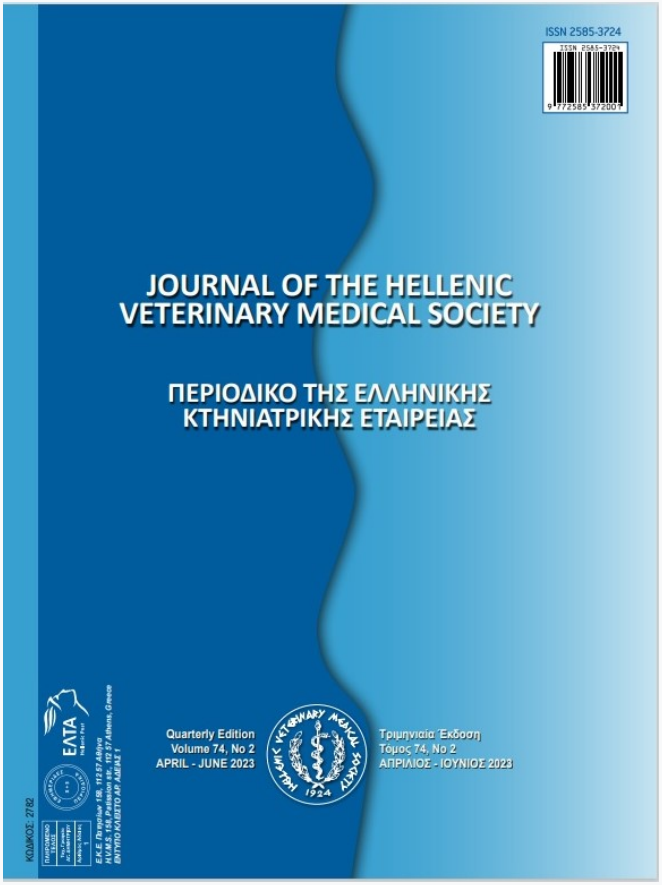
Journal of the Hellenic Veterinary Medical Society

Vol 74, No 2 (2023)



Mast Cell and VEGF Profile in Canine Soft Tissue Tumors

V Ipek, O Kaplan

doi: 10.12681/jhvms.30129

To cite this article:

Ipek, V., & Kaplan, O. (2023). Mast Cell and VEGF Profile in Canine Soft Tissue Tumors. *Journal of the Hellenic Veterinary Medical Society*, *74*(2), 5713–5720. https://doi.org/10.12681/jhvms.30129 (Original work published July 5, 2023)

J HELLENIC VET MED SOC 2023, 74 (2): 5713-5720
ΠΕΚΕ 2023, 74 (2): 5713-5720

Research article
Ερευνητικό άρθρο

Mast Cell and VEGF Profile in Canine Soft Tissue Tumors

V. Ipek[1*], O. Kaplan[2]

[1]*Mehmet Akif Ersoy University, Faculty of Veterinary Medicine, Department of Pathology*

[2]*Mehmet Akif Ersoy University, Institute of Health Sciences*

ABSTRACT: Canine soft tissue sarcomas are a complex group with a higher tendency for invasion and metastasis, making them challenging to treat. Although there are many different subtypes of these tumors, they are grouped under a single group called "soft tissue tumors", along with benign ones, because of their common mesenchymal features and morphological similarities. In addition to promoting tumor angiogenesis, proliferation, invasion, and metastasis the tumor microenvironment, which includes mast cells, also mediates the mechanism of therapeutic resistance. In this study, the presence and relationships of mast cells with VEGF in benign and malignant soft tissue tumors were investigated. As a result, mast cells were found in significantly higher numbers in malignant tumors than benign tumors. The immunostaining scores in malignant tumors were also higher in the VEGF analyses performed for the interpretation of angiogenesis. To the best of the authors' knowledge, angiogenesis and mast cells in canine soft tissue tumors are poorly characterized. This study revealed that mast cell and VEGF scores are greater in malignant tumors than benign tumors and can be used as a prognostic marker. However, no link between mast cells and VEGF immune expressions was found.

***Keywords*:** Dog; soft tissue tumor; mast cell; VEGF

Corresponding Author:
Volkan IPEK, Burdur Mehmet Akif Ersoy University, Faculty of Veterinary Medicine, Department of Pathology, Burdur, Turkey,
E-mail address: volkanipek@mehmetakif.edu.tr

Date of initial submission: 02-04-2022
Date of acceptance: 22-10-2022

INTRODUCTION

Canine soft tissue tumors are a group of tumors that originate from non-osseous connective tissues and include a variety of tumor types (Mayer and LaRue, 2005). Malignant tumors in this group include fibrosarcoma, hemangiopericytoma, myxosarcoma, undifferentiated sarcoma, liposarcoma, malignant fibrous histiocytoma, schwannoma, rhabdomyosarcoma, and neurofibrosarcoma (Thrall and EL, 1995). These tumors can form in any part of the body, but they are commonly found in the cutaneous and subcutaneous tissues (Dennis et al., 2011). Despite having different biological origins, these tumors are thought to have similar biological behaviors (Bostock and Dye, 1980, Kuntz et al., 1997, Chase et al., 2009). Accurate prognostic information is critical for the right approach to patients, therefore studies are conducted to determine various prognostic factors (Dennis et al., 2011).

