Shiraz University

IJVR

ISSN: 1728-1997 (Print)
ISSN: 2252-0589 (Online)

Vol. 26 No. 1 Ser. No. 90 2025

IRANIAN JOURNAL OF VETERINARY RESEARCH

Scientific Report

Unusual histopathological features of a uterine leiomyosarcoma in a dog

Akhila, T. R.[1]; Ganesh, T. N.[2]; Azimah, A.[3]; Kok, M. K.[4] and Salleh, A.[4, 5*]

[1]MVSc in Veterinary Surgery, KM Global Animal Hospital, 5 Jalan Industri Batu Caves, 1/1, Taman Perindustrian Batu Caves, Selangor, Malaysia; [2]Ph.D. in Veterinary Surgery, KM Global Animal Hospital, 5 Jalan Industri Batu Caves, 1/1, Taman Perindustrian Batu Caves, Selangor, Malaysia; [3]DVM, KM Global Animal Hospital, 5 Jalan Industri Batu Caves, 1/1, Taman Perindustrian Batu Caves, Selangor, Malaysia; [4]Department of Veterinary Laboratory Diagnosis, Faculty of Veterinary Medicine, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia; [5]Department of Veterinary Science, Faculty of Veterinary Medicine, Universitas Airlangga, Surabaya, Indonesia

[*]***Correspondence:*** *A. Salleh, Department of Veterinary Laboratory Diagnosis, Faculty of Veterinary Medicine, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia. E-mail: annas@upm.edu.my*

10.22099/ijvr.2025.50585.7473

(Received 28 Jun 2024; revised version 3 Feb 2025; accepted 16 Feb 2025)

Abstract

Background: Uterine leiomyosarcoma is a malignant neoplasm arising from leiomyocytes in the uterus. **Case description:** A 12-year-old mixed-breed intact female dog was presented with vaginal bleeding for the past 2 weeks. Abdominal ultrasonography revealed a mixed echogenic mass in the uterus, and right lateral abdominal radiography showed a large mid-abdominal mass. Exploratory celiotomy was performed, revealing a solid mass in the entire uterine body and both uterine horns. Ovariohysterectomy was performed and the uterine mass sample was microscopically examined. **Findings/treatment and outcome:** The mass was composed of a dense proliferation of neoplastic cells that display two morphologic features; spindloid and epithelioid appearances. Additionally, chondroid-like matrix was noted. Both components were negative for cytokeratin AE1/AE3, and strongly positive for vimentin. The spindloid component was strongly positive for α-SMA, while the epithelioid-like component was moderately positive for alpha-smooth muscle actin. These findings are suggestive of a leiomyosarcoma. Although the recovery immediately following surgery was smooth and without complications, the dog's condition worsened over the following month, ultimately leading to euthanasia. **Conclusion:** This report describes the histopathologic features and clinical outcome of an unusual variant of uterine leiomyosarcoma in a dog.

Key words: Dog, Immunohistochemistry, Pathology, Uterine leiomyosarcoma

Introduction

In small animal veterinary medicine, occurrences of neoplasms in the female reproductive tract are infrequent. Uterine tumors are uncommon, constituting only 0.3%-0.4% of all tumors found in dogs (Patsikas *et al*., 2014). Diagnostic imaging plays a crucial role in identifying abdominal masses in the uterus and planning subsequent treatment. Nonetheless, the size and characteristics of sizable uterine masses can pose challenges in interpreting abdominal radiographs and ultrasound scans (Russo *et al*., 2021). In dogs, uterine neoplasms of epithelial origin such as adenoma, carcinoma, and choriocarcinoma are rarely encountered. Genital leiomyomas and leiomyosarcomas represent the most commonly encountered mesenchymal neoplasm, and complete excision is generally curative (Agnew and Maclachlan, 2017).

This report aims to document and discuss the unusual histopathological features observed in a rare case of uterine leiomyosarcoma in a dog, including the presence of an epithelioid component and chondroid-like matrix. These findings, which are not typically associated with canine uterine leiomyosarcomas, provide new insights into the potential diversity of this tumor type in veterinary pathology.

