

## Infiltrative Rectal Adenocarcinoma in a Dog

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### ABSTRACT

**Background:** Intestinal neoplasms are uncommon in dogs and adenocarcinoma is the main histological type found. This neoplasm presents slow growth and high capacity of causing metastasis. Histologically speaking, neoplasm cells can present solid, tubular, papillary arrangement and note amorphous extra-cellular material. Clinically observed tenesmus, diarrhea, dyskinesia, hematochezia, mane, protrusion of the anus, weight loss, anorexia. The occurrence and clinicopathological aspects of tumors in dogs' gastrointestinal tract, the rectal segment, remains poorly understood. Accordingly, the aim of the present study is to report a case on infiltrative rectal adenocarcinoma diagnosed in a dog.

**Case:** A 7-year-old male dog representative of the Fila Brasileiro breed was presented to the Veterinary Hospital of University Federal Rural of Amazonia, with history of hyperthermia, anorexia, apathy and tenesmus. Imaging examinations depicted prostatomegaly. Exploratory laparotomy was performed and showed the thickening and hardening of the rectum segment. The animal was subjected to euthanasia. Necropsy showed increased rectal perimeter; the mucosa in its opening presented atypical cerebriform aspect and irregular surface, and areas dark red. The rectal segment depicted a thick wall of white color, irregular limits covering the muscular and adjacent sub-mucosa. The peri-rectal adipose tissue presented poor delimitation with the rectum, multiple greyish and reddish areas. Increased prostate and iliac lymph, and multi node of regular limits in the lungs. The histology of the rectal tissue depicted epithelium with differentiated neoformation, composed of atypical cells; nuclear anisocytosis, anisocariasis and hyperchromasia placed in small islands, cords or tubular formation. Neoplasm growth was unorganized and of infiltrative character. Some areas presented mucosal pattern cells with Signet Ring morphologic. Multiple rectal blood vessels, regional lymph nodes and lungs had neoplasm growth similar to that observed in the intestine. Mucosa also presented ulceration areas and lymphoplasmacytic infiltrate. There was fibroplasia, lymphoplasmacytic points and bleeding in the serous, as well as in the peri-rectal fat tissue. The immunohistochemical technique showed immunostaining in cytokeratin and vimentin antibodies, and in marked epithelial cells and tumor stroma markings, respectively.

**Discussion:** The intestinal tumor diagnosis in dogs is found by associating history, clinical signs, radiographic, ultrasound findings and necropsy. Only one data about the occurrence of rectal adenocarcinoma in Fila Brasileiro specimens. With regards to sex, results were similar to those record, whose males presented higher prevalence of primary rectal tumors. The macroscopic characteristic is consistent with infiltrative neoplasms; thickening was related to the presence of the tumor. The histopathological findings evidenced growing infiltrative neoplasm formed by atypical cells of tubular arrangement. Microscopy featured a chronic ulcerative colitis frame, such alteration represents one of the main risk factors for colon rectal cancer in humans. Epithelial histogenesis was confirmed through immunohistochemical results that have revealed co-expression of the cytokine epithelial marker in most tumor cells. The vimentin mesenchymal marker in the neoplasm stroma was positive, fact that can be explained by occasional immune-reaction in the anti-bodies (cytokine and vimentin) and in non-differentiated carcinomas. The prognostic was negative in the current report. Such outcome was attributed to the infiltrative character observed in the trans-operative period. Necropsy, as well as the histopathological and immunohistochemical exams, confirmed the infiltrative rectal adenocarcinoma in the dog.

**Keywords:** Adenocarcinoma, intestinal neoplasm, rectum, dog.

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## INTRODUCTION

The digestive tract in dogs can present a whole variety of neoplasms [16,19,21]; however, such neoplasms have low incidence in this animal species [6], they represent from 2% [16] to 5% of the number of described neoplasms [21]. Adenocarcinoma is the most prevalent intestinal tumor [4,5,10,13,16,20] it mainly occurs in the large intestine [5,6,21], mainly in the rectal segment [3,10,13,19].