The growth of new blood vessels (angiogenesis) is necessary for the majority of solid tumor metastases, and imbalances in the blood vessel development process lead to a variety of tumoral, inflammatory, ischemic, infectious, and immunological illnesses (Folkman et al., 1989, Rak et al., 1995). According to current research, mast cells play an important role in tumor progression by stimulating angiogenesis, particularly with the tryptase they contain (Ribatti et al., 2004). Data show that mast cell density is associated with high micro vessel density (Grimbaldeston et al., 2007) and increased inflammation (Sener and Ipek, 2021). There is also evidence that mast cells can both inhibit and stimulate tumor growth in some cases (Theoharides and Conti, 2004). Mast cells in sarcomas play a stimulating role, most likely due to their ability to secrete vascular endothelial growth factor (VEGF) (Baneth et al., 2005). Because of these controversial findings regarding the role of mast cells in tumor stroma, more clarification on the role of mast cells in tumors is required. The aim of this study was to investigate the presence of mast cells in canine benign and malignant soft tissue tumors as well as their relationship with VEGF staining intensities.

MATERIALS AND METHODS

Retrospective Study

In this retrospective study, 28 paraffin blocks with a diagnosis of canine soft tissue tumor from the tissue archive of the pathology department were used. Sections (4-5 μm) were taken from these blocks on routine and poly-L-lysine slides for histopathological and immunohistochemical analysis, respectively. Firstly, soft tissue tumors were classified histopathologically (Hendrick et al., 1998, Gross et al., 2005, Avallone et al., 2007). Accordingly, tumors were classified as fibrous tissue tumors, adipose tissue tumors, muscle tumors, perivascular wall tumors, and peripheral nerve sheath tumors. Unfortunately, immunohistochemical analysis for the differential diagnosis could not be performed. Tumors were classified as benign or malignant based on their histopathological features. VEGF (clone JH121, Thermo) antibody for angiogenesis was used immunohistochemically. Immunostaining was also done with tryptase (clone AA1, Biolegend) to reveal mast cells.

Histopathology and Immunohistochemistry

Hematoxylin and eosin staining was used for histopathological analysis. For immunohistochemical staining, after passing through the xylol and alcohol series, the slides were dipped in a 10-fold diluted citrate buffer solution (Citrate Buffer Heat-Induced Epitope Retrieval pH: 6 Thermo Scientific) for antigen retrieval and kept in the autoclave at 120 ºC for 15 minutes. Tissues were then incubated in a blocking solution (Bloxall, Vector Lab) for 15 minutes to block endogenous peroxidase activity. After phosphate buffered solution (PBS) washing for 5 minutes, 2.5% normal goat serum (Vector Lab) solution was applied for 30 minutes to prevent nonspecific antibody binding. After removing the protein block solution, slides with VEGF antibody (1/20 dilution) and tryptase antibody (1/25 dilution) were incubated at +4 ºC overnight. Then sections were washed with PBS for 2x5 minutes and were treated with secondary antibody (Amplifier antibody [Goat anti mouse], Vector Lab) and then streptavidin peroxidase solution (Vectorlab, ImmPRESS Excel Amplified HRP Polymer Reagent). Under the microscope, DAB chromogen (Vectorlab, ImmPACT® DAB EqV Substrate) was applied in a controlled manner. Sections were counterstained with Mayer's hematoxylin (Merck) for 25 seconds and covered with entellan. For the positive control tissue, canine mast cell tumor for tryptase and normal canine skin for VEGF were used. For the negative control, after protein blocking, PBS was used instead of the primary antibody.

Semi-quantitative Method

Evaluation of immunohistochemical staining was done semi-quantitatively. Tryptase-positive mast cells were counted in five high magnification fields. Mast

cell numbers ranged from 0 to 49. Then scoring was made between 0-4 as follows; 0; no mast cell, 1; ≥ 1-12 mast cells, 2; ≥ 13-24 mast cells, 3; ≥ 25-36 mast cells, and 4; > 36 mast cells. VEGF staining was scored according to the staining intensity of the cells (1; no staining, 2; weak staining, 3; moderate staining, and 4; strong staining) and then the ratio of the stained areas in the tumors (1; < 20%, 2; ≥ 20-50%, 3; > 50-80%, and 4; > 80%). The final scores were calculated from the numbers obtained by the multiplying of these two scores (1; ≤ 4, 2; ≥ 5-8, 3; ≥ 9-12, and 4; > 12). This scoring system was modified according to the study of Kamarlis et al. (2017).

