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Case Report

Primary muscle hypertrophy of the colon in a cat: a rare presentation

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Abstract

In animals and humans, intestinal muscular hypertrophy has only been observed in the small intestine, and this appears to be the first report of the disease affecting the large intestine. A rare case of primary muscle hypertrophy of the colon was found during the necropsy of a male, 3-year-old, mixed breed cat that died due to poisoning by carbamate. The necropsy revealed that the entire colon had marked circumferential thickening with narrowing of the lumen. Microscopically, diffuse intense smooth muscle hypertrophy was observed, especially of the inner circular muscle layer and muscularis mucosa. Those finds were associated with moderate multifocal to coalescing lymphocytic inflammatory infiltrate. Smooth muscle fibers of the large and small intestine showed no CDC47 expression and the percentage of nuclei in the muscle layers was similar between the colon and the duodenum. Based on macroscopic, microscopic and immunohistochemical findings, the diagnosis of diffuse primary muscle hypertrophy of the colon was confirmed.

Key words: large intestine, hypertrophy, muscle, cat.

Introduction

The natural occurrence of intestinal muscle hypertrophy is uncommon in animals (1-7) and humans (8-9). This disease is characterized by a thickening of the muscular layer due to an increase in the diameter of muscle fibers without increasing mitosis or nuclei number (10).

Intestinal muscle hypertrophy can be classified as primary or idiopathic when it is not associated with any injury that predisposes the muscle to hypertrophy, or it may be secondary when it occurs as a result of stenosis or chronic partial obstruction due to external or internal factors that do not completely obstruct the flow of intake, but this increases the level of work with a consequent hypertrophy of muscle cells (11-12). In cats, there are reports of 13 cases of muscular hypertrophy of the small intestine. Bettini et al. (12) reported four cases of idiopathic hypertrophy and five cases of secondary to lymphoma and intestinal adenocarcinoma. Diana et al. (13) reported four cases of intestinal muscle hypertrophy, two associated with chronic enteritis and two with lymphoma and obstruction by a foreign body. In animals and humans, intestinal muscular hypertrophy has only been observed in the small intestine (1-8; 12,13). This appears to be the first report of the disease affecting the large intestine.

The objective of this report is to describe the pathological findings of primary muscle hypertrophy in the colon of a cat.

Case Report

A male, 3-year-old, mixed breed cat with a history of sudden death was admitted to the Pathology Department at the Veterinary Hospital of *Universidade Federal de Minas Gerais* (UFMG) for necropsy. According to the owner, the animal was well, eating, urinating and defecating properly. The cat had no medical history of changes in the gastrointestinal tract or any other disease. The owner suspected that the cat had eaten rat poison because he suddenly presented sialorrhoea and muscle tremors and died before the clinical examination. Because poisoning was suspected, a necropsy was performed.

At necropsy, the animal showed good nutritional condition. The entire colon had marked circumferential thickening, intense roughness, areas with detachment of the mucosa and intense lumen narrowing (Fig. 1). Throughout the intestine, there was a discrete amount of soft and yellowish feces. The small intestine showed only slight roughness of the mucosa, and the mesenteric lymph nodes did not show any apparent alteration. The lungs and kidneys were moderately congested, and the liver had multifocal pale areas with evident lobular pattern. Inside the stomach, there was a discrete amount of intake with a discrete number of small black granules. The other organs presented no gross lesions.

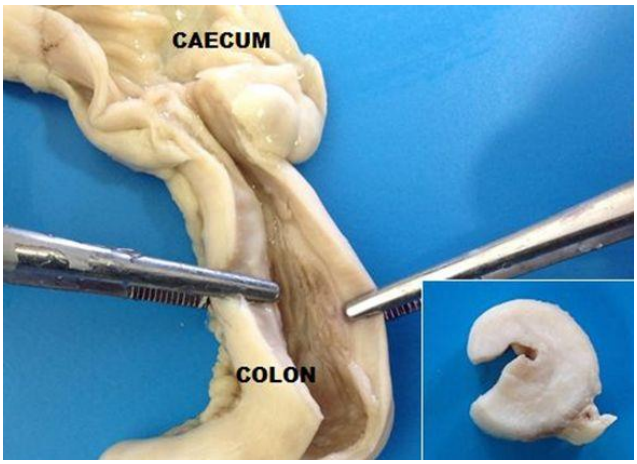

Figure 1. Idiopathic muscle hypertrophy of the colon in a cat. Macroscopy of the colon with intensely thick wall and moderately rough mucosa.