Rectum adenocarcinoma is an uncommon malignant intestinal neoplasm in dogs [1,7,16]. It originates in mucosa epithelial cells [11] and is of slow growth, despite its high capacity to cause metastasis in regional lymph nodes [8,10], in the spleen, liver, pancreas [8] and lungs [8,13]. Besides, it can be infiltrative, ulcerative and proliferative. The infiltrative type invades the rectal wall and causes fibrosis, and peristalsis obstruction and flaw [8,11]. The ulcerative type leads to strong, ulcerate and thick injuries, whereas the proliferative type has pedunculated, multiple and friable character [11].

Histologically speaking, neoplasm cells can present solid [7], tubular [7,13,10], papillary [5] or tubopapillary [3,7] arrangement. Moreover, it is possible observing amorphous extra-cellular material, which is characteristic of mucin in the tubular lumen [3,10,13]. Disorders such as tenesmus, diarrhea, dyskinesia [4,5,14,20], hematochezia [4,5], mane [3,4] and protrusion of the anus may occur. Other clinical signs such as weight loss, anorexia and apathy can be seen in the late stages of neoplastic growth [14].

The Brazilian reality about the occurrence and clinicopathological aspects of intestinal tumors in dogs, mainly in the rectal segment, remains little known. It likely happens due to its hard diagnostic and unspecific clinical manifestations, which can be misinterpreted, just as other diseases in the digestive tract. Accordingly, the aim of the present study was to report the clinical, macroscopic, histological and immunohistochemical aspects of an infiltrative rectal adenocarcinoma case in a canine specimen.

## CASE

A 7-year-old male canine specimen representative of the Fila Brasileiro breed was presented at the Veterinary Hospital of Universidade Federal Rural da Amazônia (UFRA) with clinical signs of hyperthermia, appetite loss, anorexia, apathy and tenesmus. Among the performed complementary exams, abdominal

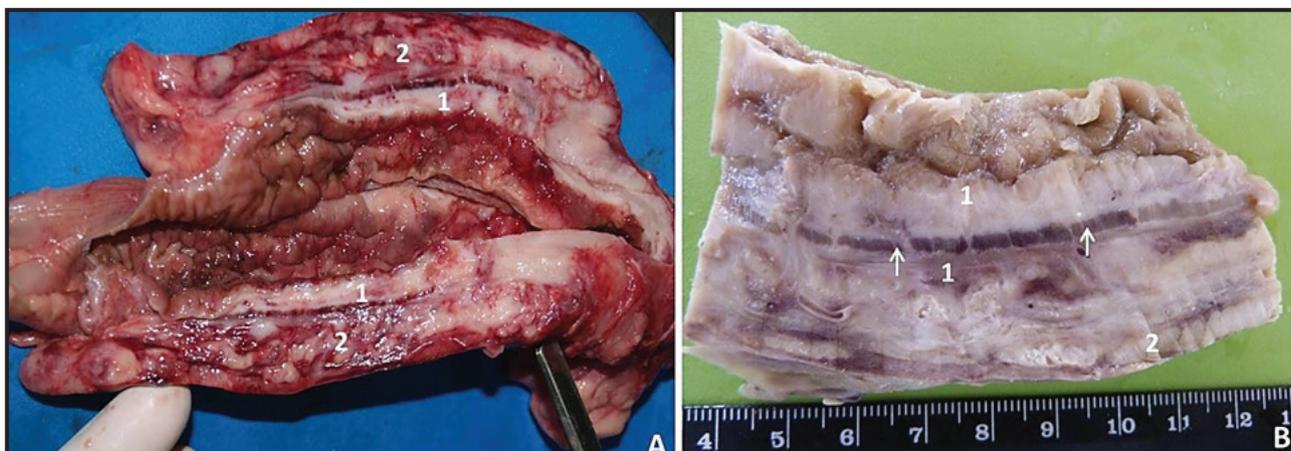
ultrasound depicted prostate with increased volume and iliac lymph nodes. The radiographs of the pelvic region showed descendant colon dilatation filled with gas content and it is narrowing due to the obstruction of its caudal portion, cranially to the prostate, which suggests prostatomegaly.

Based on these exams, the choice was made to perform a surgical exploratory laparotomy protocol, which evidenced thickening and diffuse rigidity of the rectum. However, it was not possible conducting any surgical intervention in order to recover the rectal function. The animal was subjected to euthanasia and sent to the Animal Pathology Laboratory of UFRA after veterinary advice and the owner's authorization, to undergo necroscopy and histopathological examinations.