Case description

A 12-year-old mixed-breed intact female dog, weighing 13 kg, was presented to KM Global Animal Hospital with the history of vulvar bleeding for the past 2 weeks, inappetence, and general dullness. Abdominal palpation revealed a markedly distended abdomen, especially prominent in the mid-abdominal region, where a large, firm mass was detected. Complete blood count showed non-regenerative anemia and severe neutrophilia and leukocytosis (Table 1), while selected serum biochemical values were within reference limits.

Table 1: Hematology and serum biochemistry of the dog with an abdominal distension

Parameters (unit)	Reference values	Results
Red blood cell count (10^{12}/L)	5.65-8.87	4.19
Hematocrit (%)	37.3-61.7	24.00
Hemoglobin (g/dL)	13.1-20.5	8.7
White blood cell count (10^9/L)	5.05-16.76	38.65
Neutrophil count (10^9/L)	2.95-11.64	29.53
Lymphocyte count (10^9/L)	1.05-5.10	5.22
Monocyte count (10^9/L)	0.16-1.23	3.23
Platelet count (K/µL)	148-484	230
Creatinine (µmol/L)	44-159	64
Urea (mmol/L)	2.5-9.6	4.1
Alanine aminotransferase (U/L)	10-125	100
Alkaline phosphatase U/L	23-212	200

Radiography revealed a sizable, well-defined mass measuring 15 cm in diameter located in the mid-abdominal region (Fig. 1). Thoracic radiography showed no signs of metastasis. Ultrasonography revealed a large mass with mixed echogenicity, showing no distinct hyperechoic and hypoechoic regions. No fluid or calcification was identified within the mass. Exploratory celiotomy was performed under general anaesthesia, which included pre-medication with meloxicam (0.2 mg/kg), diazepam (0.2 mg/kg), and tramadol (4 mg/kg). Induction was achieved using propofol (10 mg/kg, administered to effect), followed by maintenance with isoflurane (2%) and additional propofol (10 mg/kg). A solid mass was observed in the uterine body and both uterine horns, necessitating ovariohysterectomy. The abdominal cavity was thoroughly lavaged, and the surgical incision was sutured in the routine manner.

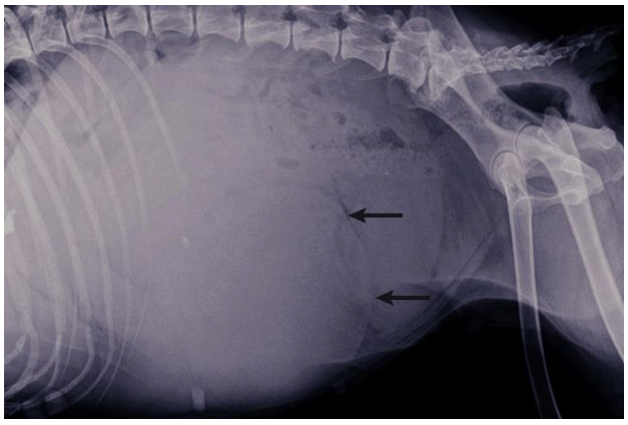

Fig. 1: Right lateral radiographic view of a dog with abdominal distension showing the presence of a large radiopaque mass 15 cm in diameter in the abdominal cavity (arrows)

Following removal of the mass, it was immediately examined and a sample of the mass was collected and fixed in 10% neutral-buffered formalin (Mazlan *et al*., 2021). Subsequently, it underwent routine tissue processing and sectioning for histopathology. The tissue sections were stained using the routine Hematoxylin and Eosin (H&E) stain. They were also subjected to indirect immunohistochemistry (IHC) for cytokeratin AE1/AE3, vimentin, and alpha-smooth muscle actin (α-SMA). This involved deparaffinization, rehydration, and heat-induced epitope retrieval in citrate buffer. Non-specific binding sites were blocked, followed by incubation with ready-to-use primary antibodies sourced from Roche Ventana (Fraser *et al*., 2024). The details of the antibodies are summarized in Table 2. After washing, a ready-to-use secondary antibody was applied, and the sections were then counterstained with methylene blue.