Statistical Analysis

For statistical analysis, the Minitab 16 package program (version 16.1.1) was used. The Ryan-Joiner normality test was used to determine whether the obtained scores showed a normal distribution. After scoring, differences in immunostaining between benign and malignant tumors were statistically evaluated using two-sample T-tests to determine whether the results were significant. In addition, the Pearson correlation test was used to determine the correlation between mast cells and VEGF in all tumors. The correlation coefficient r was evaluated as follows: 0–0.3 insignificant correlation, 0.3–0.5 low correlation, 0.5–0.7 moderate correlation, 0.7–0.9 high correlation, and 0.9–1 very high correlation (Hinkle et al., 2003, Mulaka, 2012).

RESULTS

Histopathologically, tumors were diagnosed as fibroma (n:7), leiomyoma (n:2), fibrosarcoma (n:5), myxosarcoma (n:1), fibromyxosarcoma (n:1), undifferentiated sarcoma (n:4), malignant peripheral nerve sheath tumor (PNST) (n:2), malignant perivascular wall tumor (PWT) (n:2), and liposarcoma (n:4) (Figure-1).

Positive immunostaining of mast cells using anti-tryptase antibody were scored between 0-4. Accordingly, in fibroma cases, the score was 1 in six cases and 2 in one case. In leiomyoma cases, score 0 in one case, and 1 in one case. In fibrosarcoma cases, the score was 1 in two cases, 2 in one case, 3 in one case, and 4 in one case. The score is 0 and 1 in myxosarcoma case and fibromyxosarcoma case, respectively. In the undifferentiated sarcoma cases, the score was 1 in one case, 2 in two cases, and 3 in one case. In the malignant PNST cases, the score was 1 in one case and 2 in the other case. In the malignant PWT cases, the score was 1. Finally, in the liposarcoma cases, the score was 1 in three cases, and 2 in one case (Figure-2).

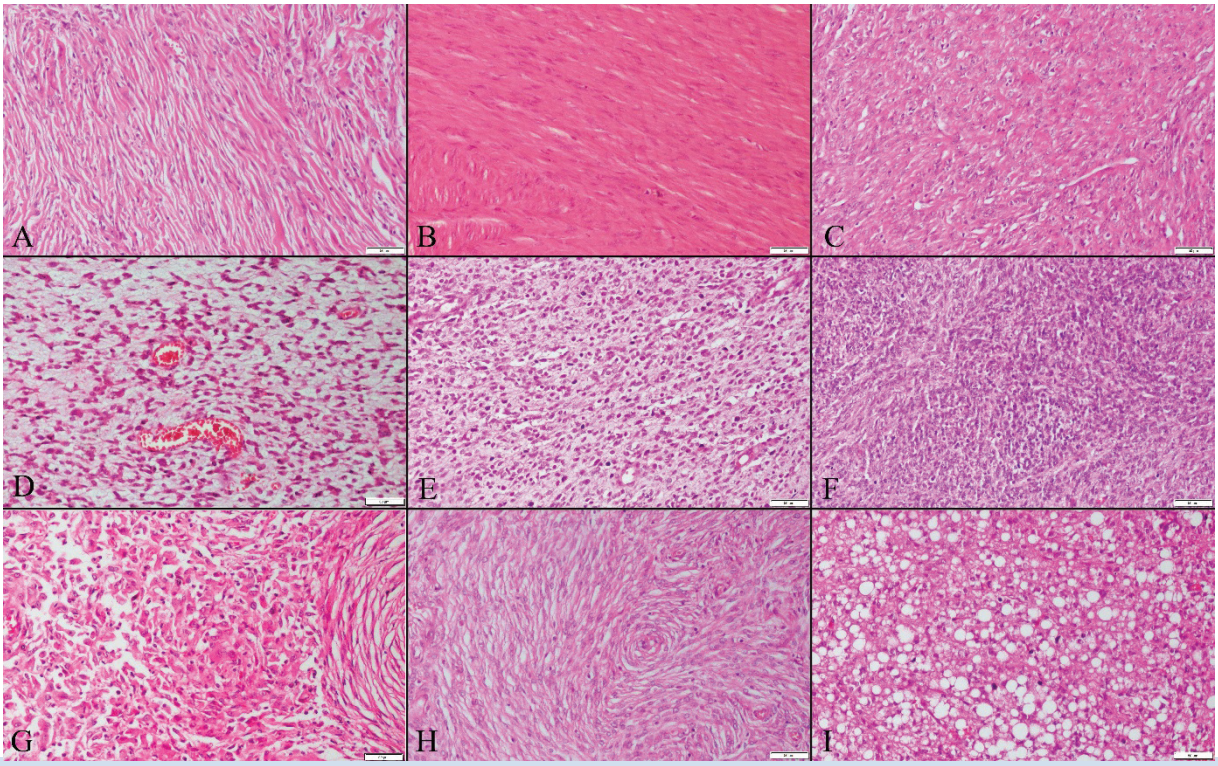


Figure-1: Histopathological appearances of the case of fibroma (A), leiomyoma (B), fibrosarcoma (C), myxosarcoma (D), fibromyxosarcoma (E), undifferentiated sarcoma (F), malignant PNST (G), malignant PWT (H), and liposarcoma (I)). H&E Bars: 50 µm

