficient  $r = 0.10$ ,  $p = 0.5029$ ). This discrepancy may be explained by multiple factors. It is well known that mast cell density varies in allergic reactions and autoimmune processes. Given that this parameter does not correlate with TNM classification and the sample size is limited, its clinical relevance in oncogenesis cannot be considered definitive. Nevertheless, these findings highlight the need for broader patient evaluation to identify additional factors associated with increased mast cell density.

A notable correlation was identified between the number of MCs in the tumor and T stage according to the TNM classification (Spearman coefficient  $r = 0.55$ ,  $p = 0.0002$ ; Table 3). When mean values were considered, a higher T stage was associated with a higher mast cell count. However, this parameter is influenced by tumor size and changes when normalized per unit area (Table 4). When mast cell density per  $\text{mm}^2$  was analyzed, differences between T1b and T2 stages were minimal, which may reflect a transitional biological state of the tumor. This emphasizes the need to better understand the role of MCs in the pathogenesis of MTC and how it evolves with tumor progression.

When the percentage ratio of MCs to tumor cells was calculated, there was a further change in the data (Table 5): the mean value of this parameter was higher in patients with T1b tumors than in those with T2 tumors. These findings

underscore a possible role of MCs in either the progression or regression of medullary carcinoma.

Beyond quantitative differences, distinct features of co-localization of MCs with other cells in the tumor microenvironment, as well as with atypical tumor cells, were observed. Even with monoplex tryptase staining, close spatial proximity between MCs and one or more immunocompetent cells was evident within the tumor stroma. Further phenotypic characterization is likely to reveal critical details of immunogenesis within the local tissue microenvironment. Moreover, in each examined patient, the interaction between MCs and atypical tumor cells exhibited individual-specific patterns, both in terms of co-localization frequency and contact surface area. Tryptase is secreted from a specific pole of the MCs in a polarized manner, followed by targeted accumulation within the extracellular matrix in the microenvironment of atypical cells. Some extracellular secretory granules showed a loss of tryptase immunopositivity, which may be attributed to the active transport of this specific protease toward its biological targets.

## DISCUSSION

### Summary of Primary Results

The conducted study is the first to demonstrate the feasibility of immunohistochemical detection of tryptase-positive mast cells in the tumor microenvironment of sporadic

**Table 3.** Number of detected mast cells in tumors according to the T stage by TNM classification

Descriptive statistics	Intratumoral mast cell count, <i>n</i>			
	T1a	T1b	T2	T3a
Mean ± standard deviation	41.5±56.9	215±208	765±918	1370±1510
Median [Q1; Q3]	22.5 [7.0; 37.3]	162 [86; 214]	380 [97; 1100]	937 [531; 1770]
Min–max range	6–174	1–681	29–2540	65–3530

**Table 4.** Number of mast cells per unit tumor area according to the T stage by TNM classification

Descriptive statistics	Intratumoral mast cell density, cells/ $\text{mm}^2$			
	T1a	T1b	T2	T3a
Mean ± standard deviation	1.96±2.02	3.57±3.75	3.74±3.88	5.87±3.50
Median [Q1; Q3]	1.46 [0.84; 2.04]	2.14 [1.15; 4.23]	2.35 [1.01; 5.28]	5.60 [3.80; 7.68]
Min–max range	0.27–6.65	0.088–13.100	0.34–10.90	2.05–10.20