Table 2: Antibody details for indirect immunohistochemistry

Antibody	Clone	Species	Validation
Cytokeratin AE1/AE3	AE1/AE3	Mouse	Dog uterus
Vimentin	V9	Mouse	Dog uterus
Alpha-smooth muscle actin	1A4	Mouse	Dog uterus

The dog was maintained under parenteral fluid Ringers lactate and received antibiotics (Amoxicillin), analgesics (Tramadol), and an anti-inflammatory drug (Meloxicam) for one week. The dog showed a smooth recovery from surgery, evidenced by a return to normal appetite, increased activity level, and stable vital signs within the first few days postoperatively.

Findings/treatment and outcome

Gross and histopathological findings

The mass was examined, revealing a cream-colored, irregular cut surface (Fig. 2). The histopathological examination revealed extensive areas of necrosis within the mass. In viable areas, a biphasic pattern was observed characterized by a component populated by neoplastic spindle-shaped cells, and component populated by epithelioid neoplastic cells (Fig. 3a). In the epithelioid-like component, the neoplastic cells were arranged in sheets. These cells appeared large and markedly pleomorphic, with a polygonal shape, abundant eosinophilic cytoplasm, large nuclei, and one to two prominent nucleoli (Fig. 3b). Binucleation and giant nuclei were occasionally encountered. The mitotic count among these cells was 59 in 10 high-power fields (2.37 mm²). The spindle-shaped cells were arranged in an interwoven pattern, displaying moderate amount of eosinophilic cytoplasm, with small to large, round to oval hyperchromatic nuclei, each containing one or two small nucleoli (Fig. 3c). Binucleation was rarely noted. The mitotic count among these cells was 1 in 10 high-power fields (2.37 mm²). Additionally, a few chondroid-like matrix was observed (Fig. 3d). Lymphocytes and neutrophils were noted in low numbers within the spindle area, with slightly higher numbers in the epithelioid area. In both areas, these inflammatory cells were primarily distributed around blood vessels. Degenerated neutrophils were also observed in the areas of necrosis.

When using the epithelial marker CK AE1/AE3, cells from both components were negative (Fig. 4a). In contrast, when using vimentin, all neoplastic cells in both spindle and epithelioid-like components showed strong intracytoplasmic immunopositivity (Fig. 4b). In contrast,

with α-SMA, all spindle cells showed strong intracytoplasmic immunopositivity (Fig. 4c), while the epithelioid-like cells showed moderate intracytoplasmic immunopositivity (Fig. 4d).

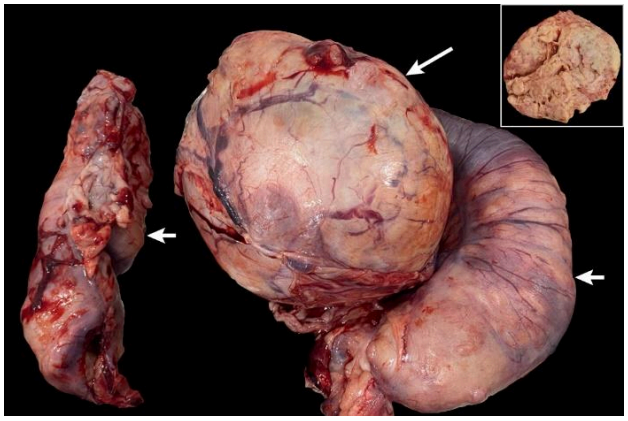

Fig. 2: Post-surgical examination of the excised female reproductive organs revealed generalized distension of the uterine body (long arrow) and both uterine horns (short arrows) by neoplastic tissue. The cut surface of the mass shows a cream-colored, irregular surface (inset)

Diagnosis and case progression

Based on the histopathological and immunohistochemical features, a diagnosis of uterine leiomyosarcoma was made. Three weeks post-surgery, the dog underwent reassessment, during which clinical and haematological examinations appeared normal. However, by the fourth week, the dog presented with signs of paralysis, weakness, lethargy, and inappetence persisting for one week. Upon examination, a large, hard mass was palpated in the right abdominal area that was suspected to be a regrowth of the neoplasm. Given the deteriorating condition of the dog and upon request by the owners, euthanasia was performed. The owner declined to conduct a necropsy on the carcass.