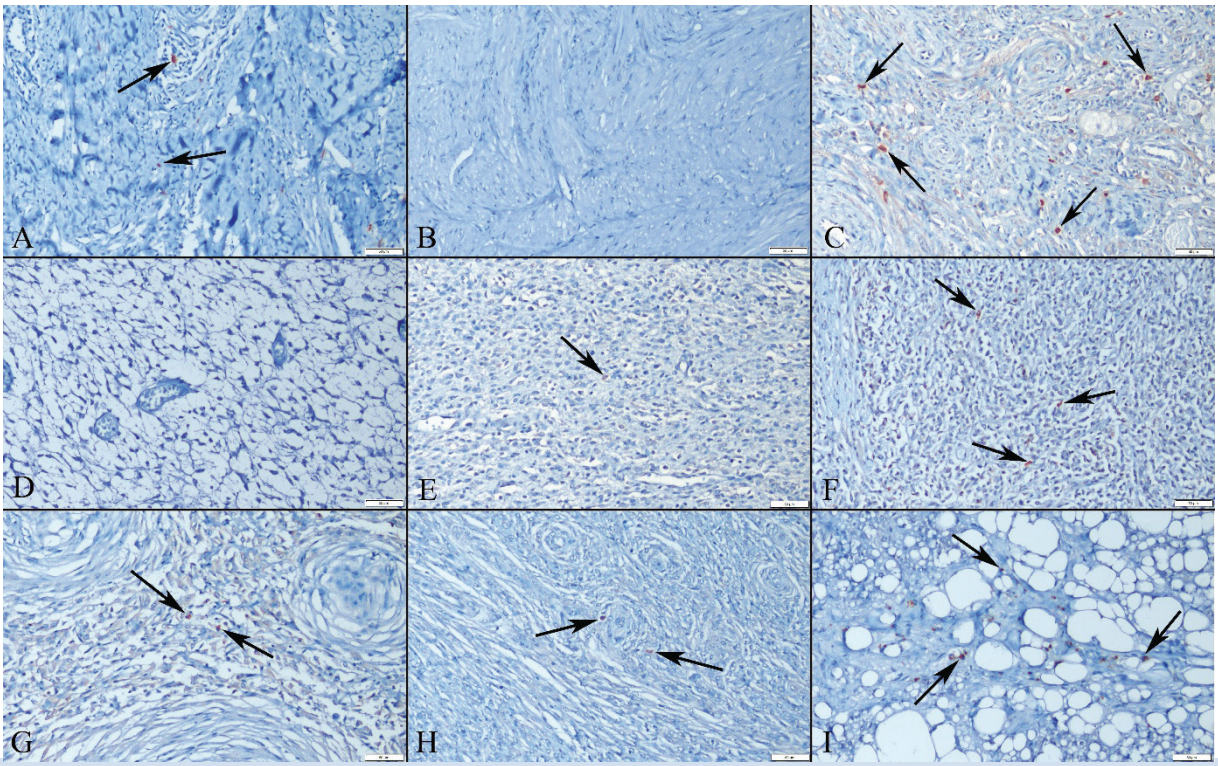


Figure-2: Anti-tryptase positive mast cells in fibroma (A), leiomyoma (B), fibrosarcoma (C), myxosarcoma (D), fibromyxosarcoma (E), undifferentiated sarcoma (F), malignant PNST (G), malignant PWT (H), and liposarcoma (I)). DAB, Bars: 50 µm.