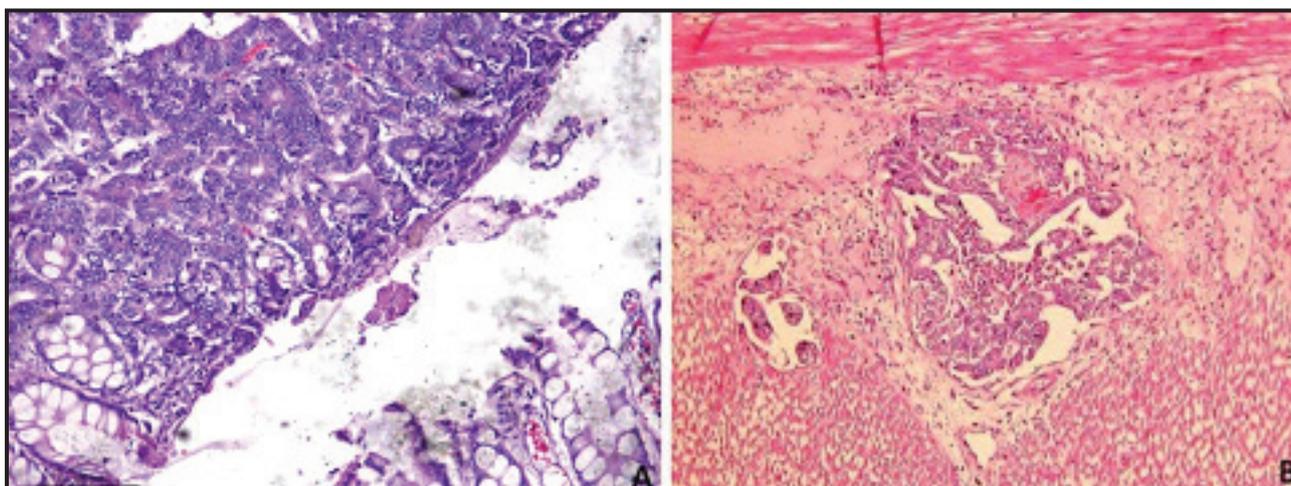
Necroscopy showed increased rectal perimeter; the mucosa in its opening presented atypical cerebroid aspect and irregular surface, and areas of multiple colors, from light to dark red (Figure 1A). The longitudinal cut of the rectal segment depicted a thick wall of opaque white color, irregular and unprecise limits covering the muscular and adjacent sub-mucosa (Figure 1). The peri-rectal adipose tissue presented poor delimitation with the rectum (Figure 1B), little elastic consistency and multiple poor-defined greyish and reddish areas in the cut (Figure 1A). Increased prostate and iliac lymph nodes were observed, as well as smooth surface, and multi nodes of regular limits in the lungs (up to 2 mm).

Rectum fragments were collected during necropsy and fixed in 10% buffered formalin. Subsequently, the samples were cleaved, routinely processed, included in paraffin, cut at thickness of 5 micrometers and inked with hematoxylin and eosin (HE) in order to perform the histopathological examinations.

The histology of the rectal tissue depicted epithelium with differentiated neof ormation, mainly in the crypts, composed of atypical cells; nuclear anisocytosis, anisocariasis and hyperchromasia placed in small islands, cords or tubular formations (Figure 2A). Neoplasm growth was unorganized and of infiltrative character; it established itself in the sub-mucosa, in the inner muscular layer, in the lymphatic vessels of intermuscular connective tissues (Figure 2B) and in the lumen of sub-mucosa capillaries. Some areas presented mucosal pattern cells with Signal Ring morphology; besides, mitotic figures were frequent. Multiple rectal blood vessels, regional lymph nodes and lungs had neoplasm growth similar to that observed in the intestine.



**Figure 1.** Macroscopic aspect of rectal adenocarcinoma in a dog. A- Atypical cerebroid aspect of the mucosa with multiple red areas. The thickened and mostly opaque white rectal wall on the surface of the cut (1). Peri-rectal fat (2) with poorly delimited multiple greyish and reddish areas. B- Thick and whitish intestinal wall with better visualization of the muscular due to hypertrophy (arrows) and discontinuity in neoplasm infiltration areas. There is no perception about the limits of the rectum (1) and the peri-rectal adipose tissue (2).