Discussion

This report describes a case of uterine leiomyosarcoma in a dog, which presented with notable clinical signs and specific histopathological features. Following surgery, the dog showed an initial smooth recovery; however, a month later, it exhibited signs of deterioration, including paralysis, weakness, and a palpable abdominal mass. While neoplasms of the female reproductive system are common in dogs, they are predominantly encountered in the vagina and vulva (Richards *et al*., 2001). Uterine neoplasms, on the other hand, are considered rare, accounting for only 0.4% of

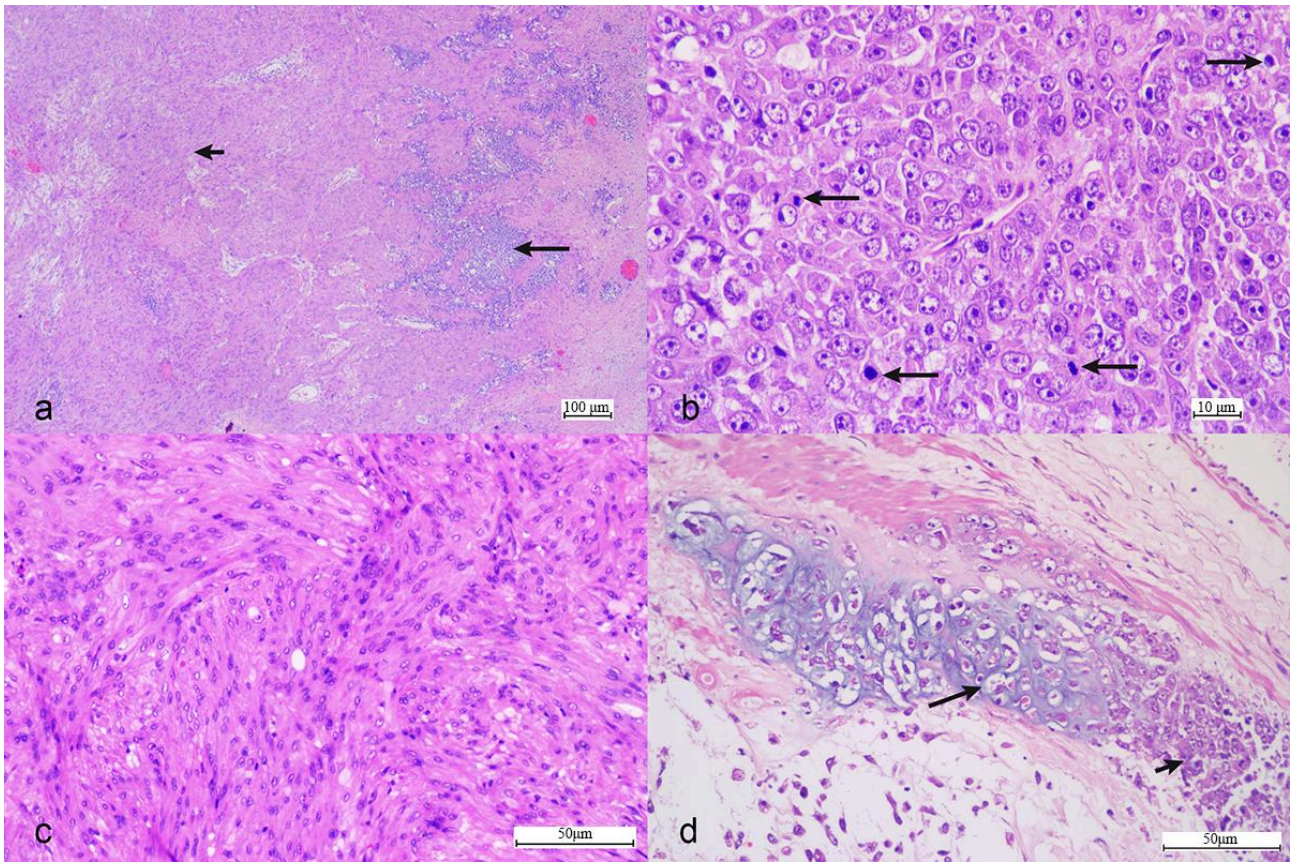

Fig. 3: Histopathology of a uterine neoplasm in a dog. (**a**) Neoplastic spindle cells (short arrow) and epithelioid-like cells (long arrow), (H&E, scale bar, 100 µm), (**b**) Epithelioid-like cells with marked pleomorphism, large nuclei, prominent nucleoli, and high mitosis (arrows), (H&E, scale bar, 10 µm), (**c**) Neoplastic mesenchymal cells displaying homogeneity, with spindle-shaped cells, moderate cytoplasm, and round to oval nuclei, (H&E, scale bar, 50 µm), and (**d**) Presence of chondroid-like matrix (long arrow) within the uterine neoplasm. Note the presence of the neoplastic epithelioid-like cells (short arrow), (H&E, scale bar, 50 µm)