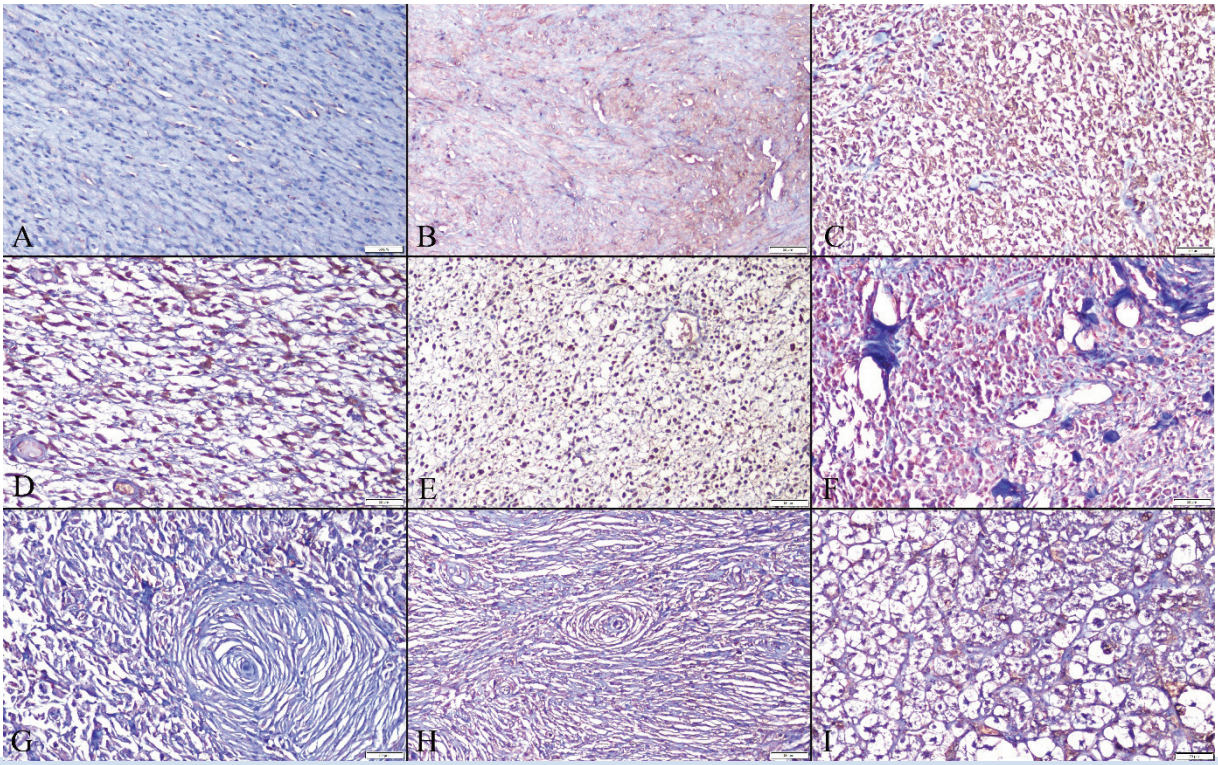


Figure-3: Anti-VEGF positivity in fibroma (A), leiomyoma (B), fibrosarcoma (C), myxosarcoma (D), fibromyxosarcoma (E), undifferentiated sarcoma (F), malignant PNST (G), malignant PWT (H), and liposarcoma (I). DAB, Bars: 50 µm

VEGF scoring was performed according to the staining intensity of tumoral cells and the stained cell area and total scores were obtained. Accordingly, the score was 1 in five, 2 in two of the fibroma cases, and 1 in all leiomyoma cases. In the fibrosarcoma, the scores were 1 in one case, 2 in two cases, and 3 in

two cases. The score was 3 in the myxosarcoma and fibromyxosarcoma cases. In the undifferentiated sarcoma, the score was 1 in one case, 2 in two cases, and 3 in the other case. The score was 1 in the malignant PNST cases. In the malignant PWT cases, the score was 2 in one case and 4 in the other case. Finally, in the liposarcoma cases, the score was 1 in three cases and 3 in another case (Figure-3). Detailed information about histopathological diagnoses and immunohistochemical scores is summarized in Table 1.

Statistically, mast cell counts in malignant tumors were found to be significantly higher than in benign tumors in a two-sample T-test ($P < 0.05$). The VEGF score was also significantly higher in malignant tumors ($P < 0.01$). Pearson correlation analysis revealed an insignificant correlation was found between mast cell and VEGF scores ($r= 0.201$). When the correlation between mast cell scores and VEGF scores was examined only in benign tumors, a moderate correlation was observed ($r= 0.567$), but no correlation was found between mast cell scores and VEGF scores in malignant tumors ($r= 0.025$).

DISCUSSION

Inflammation in a tumor microenvironment is a prominent feature of cancer (Caso et al., 2009, Tu et al., 2016). A tumor microenvironment is formed by immune cells, leukocytes, fibroblasts, and vascular endothelial cells that interact with tumor cells to influence tumor formation, growth, and metastasis (Teng et al., 2016). Mast cells are a major source of

Table-1. Information of the cases, histopathological diagnoses and immunohistochemical scores

No	Species	Race	Gender	Age (year)	Localization	Histopathological Diagnosis	Mast Score	VEGF
1.	Dog	Crossbreed	Female	14	Breast	Fibroma	1	1
2.	Dog	Beagle	Male	6	Hip	Fibroma	1	2
3.	Dog	Beagle	Male	6	Hip	Fibroma	2	2
4.	Dog	Russian poodle	Male	10	Gum	Fibroma	1	1
5.	Dog	Crossbreed	Female	12	Vagina	Fibroma	1	1
6.	Dog	Golden retriever	Female	6	Breast skin	Fibroma	1	1
7.	Dog	Brazilian Mastiff	Male	10	Hip	Fibroma	1	1
8.	Dog	Crossbreed	Female	11	Vagina	Leiomyoma	1	1
9.	Dog	Terrier	Female	16	Vagina	Leiomyoma	0	1
10.	Dog	Golden retriever	Male	5.5	Thyroid, neck region	Fibrosarcoma	4	2
11.	Dog	Rottweiler	Male	4	Right forearm	Fibrosarcoma	1	2
12.	Dog	Setter	Male	3	Left front leg	Fibrosarcoma	1	1
13.	Dog	Crossbreed	Female	16	Gum	Fibrosarcoma	3	3
14.	Dog	Golden retriever	Male	10	Gum	Fibrosarcoma	2	3
15.	Dog	Golden retriever	Female	9	Left scapular region	Myxosarcoma	0	3
16.	Dog	Golden retriever	Male	10	Maxilla	Fibromyxosarcoma	1	3
17.	Dog	Crossbreed	Male	6	Lower eyelid	Undifferentiated Sarcoma	2	1
18.	Dog	Rottweiler	Male	8 month	Lumbar region	Undifferentiated Sarcoma	3	3
19.	Dog	German shepherd dog	Male	6	Gum	Undifferentiated Sarcoma	2	2
20.	Dog	Crossbreed	Male	13	Spleen	Undifferentiated Sarcoma	1	2
21.	Dog	Terrier	Male	12	Perianal region	Malign PNST	1	1
22.	Dog	Husky	Female	15	Tarsal region	Malign PNST	2	1
23.	Dog	Crossbreed	Male	9	Perianal region	Malign PWT	1	2
24.	Dog	Crossbreed	Female	13	Left hind limb	Malign PWT	1	4
25.	Dog	Terrier	Female	10	Liver	Liposarcoma	2	1
26.	Dog	Crossbreed	Female	7	Abdominal muscle	Liposarcoma	1	1
27.	Dog	Crossbreed	Female	12	Femoral lateral muscle	Liposarcoma	1	3
28.	Dog	Husky	Female	7	Coccyx	Liposarcoma	1	1