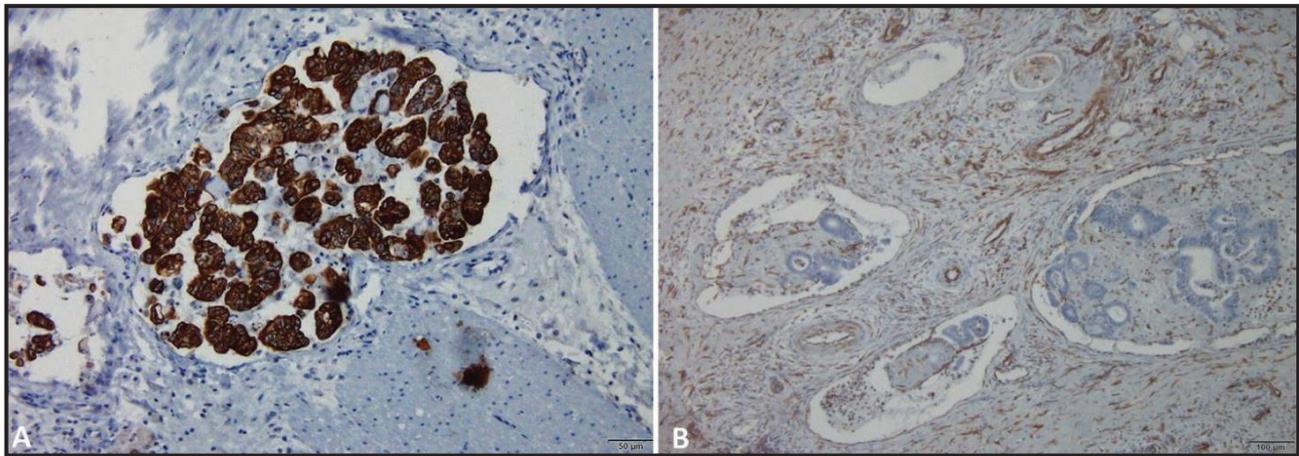
**Figure 2.** Histology of rectal adenocarcinoma in dogs. A- The rectal mucosa presents the neoplasm growth of atypical epithelial cells with nuclear anisocytosis, anisocariosis and hyperchromasia in tubular formation [Bar= 100  $\mu$ m]. B- The connective tissue between the muscle layers present lymphatic vessels with neoplasm growth of tubular pattern epithelial cells [Bar= 100  $\mu$ m].

Mucosa also presented ulceration areas and lymphoplasmacytic infiltrate in the slide itself. There was fibroplasia, lymphoplasmacytic points and bleeding (chronic hemorrhagic ulcerative colitis) in the serous, as well as in the peri-rectal fat tissue, these symptoms were associated with collections of piocytes, which characterize severe purulent soapstone.

The histological cuts of the tumor were subjected to the immunohistochemical technique, which was performed in the Animal Pathology Laboratory of Universidade Estadual Paulista (UNESP), Jaboticabal Campus. Cytokeratin antibodies at dilution 1:200 (clone AE1/AE3, DakoCytomation, COD. M3515)<sup>1</sup> and vimentin at dilution 1:150 (clone V9, DakoCytomation, COD. M0725)<sup>1</sup> were used. Antigenic recovery was performed through heating by using Pascal pressure

chamber (DakoCytomation, model S2800)<sup>1</sup>, in sodium citrate buffer solution 10 mM (pH 6.0); the inhibition of unspecific reactions was conducted through double blocking.

Streptavidin-biotin-peroxidase (kit LSAB +System HRP, DakoCytomation, COD. K0690-1)<sup>1</sup> was used to reveal the reactions; the cuts were counter stained with Harris hematoxylin. A remarkable marking degree was observed in immunohistochemical evaluation of the rectal adenocarcinoma by using AE1/AE2 cytokeratin; tumor epithelial cells were immune-marked in the rectal tissue layers in blood vessels (Figure 3A). Vimentin V9 immune-marks were observed in the tumor stroma, although it was absent in the epithelial cells (Figure 3B).



**Figure 3.** Rectal adenocarcinoma in a dog. Immunohistochemistry of AE1/AE2 cytokine with contrasting hematoxylin. A- All the neoplastic epithelial cells were positive for cytokeratin (HE) [Bar= 50 µm]. B- Immunostaining of the tumor stroma for vimentin V9, with contrasting hematoxylin [Bar= 100 µm].