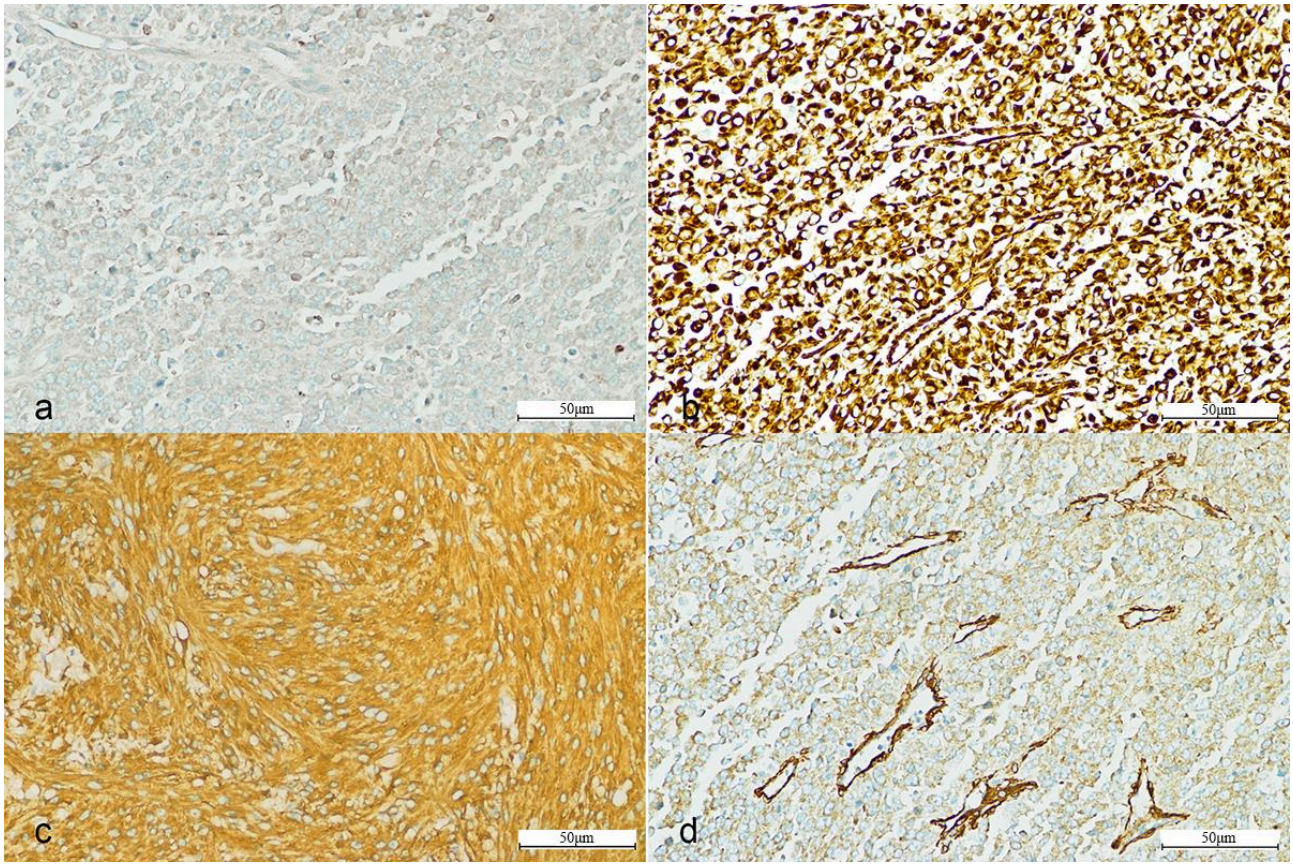

Fig. 4: Immunohistochemistry of a uterine neoplasm in a dog. (**a**) Neoplastic epithelioid-like cells are negative for CK AE1/AE3, (CK AE1/AE3, scale bar, 50 µm), (**b**) Neoplastic epithelioid-like cells are strongly positive for vimentin, (Vimentin, scale bar, 50 µm), (**c**) All neoplastic spindle cells show strong immunopositivity for α-SMA, (α-SMA, scale bar, 50 µm), and (**d**) Neoplastic epithelioid-like cells show moderate immunopositivity for α-SMA, (α-SMA, scale bar, 50 µm)