histamine, which influences tumor development via the H1 and H2 receptors (Fitzsimons et al., 1997). Mast cells exert immunosuppression and promote tumor development by producing tumor necrosis factor alpha (TNF-α) and IL-10, both of which are critical in fostering immune tolerance mediated by regulatory T (Treg) cells (Grimbaldeston et al., 2007, Ullrich et al., 2007). For this purpose, we evaluated the presence and grade of mast cells in benign and malignant canine soft tissue tumors. We recently discovered a substantial rise in calprotectin positive leukocytes in canine soft tissue sarcomas which might be a prognostic marker (Savas and Ipek, 2021). Mast cells were also shown to be considerably higher in canine soft tissue sarcomas compared to their benign counterparts in this study.

Because of their histological characteristics, differential diagnosis of soft tissue tumors can be difficult (Hendrick et al., 1998, Goldschmidt and Hendrick, 2002). In our study, four mesenchymal tumors were diagnosed as undifferentiated since a definitive diagnosis could not be achieved with histopathology. Furthermore, two PWT and two PNST were diagnosed. PWTs are neoplasms originating from diverse cellular components of the vascular wall, with the exception of endothelium (Weiss and Goldblum, 2001). PNST develops from many types of peripheral nerve cells (Koestner and Higgins, 2002). PWTs are among the tumors to consider while making a diagnosis of peripheral nerve sheath tumors. PWTs have perivascular helical structures whereas PNSTs have similar structures surrounding collagen fibrils rather than capillaries (Gaitero et al., 2008). In our study, the PWT distinction was made with the absence of vessels in the center of the spiral structures observed in a PNST case. In the other PNST, differentiation was made by the presence of the Antoni-A pattern. Immunohistochemical characteristics can also be used in the differential diagnosis of soft tissue tumors (Avallone et al., 2007), however , immunohistochemical staining was not possible in our study, therefore only the histological features were evaluated.

Mast cells secrete several pro-angiogenic factors, including fibroblast growth factor-2, VEGF, IL-8, TNF-α, TGF-β and nerve growth factor (Möller et al., 1998, Abdel-Majid and Marshall, 2004). Mast cells migrate in response to VEGF and placental growth factor-1 both in vivo and in vitro (Gruber et al., 1988, Detmar et al., 1998). VEGF can operate as both an angiogenic and chemotactic factor for mast cells in this environment, generating an autocrine cycle of mast cell proliferation. Mast cells contain preformed active serine proteases, such as tryptase and chymase, in secretory granules (Metcalfe et al., 1997). In vitro, tryptase stimulates the endothelial cell proliferation and promotes vascular channel development . Mast cell-deficient W/Wv mice exhibit a reduced incidence of tumor angiogenesis (Starkey et al., 1988). Mast cells have been demonstrated to increase in number during angiogenesis in vascular tumors. Furthermore, enhanced neovascularization is associated with mast cell accumulation in hematological and solid tumors (Tth et al., 2000, Ribatti et al., 2003). In our study, despite the increased mast cell density and VEGF staining characteristics in malignant tumors, no correlation was found between mast cells and VEGF. This lends credence to the notion that VEGF release from tumor cells is not only dependent on mast cells but also on a variety of other tissue factors.