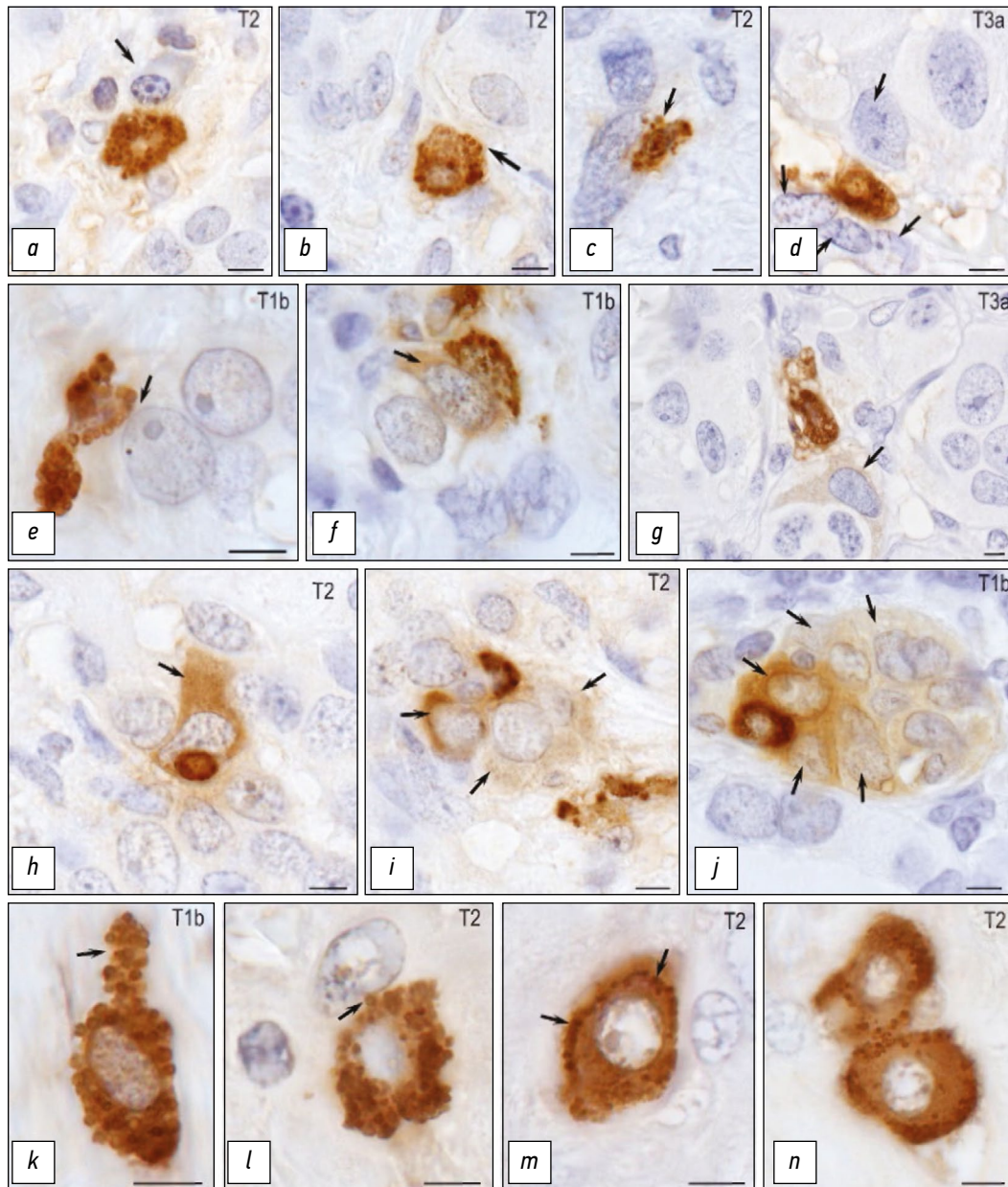
**Table 5.** Percentage ratio of mast cells to tumor cells according to the T stage by TNM classification

Descriptive statistics	Proportion of mast cells relative to tumor cells, %			
	T1a	T1b	T2	T3a
Mean ± standard deviation	0.07±0.07	0.12±0.10	0.10±0.10	0.16±0.10
Median [Q1; Q3]	0.04 [0.03; 0.11]	0.08 [0.03; 0.19]	0.07 [0.04; 0.15]	0.14 [0.10; 0.19]
Min–max range	0.01–0.20	0.00–0.34	0.00–0.24	0.05–0.29

MTC using a CNN-based model. Quantitative differences in mast cell count were identified across different disease stages according to the TNM classification (8th edition). The observed features indicate the need for further study of the histotopographic and cytological characteristics of tryptase-positive MCs, as well as the application of additional markers for more detailed immunophenotyping of surrounding cells and matrix structures.