all canine tumors (Serin *et al*., 2010; Patsikas *et al*., 2014). The most frequently reported histopathological type of uterine tumors is leiomyoma, which comprises 85% to 95% of cases and is typically benign (Mohan *et al*., 2018; Kazmierczak *et al*., 2023).

The clinical manifestations of uterine neoplasms in dogs vary depending on factors such as tumor size, the presence of metastases, and concurrent conditions such as mucometra or pyometra (Pena *et al*., 2006; Tsioli *et al*., 2011). Clinical signs of uterine neoplasms often include the presence of an abdominal mass or abdominal distension, lethargy, haemorrhagic vaginal discharge, along with signs such as vomiting, loss of appetite, tenesmus, dullness, and dysuria (Serin *et al*., 2010; Tsioli *et al*., 2011; Barozzi *et al*., 2021; Kazmierczak *et al*., 2023). In the present case, the dog showed a subset of these signs - specifically abdominal mass, dullness, inappetence, and vaginal bleeding - aligning partially with documented presentations. Surgery remains the primary treatment approach for uterine neoplasms in dogs. However, there is a lack of studies investigating the effectiveness of supplementary chemotherapy or radiotherapy in canines (Kazmierczak *et al*., 2023), likely due to the rarity of this disease in the species.

The marked neutrophilia and leukocytosis observed in this case are notable findings that likely reflect an inflammatory response. The presence of lymphocytes and neutrophils, particularly in the vascular areas of the tumor, further supports the notion of an inflammatory milieu associated with the neoplasm. The relationship between inflammation and neoplasia is complex; inflammation may either precede or accompany tumor development (Greten and Grivennikov, 2019). For instance, canine mammary tumors have been shown to be induced by inflammatory cells that promote tumor formation (Carvalho *et al*., 2016). However, it is important to note that most tumors are not preceded by an inflammatory response (Greten and Grivennikov, 2019). This inflammation represents the interaction between the body and the tumor, a phenomenon known as tumor-elicited inflammation (Grivennikov *et al*., 2010).

In the present case, metastasis was not evident before the surgical procedure. However, the clinical signs observed by the owner a month after the surgery may suggest the possibility of metastasis. Unfortunately, this could not be confirmed as a necropsy was not performed.

Uterine leiomyosarcoma were previously reported in dogs. Their histopathological features range from well-differentiated spindle cells arranged in interweaving fascicles, with the cells showing eosinophilic cytoplasm and oval to elongate, nuclei that lack pleomorphism. Atypia may present, while mitotic count is typically low (Tsioli *et al*., 2011; Kazmierczak *et al*., 2023). This

description is common for well-differentiated uterine leiomyosarcoma, and fits well with the spindle component observed in this case. However, it does not match the epithelioid-like component seen in this case, thus, raising a suspicion of a different variant of leiomyosarcoma.

Unfortunately, subclassification of uterine leiomyosarcoma has not been described in veterinary medicine. The epithelioid-like component seen in this case is almost similar to an epithelioid leiomyoma previously described in a Sumatran rhinoceros (Salleh *et al*., 2020), with the exception in term of the number of mitotic figures. Based on the human medical nomenclature by World Health Organization (WHO), it is suggestive that the epithelioid component seen in this case is an epithelioid leiomyosarcoma. The current WHO subclassification of leiomyosarcoma includes: spindle cell leiomyosarcoma, epithelioid leiomyosarcoma, and myxoid leiomyosarcoma (Kurman *et al*., 2014).