In previous human research, greater numbers of mast cells were seen in nerve sheath tumors (Olsson, 1968, Giorno and Claman, 1988). Mast cells are more common in neurofibromas than in neurilemomas and malignant schwannomas (Isaacson, 1976). According to Johnson et al (1989) this distinction is quite important because neurofibromas can only be distinguished from neurilemmomas and malignant schwannomas only by the amount of mast cells. Vasconcelos et al (2019) has demonstrated the prognostic importance of mast cells in human malignant PNSTs. On the other hand, Enzinger and Weiss (1988) found that mast cells relatively unimportant in the differential diagnosis of soft tissue tumors and they emphasized that they occur more frequently in synovial sarcomas. Malignant fibrous histiocytomas also have high mast cell numbers (Parwaresch et al., 1985). Moreover, mast cells have been identified as an important prognostic marker (Ueda et al., 1988). Mast cells were found to be significantly higher in sarcomas than in benign tumors in our study, suggesting that they might be a prognostic marker, albeit there was no correlation between mast cell and VEGF scores. However, further studies are needed to compare mast cells with larger numbers of samples and more prognostic data.

ACKNOWLEDGEMENT

This study was supported by Burdur Mehmet Akif Ersoy University Scientific Research Projects Commission. (Project No: 0679-MP-20)

REFERENCES

Abdel-Majid RM, Marshall JS (2004) Prostaglandin E2 Induces Degranulation-Independent Production of Vascular Endothelial Growth Factor by Human Mast Cells. J. Immunol. 172: 1227–1236.

Avallone G, Helmbold P, Caniatti M, Stefanello D, Nayak RC, Roccabianca P (2007) The spectrum of canine cutaneous perivascular wall tumors: morphologic, phenotypic and clinical characterization. Vet. Pathol. 44: 607–620.

Baneth V, Raica M, Cîmpean AM (2005) Assessment of angiogenesis in soft-tissue tumors. Rom. J. Morphol. Embryol. 46: 323–327.

Bostock DE, Dye MT (1980) Prognosis after surgical excision of canine fibrous connective tissue sarcomas. Vet. Pathol. 17: 581–588.

Caso G, Barry C, Patejunas G (2009) Dysregulation of CXCL9 and reduced tumor growth in Egr-1 deficient Mice. J. Hematol. Oncol. 2: 7.

Chase D, Bray J, Ide A, Polton G (2009) Outcome following removal of canine spindle cell tumours in first opinion practice: 104 cases. J. Small Anim. Pract. 50: 568–574.

Dennis MM, McSporran KD, Bacon NJ, Schulman FY, Foster RA, Powers BE (2011) Prognostic Factors for Cutaneous and Subcutaneous Soft Tissue Sarcomas in Dogs. Vet. Pathol. 48: 73–84.

Detmar M, Brown LF, Schön MP, Elicker BM, Velasco P, Richard L, Fukumura D, Monsky W, Claffey KP, Jain RK (1998) Increased microvascular density and enhanced leukocyte rolling and adhesion in the skin of VEGF transgenic mice. J. Invest. Dermatol. 111: 1–6.

Enzinger F, Weiss S (1988) Malignant fibrohistiocytic tumors. In: S Weiss and F Enzinger, editor. Soft Tissue Tumors St. Louis, Missouri: Mosby. p. 269–300.

Fitzsimons C, Molinari B, Duran H, Palmieri M, Davio C, Cricco G, Bergoc R, Rivera E (1997) Atypical association of H1 and H2 histamine receptors with signal transduction pathways during multistage mouse skin carcinogenesis. Inflamm. Res. 1997 468 46: 292–298.

Folkman J, Watson K, Ingber D, Hanahan D (1989) Induction of angiogenesis during the transition from hyperplasia to neoplasia. Nature 339: 58–61.

Gaitero L, Añor S, Fondevila D, Pumarola M (2008) Canine Cutaneous Spindle Cell Tumours with Features of Peripheral Nerve Sheath Tumours: A Histopathological and Immunohistochemical Study. J. Comp. Pathol. 139: 16–23.

Giorno R, Claman HN (1988) Mast cells and neurofibromatosis. Neurofibromatosis 1: 100–104.

Goldschmidt M, Hendrick M (2002) Tumors of the skin and soft tissues. In: D Meuten, editor. Tumors in domestic animals Ames, Iowa: Iowa State Univ Pr. p. 109–111.

Grimbaldeston MA, Nakae S, Kalesnikoff J, Tsai M, Galli SJ (2007) Mast cell–derived interleukin 10 limits skin pathology in contact dermatitis and chronic irradiation with ultraviolet B. Nat. Immunol. 2007 810 8: 1095–1104.

Gross T, Ihrke P, Walder E, Affolter V (2005) Skin diseases of the dog and cat: clinical and histopathologic diagnosis. Oxford: Blackwell.

Gruber B, Marchese M, Kiely J, Schwartz L, Schecter N (1988) 520 Human mast cell products degrade connective tissue matrix. J Allergy Clin Immunol 81: 298.

Hendrick M, Mahaffeey E, Moore F, Vos J, Walder E (1998) Histological classification of mesenchymal tumors of skin and soft tissues of domestic animals, Vol. 2. Washington DC: Armed Forces Institute of Pathology.

Hinkle D, Wiersma W, Jurs S (2003) Applied Statistics for the Behavioral Sciences. Boston: Houghton Mifflin.

Isaacson P (1976) Mast cells in benign nerve sheath tumours. J. Pathol. 119: 193–196.