## Interpretation

The obtained results are consistent with the findings of Mo et al. [21] and Meng et al. [22], who demonstrated that a high mast cell density is an independent predictor of long-term progression-free survival in patients with pancreatic neuroendocrine tumors. Given that MTC is a neuroendocrine tumor, the correlations we observed between MC count and disease stage may carry prognostic value.



**Fig. 2.** Histotopographic and cytological features of tryptase-positive tumor-associated mast cells in sporadic medullary thyroid carcinoma: T1b, T2, T3a, tumor stages according to the TNM classification (8th edition); *a*, colocalization of a mast cell with a plasma cell (arrow) in the tumor microenvironment; *b*, adjacency of a mast cell to an atypical cell showing signs of secretory activity (arrow); *c*, targeted secretion of autonomous tryptase-positive granules by a mast cell toward a tumor cell (arrow); *d*, colocalization of a mast cell with multiple atypical cells (arrow); *e*, mature secretory granules in a mast cell adjacent to a tumor cell (arrow); *f*, adjacency of a tryptase-positive mast cell to an atypical cell (arrow); *g-j*, various patterns of tryptase-mediated juxtacrine interaction between mast cells and tumor cells, accompanied by tryptase transport into cytoplasmic compartments (arrow); *k*, elongated mast cell with a large cytoplasmic process (arrow), filled with tryptase-positive secretory granules showing peripheral intragranular localization of the protease; *l*, round mast cell filled with mature secretory granules, exhibiting whole-granule secretion directed toward targets in the tumor microenvironment (arrow); *m*, mast cell with predominant peripheral localization of secretory granules (arrow); *n*, interaction between two mast cells. Scale bar, 5  $\mu$ m.

Of particular interest are the detected patterns of co-localization of mast cells with other components of the tumor microenvironment and the observed features of their secretory activity, which collectively suggest an active role of MCs in tumor biology.

### Study Limitations

This study has several limitations, including a relatively small sample size (43 patients) and uneven distribution across TNM stages. In addition, the retrospective design and participation of a limited number of sites may restrict the generalizability of the results.

## CONCLUSION

Our study demonstrated the presence of mast cells in the stroma of medullary thyroid carcinoma and revealed quantitative differences associated with tumor node size. The obtained results indicate the need for expanding the study cohort and improving its uniformity. Nevertheless, the data suggest a relationship between TNM stage and the number of tryptase-positive MCs in the tumor stroma. Recalculating the data as percentages, taking into account staging characteristics, suggests the existence of transitional forms of sporadic MTC, in which the tumor may alter its malignant potential under the influence of the immune microenvironment.

This work may serve as a starting point for in-depth investigation of the role of MCs in the development of sporadic MTC, the understanding of which may contribute to the identification of new therapeutic targets. In addition, the active interaction of MCs with atypical sporadic MTC cells and other components of the tumor microenvironment, as identified in this study, may be considered a potential criterion for interpreting the biological effects of MCs on the tumor and warrants further analysis for the development of diagnostic algorithms and improvement of prognostic objectivity.

## ADDITIONAL INFORMATION

**Author contributions:** E.V. Bondarenko: conceptualization, writing—original draft, writing—review & editing; M.V. Balyasin: formal analysis, writing—original draft, writing—review & editing; A.A. Kostin: formal analysis, writing—review & editing; A. Chevais: investigation, writing—original draft, writing—review & editing; A.V. Alekhnovich: formal analysis, writing—review & editing; F.M. Abdulkhabirova: investigation, writing—original draft, writing—review & editing; D.A. Atiakshin: conceptualization, investigation, writing—original draft, writing—review & editing. All the authors approved the version of the manuscript to be published and agreed to be accountable for all aspects of the work, ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

**Ethics approval:** The study was reviewed and approved by the Local Ethics Committee of the Endocrinology Research Center (Minutes No. 1 of January

15, 2025). Before enrollment, all participants provided written informed consent approved by the Ethics Committee as part of Minutes No. 1 of January 15, 2025.