The observed disparity in mitotic figures between the epithelioid and spindle components is noteworthy. The low mitotic index of the spindle component suggests a less aggressive nature. In contrast, the high mitotic count for the epithelioid component indicates a higher level of cellular proliferation, which is often associated with more aggressive tumor behavior. Higher mitotic activity typically correlates with an increased likelihood of metastatic spread, tumor recurrence, and a poorer clinical outcome (Bray, 2016). In human, epithelioid leiomyosarcoma of uterus are known to be aggressive (Chapel *et al*., 2022). Furthermore, tumor-related deaths in dogs with soft tissue sarcoma are more likely with >10% tumor necrosis (Liptak and Forrest, 2007). Therefore, the marked mitotic activity in the epithelioid component and extensive tumor necrosis may explain the dog's clinical decline observed postoperatively.

The presence of an epithelioid component and chondroid-like matrix within this leiomyosarcoma complicated the diagnostic process. Routine histopathology initially raised suspicion for uterine carcinosarcoma; however, IHC was crucial in ruling it out. The IHC results demonstrated that the neoplastic cells were negative for CK AE1/AE3, a common marker for epithelial-origin tumors, but positive for vimentin and α-SMA, which are markers indicative of mesenchymal origin and smooth muscle differentiation, respectively. This IHC profile supported the diagnosis of leiomyosarcoma over carcinosarcoma (Miettinen, 2014). The unique feature of a chondroid-like matrix may suggest a more complex biological behavior and highlights the importance of IHC in distinguishing between histopathologically similar neoplasms. Additionally, factors within the tumor microenvironment, such as hypoxia, inflammation, and specific growth factors, can influence tumor cell differentiation and extracellular matrix production (Maruyama *et al*., 2023), potentially contributing to the tumor's unique histopathological appearance.

While ultrasonography and radiographic imaging are valuable diagnostic tools for detecting uterine neoplasms (Hagman, 2014), histopathological examination is essential for arriving at a definitive diagnosis of uterine leiomyosarcoma. Routine histopathology is typically adequate for diagnosis. However, with rare neoplasms and different variants being reported in uterine neoplasms of animals (Boisclair and Dore, 2001; Percival *et al*., 2018; Salleh *et al*., 2020). Employment of IHC may aid in arriving at an accurate diagnosis. This report describes the histopathologic features and clinical outcome of an unusual uterine leiomyosarcoma in a dog, distinguished by the presence of both epithelioid differentiation and a chondroid-like matrix, features that likely contributed to its aggressive biological behavior.

Acknowledgements

The authors would like to thank all the staff of KM Global Animal Hospital, Malaysia for their support in treating this case.

Conflict of interest

The authors declared no conflicts of interest with respect to the research, authorship or publication of this article.

References

Agnew, DW and Maclachlan, NJ (2017). Tumors of the genital systems. In: Meuten, DJ (Ed.), *Tumors in domestic animals*. (5th Edn.), Ames, Iowa, John Wiley & Sons. PP: 689-722.

Barozzi, MCM; Saba, CF and Gendron, KP (2021). CT characteristics of uterine and vaginal mesenchymal tumours in dogs. J. Small Anim. Pract., 62: 293-299.

Boisclair, J and Dore, M (2001). Uterine angiolipoleiomyoma in a dog. Vet. Pathol., 38: 726-728.

Bray, JP (2016). Soft tissue sarcoma in the dog - Part 1: A current review. J. Small Anim. Pract., 57: 510-519.

Carvalho, MI; Silva-Carvalho, R; Pires, I; Prada, J; Bianchini, R; Jensen-Jarolim, E and Queiroga, FL (2016). A comparative approach of tumor-associated inflammation in mammary cancer between humans and dogs. Biomed. Res. Int., 2016: 4917387.

Chapel, DB; Nucci, MR; Quade, BJ and Parra-Herran, C (2022). Epithelioid leiomyosarcoma of the uterus: modern outcome-based appraisal of diagnostic criteria in a large institutional series. Am. J. Surg. Pathol., 46: 464-475.

Fraser, C; Keong Kok, M; Shameha Abdul Razak, I; Puspitasari, Y and Salleh, A (2024). Diagnostic challenge in veterinary pathology: Metastatic mammary tumor in a female tiger (*Panthera tigris*). Vet. Pathol., 61: 508-511.

Greten, FR and Grivennikov, SI (2019). Inflammation and cancer: triggers, mechanisms, and consequences. Immunity. 51: 27-41.