Johnson MD, Kamso-Pratt J, Federspiel CF, Whetsell WO (1989) Mast cell and lymphoreticular infiltrates in neurofibromas. Comparison with nerve sheath tumors. Arch. Pathol. Lab. Med. 113: 1263–1270.

Kamarlis RK, Lubis MN, Hernowo BS, Harapan H, Kar AS (2017) Immunoexpression of P63 and SOX2 in triple-negative breast cancers, Indonesia. F1000Research 6: 1780.

Koestner A, Higgins R (2002) Tumors of the nervous system. In: D Meuten, editor. Tumors in domestic animals Ames: Iowa State Press. p. 697–738.

Kuntz CA, Dernell WS, Powers BE, Devitt C, Straw RC, Withrow SJ (1997) Prognostic factors for surgical treatment of soft-tissue sarcomas in dogs: 75 cases (1986-1996). J. Am. Vet. Med. Assoc. 211: 1147–1151.

Mayer MN, LaRue SM (2005) Soft tissue sarcomas in dogs. Can. Vet. J. 46: 1048.

Metcalfe DD, Baram D, Mekori YA (1997) Mast cells. Physiol. Rev. 77: 1033–1079.

Möller A, Henz BM, Grützkau A, Lippert U, Aragane Y, Schwarz T, Krüger-Krasagakes S (1998) Comparative cytokine gene expression: regulation and release by human mast cells. Immunology 93: 289–295.

Mulaka M (2012) A guide to appropriate use of correlation coefficient in medical research. Malawi Med. J. 24: 69–71.

Olsson Y (1968) Mast Cells in the Nervous System. Int. Rev. Cytol. 24: 27–70.

Parwaresch MR, Homy HP, Lennert K (1985) Tissue mast cells in health and disease. Pathol. - Res. Pract. 179: 439–461.

Rak JW, St. Croix BD, Kerbel RS (1995) Consequences of angiogenesis for tumor progression, metastasis and cancer therapy. Anticancer. Drugs 6: 3–18.

Ribatti D, Crivellato E, Roccaro AM, Ria R, Vacca A (2004) Mast cell contribution to angiogenesis related to tumour progression. Clin. Exp. Allergy 34: 1660–1664.

Ribatti D, Ennas MG, Vacca A, Ferreli F, Nico B, Orru S, Sirigu P (2003) Tumor vascularity and tryptase-positive mast cells correlate with a poor prognosis in melanoma. Eur. J. Clin. Invest. 33: 420–425.

Savas O, Ipek V (2021) Investigation of Calprotectin Positive Leukocytes in Canine Soft Tissue Tumors. J. Res. Vet. Med. 40: 77–87.

Sener S, Ipek V (2021) Investigation of brain mast cells in ovine encephalitic listeriosis. Biotech. Histochem. .

Starkey JR, Crowle PK, Taubenberger S (1988) Mast-cell-deficient W/Wv mice exhibit a decreased rate of tumor angiogenesis. Int. J. cancer 42: 48–52.

Teng F, Tian WY, Wang YM, Zhang YF, Guo F, Zhao J, Gao C, Xue FX (2016) Cancer-associated fibroblasts promote the progression of endometrial cancer via the SDF-1/CXCR4 axis. J. Hematol. Oncol. 9: .

Theoharides TC, Conti P (2004) Mast cells: the Jekyll and Hyde of tumor growth. Trends Immunol. 25: 235–241.

Thrall D, EL G (1995) Soft-tissue sarcomas. Semin Vet Med Surg (Small Anim) 10: 173–179.

Tth T, Tth-Jakatics R, Jimi S, Takebayashi S, Kawamoto N (2000) Cutaneous malignant melanoma: Correlation between neovascularization and peritumor accumulation of mast cells overexpressing vascular endothelial growth factor. Hum. Pathol. 31: 955–960.

Tu Z, Xiao R, Xiong J, Tembo KM, Deng X, Xiong M, Liu P, Wang M, Zhang Q (2016) CCR9 in cancer: Oncogenic role and therapeutic targeting. J. Hematol. Oncol. 9: .

Ueda T, Aozasa K, Tsujimoto M, Yoshikawa H, Kato T, Ono Ke, Matsumoto Ke (1988) Prognostic Significance of Mast Cells in Soft Tissue Sarcoma. Cancer 62: 2419.

Ullrich SE, Nghiem DX, Khaskina P (2007) Suppression of an Established Immune Response by UVA: A Critical Role for Mast Cells. Photochem. Photobiol. 83: 1095.

Vasconcelos RAT de, Guimarães Coscarelli P, Vieira TM, Noguera WS, Rapozo DCM, Acioly MA (2019) Prognostic significance of mast cell and microvascular densities in malignant peripheral nerve sheath tumor with and without neurofibromatosis type 1. Cancer Med. 8: 972–981.

Weiss S, Goldblum J (2001) Perivascular tumors. In: S Weiss and J Goldblum, editor. Soft tissue tumors St. Louis MO: Mosby. p. 985–1035.