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**Statement of originality:** This study is an original research work. All data presented in the manuscript were obtained for the first time during this study. No previously published or obtained materials were used.

**Data availability statement:** The authors provide limited access to the data upon reasonable request. The restriction is due to the need to protect patients' personal medical data in accordance with the requirements of the Local Ethics Committee and the legislation of the Russian Federation on personal data. Requests must include the purpose of data use, the intended analytical methods, and information on the researcher's institutional affiliation. Please send your requests to the corresponding author, Ekaterina V. Bondarenko (ekaterinabondarenko@inbox.ru). Upon request, access may be granted to the trained CNN model weights in .onnx format and to a depersonalized dataset containing binary masks of annotated regions (694 images, 512×512 pixels).

**Generative AI:** No generative artificial intelligence technologies were used to prepare this article.

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## ДОПОЛНИТЕЛЬНАЯ ИНФОРМАЦИЯ

**Вклад авторов.** Е.В. Бондаренко — определение концепции, написание черновика рукописи, пересмотр и редактирование рукописи; М.В. Баясин — анализ данных, написание черновика рукописи, пересмотр и редактирование рукописи; А.А. Костин — анализ данных, пересмотр и редактирование рукописи; А. Шевэ — проведение исследования, написание черновика рукописи, пересмотр и редактирование рукописи; А.В. Алехнович — анализ данных, пересмотр и редактирование рукописи; Ф.М. Абдулхабилова — проведение исследования, написание черновика рукописи, пересмотр и редактирование рукописи; Д.А. Атякшин — определение концепции, проведение исследования, написание черновика рукописи, пересмотр и редактирование рукописи. Все авторы одобрили рукопись (версию для публикации), а также согласились нести ответственность за все аспекты настоящей работы, гарантируя надлежащее рассмотрение и решение вопросов, связанных с точностью и добросовестностью любой её части.

**Этическая экспертиза.** Исследование рассмотрено и одобрено локальным этическим комитетом ФГБУ «НМИЦ эндокринологии им. академика И.И. Дедова» Минздрава России, протокол № 1 от 15 января 2025 года. До включения в исследование все участники добровольно подписали форму информированного согласия, утверждённую этическим комитетом в составе протокола № 1 от 15 января 2025 года.

**Источники финансирования.** Научное исследование проведено при поддержке Государственного задания «Гормонально-метаболические и молекулярно-клеточные характеристики заболеваний щитовидной железы, как основа для разработки инновационных методов диагностики, лечения и профилактики» Рег. № НИОКТР 123021300097-0 ФГБУ «НМИЦ эндокринологии им. академика И.И. Дедова» Минздрава России.

**Раскрытие интересов.** Авторы заявляют об отсутствии отношений, деятельности и интересов за последние три года, связанных с третьими лицами (коммерческими и некоммерческими организациями), интересы которых могут быть затронуты содержанием статьи.

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**Доступ к данным.** Авторы предоставляют ограниченный доступ к данным по обоснованному запросу. Ограничение обусловлено

необходимостью защиты персональных медицинских данных пациентов в соответствии с требованиями локального этического комитета и законодательства Российской Федерации о персональных данных. Запросы должны содержать описание цели использования данных и планируемых методов анализа, а также информацию об институциональной принадлежности исследователя. Запросы следует направлять автору, ответственному за переписку: Бондаренко Екатерине Владимировне (ekaterinabondarenko@inbox.ru). По запросу предоставляется доступ к весам обученной CNN-модели в формате .onnx и обезличенному датасету с бинарными масками аннотированных областей (694 изображения размером 512×512 пикселей).

**Генеративный искусственный интеллект.** При создании настоящей статьи технологии генеративного искусственного интеллекта не использовались.

**Рассмотрение и рецензирование.** Настоящая работа подана в журнал в инициативном порядке и рассмотрена по обычной процедуре. В рецензировании участвовали два внешних рецензента, член редакционной коллегии и научный редактор издания.

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