Grivennikov, SI; Greten, FR and Karin, M (2010). Immunity, inflammation, and cancer. Cell. 140: 883-899.

Hagman, R (2014). Diagnostic and prognostic markers for uterine diseases in dogs. Reprod. Domest. Anim., 49: 16-20.

Kazmierczak, J; Sugai, NJ; Withowski, KE; Jonatan, A;

LeRoith, T and Cecere, JT (2023). Case report: A case of oviductal and uterine leiomyosarcoma in an 11-year-old dog. Front. Vet. Sci., 10: 1227799.

Kurman, RJ; Carcangiu, ML and Herrington, CS (2014). *WHO classification of tumours of female reproductive organs*. 4th Edn., Lyon, France: International Agency for Research on Cancer.

Liptak, JM and Forrest, LJ (2007). Soft tissue sarcomas. In: Withrow, SJ and MacEwen, EG (Eds.), *Withrow and MacEwen's small animal clinical oncology*. (4th Edn.), St. Louis, Missouri: Elsevier. PP: 425-454.

Maruyama, S; Yamazaki, M; Abé, T; Cheng, J; Saku, T and Tanuma, JI (2023). Hypoxia-induced biosynthesis of the extracellular matrix molecules, perlecan and fibronectin, promotes the growth of pleomorphic adenoma cells *in vitro* models. Biomedicines. 11: 2981.

Mazlan, M; Khairani-Bejo, S; Hamzah, H; Nasruddin, NS; Salleh, A and Zamri-Saad, M (2021). Pathological changes, distribution and detection of *Brucella melitensis* in foetuses of experimentally-infected does. Vet. Q., 41: 36-49.

Miettinen, M (2014). Immunohistochemistry of soft tissue tumours–review with emphasis on 10 markers. Histopathology. 64: 101-118.

Mohan, D; Maiti, SK; Shivaraju, S; Ramith, KR; Sangeetha, P; Bindhuja, BV and Kumar, N (2018). Surgical management of multiple uterine tumours in a dog. MOJ Anat. Physiol., 5: 117-118.

Patsikas, M; Papazoglou, LG; Jakovljevic, S; Papaioannou, NG; Papadopoulou, PL; Soultani, CB; Chryssogonidis, IA; Kouskouras, KA; Tziris, NE and Charitanti, AA (2014). Radiographic and ultrasonographic findings of uterine neoplasms in nine dogs. J. Am. Anim. Hosp. Assoc.; 50: 330-337.

Pena, FJ; Gines, JA; Duque, J; Vieitez, V; Martinez-Perez, R; Madejón, L; Martinez, IN; Moran, JM and Fernández-García, S (2006). Endometrial adeno-carcinoma and mucometra in a 6-year-old Alaska Malamute dog. Reprod. Domest. Anim., 41: 189-190.

Percival, A; Singh, A; Zur Linden, RA; Watrous, G; Patten, S; Valverde, A and Ratsep, E (2018). Massive uterine lipoleiomyoma and leiomyoma in a miniature poodle bitch. Can. Vet. J., 59: 845-850.

Richards, HG; McNeil, PE; Thompson, H and Reid, SWJ (2001). An epidemiological analysis of a canine-biopsies database compiled by a diagnostic histopathology service. Prev. Vet. Med., 51: 125-136.

Russo, M; England, GCW; Catone, G and Marino, G (2021). Imaging of canine neoplastic reproductive disorders. Animals. 11: 1213.

Salleh, A; Zainuddin, ZZ; Mohamed Tarmizi, MR; Yap, KC and Zamri-Saad, M (2020). Two variants of uterine leiomyoma in Malaysia's last Sumatran rhinoceros (*Dicerorhinus sumatrensis*). Vet. Q., 40: 250-257.

Serin, G; Aydogan, A; Yaygingul, R and Tunca, R (2010). Uterine leiomyosarcoma in a dog: a case report. Vet. Med. (Praha), 55: 405-408.

Tsioli, VG; Gouletsou, PG; Loukopoulos, P; Zavlaris, M and Galatos, AD (2011). Uterine leiomyosarcoma and pyometra in a dog. J. Small Anim. Pract., 52: 121-124.