### DISCUSSION

The intestinal tumor diagnosis in dogs is found by associating history and clinical signs, complementary exams, mainly imaging examinations, and histopathological analysis [17]. The radiographic and ultrasound findings of the herein reported case were not relevant for tumor detection, since they just depicted suggestive signs of lymphadenomegaly and prostatomegaly, which were proved real during the necropsy. Thus, the anatomopathological findings were essential for the neoplasm diagnosis [3,10].

Although the clinical manifestations of hyperthermia, weight loss, anorexia, apathy and tenesmus are unspecific, they corroborate the symptomatology described for rectal tumors, mainly in the latest stages of the disease [4,14,20].

Only one data about the occurrence of rectal adenocarcinoma in Fila Brasileiro specimens was published so far [14]. This disease was also detected in the herein reported case. However, other cases in the literature describe a larger number of rectal neoplasms in the German Shepherd breed, in dogs of undefined breed and in Doberman Pinscher [4].

With regards to sex and age, results were similar to those recorded by Danova *et al.* [4], in a study conducted with 23 canine specimens, whose males presented higher prevalence of primary rectal tumors than females; the animals were 8 years old, on average. On the other hand, Jelinek [10] proved that females prevailed among the fourteen dogs diagnosed with these neoplasms.

The macroscopic characteristic is consistent with infiltrative neoplasms [8,11], since all the

intestinal layers were affected; thickening was related to the presence of the tumor, which caused lumen space reduction with possible obstruction frame, fact that can be explained by the tenesmus found in the herein assessed animal.

The histopathological findings comply with others described by Damasceno *et al.* [3], who evidenced growing infiltrative neoplasm formed by atypical cells of tubular arrangement. However, the aforementioned authors found cells with tubopapillary disposition, but Ferreira *et al.* [5] detected the papillary sub-type in the descending colon in a canine specimen belonging to the Beagle breed. It highlights the importance of the uncommon occurrence of the type of tubular neoplasms in the large intestine.

Microscopy featured a chronic ulcerative colitis frame; according to the National Cancer Institute [9], such alteration represents one of the main risk factors for colon rectal cancer in humans. Other studies conducted with dogs also showed colitis associated with adenocarcinoma, although in the descending colon [5]. Therefore, the chronic intestinal inflammatory process can also be a risk factor for canine species.

Metastases were detected in the blood vessels of the sub-mucosa, in the lymphatic vessels of the intermuscular connective tissue of the rectum, in the lungs and in regional lymph nodes, which are rare sites for neoplasm cells of rectal adenocarcinoma, since the reported cases so far refer to metastases in regional lymph nodes [8,10], spleen, liver, pancreas [8] and lungs [8,13].

Epithelial histogenesis was confirmed through immunohistochemical results that have revealed

co-expression of the cytokine epithelial marker in most tumor cells, thus showing that the immunohistochemical technique has the potential to add to the histopathological diagnosis. However, its use to diagnose rectal adenocarcinoma in dogs remains scarce [3,10]. The vimentin mesenchymal marker in the neoplasm stroma was positive, fact that can be explained by occasional immune-reaction in the anti-bodies (cytokine and vimentin) and in non-differentiated carcinomas [15,18].

Well-differentiated breast neoplasm in dogs can be positive in cytokeratin, but not in vimentin, whereas anaplastic carcinomas express these two anti-bodies [15]. Therefore, they can be important markers for non-differentiated carcinomas that often lead to non-conclusive clinical and histopathological diagnoses [18].

Another possible interpretation for the intense vimentin expression in the present study is related to the neoplasm site, because the rate of vimentin positivity in this neoplasia in other tumors such as the

rectal carcinoma in humans is approximately five times higher than in other gastro-intestinal carcinoids [12].

The prognosis of rectum adenocarcinoma is reserved and related to the macroscopic aspects of the tumor, so that the survival rate is higher for polypoid, pendunculated and isolated masses [12]. The prognostic was negative in the current report, it was not possible adopting the surgical treatment to remove the tumor. Such outcome was attributed to the infiltrative character observed in the trans-operative period.

The recorded clinical signs of necropsy, the histopathological and immunohistochemical results did not meet the differentiated rectal adenocarcinoma case of the infiltrative type found in a canine specimen.

#### MANUFACTURER

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**Declaration of interest.** The authors report no conflicts of interest.

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