Fragments of the entire gastrointestinal tract, lymph nodes and other abdominal and thoracic organs were fixed in 10% neutral phosphate-buffered formalin, embedded in paraffin, cut at 5 µm, and stained with haematoxylin-eosin (HE) for histopathological analysis. Histological sections of the duodenum and colon stained by HE were subjected to histomorphometric analysis. Ten random fields of the inner and outer muscular layer in the duodenum and colon were analysed with the aid of a 121-point graticule attached to the microscope under the 40× objective for comparing the percentage of nuclei in the leiomyocytes of the normal segment with those of the altered segment. The mean percentage of nuclei in each layer of each segment was obtained. Besides, the thickness of intestinal wall and the percentage of each layer (mucosa, submucosa and muscular) was determined at 10 points with the aid of a micrometer eyepiece and under the X4 objective. The values of the thickness were converted to mm using a micrometric slide.

Fragments of the liver, kidney, abdominal fat and gastric contents were sent under refrigeration to the Toxicological Research Laboratory at the Veterinary School of UFMG for drug testing.

Microscopically, the entire colon showed diffuse and intense smooth muscle hypertrophy characterized by an intense increase in the thickness of the inner circular muscular layer and muscularis mucosa (Fig. 2A). In some regions, the smooth muscle fibers of the inner circular layer and muscularis mucosa were misdirected and interposed (Fig. 2B), making it difficult to individualize the layers. In addition, moderate multifocal to coalescing lymphocytic inflammatory infiltrate with mild fibroplasia was observed in all layers of the colon (Fig. 2C and E). Unlike the large intestine, the small intestine (duodenum, jejunum, terminal ileum) did not show any alteration of smooth muscle layers. The only change was the presence of a discrete to moderate lymphocytic inflammatory infiltrate in the mucosa and submucosa. The mean nuclei percentage of leiomyocytes was similar in the large and small intestines. The inner and outer muscle layers of the small intestine had a mean nuclei percentage of 16.69 ± 3.77 and 24.24 ± 4.92, respectively, while the layers of the large intestine had a mean percentage of 18.92 ± 1.96 and 18.84 ± 5.31, respectively. However, the large intestine was 3.5X thicker than the small intestine (4.42 ± 0.61 mm and 1.27 ± 0.11 mm, respectively), with a higher percentage of muscular in comparison with the small intestine [large intestine = 34.55 ± 6.17% (mucosa); 9.78 ± 5.08% (submucosa); 55.66 ± 3.85% (muscular); small intestine = 57.27 ± 5.91% (mucosa); 5.58 ± 0.94% (submucosa); 37.14 ± 6.29% (muscular)].

The liver showed moderate multifocal vacuolar degeneration and the lungs and kidneys had moderate congestion. The rest of the digestive tract, mesenteric lymph nodes and other organs showed no significant microscopic alterations.

The cell proliferation rate of the small and large intestine was analysed by CDC47 immunohistochemistry expression. For this, the biotin-streptavidin peroxidase (Streptavidin Peroxidase; Lab Vision Corp., Fremont, CA, USA) technique was used. Antigenic recovery by heat at 98^{O}C using a retrieval solution was performed for 20 minutes. Histological sections were incubated overnight in a humidified chamber with anti-CDC47 antibody (1:50) (47DC141; Neomarkers, Fremont, CA, USA). The sections were incubated for 30 minutes for each of the following steps: blocking endogenous peroxidase, blocking serum

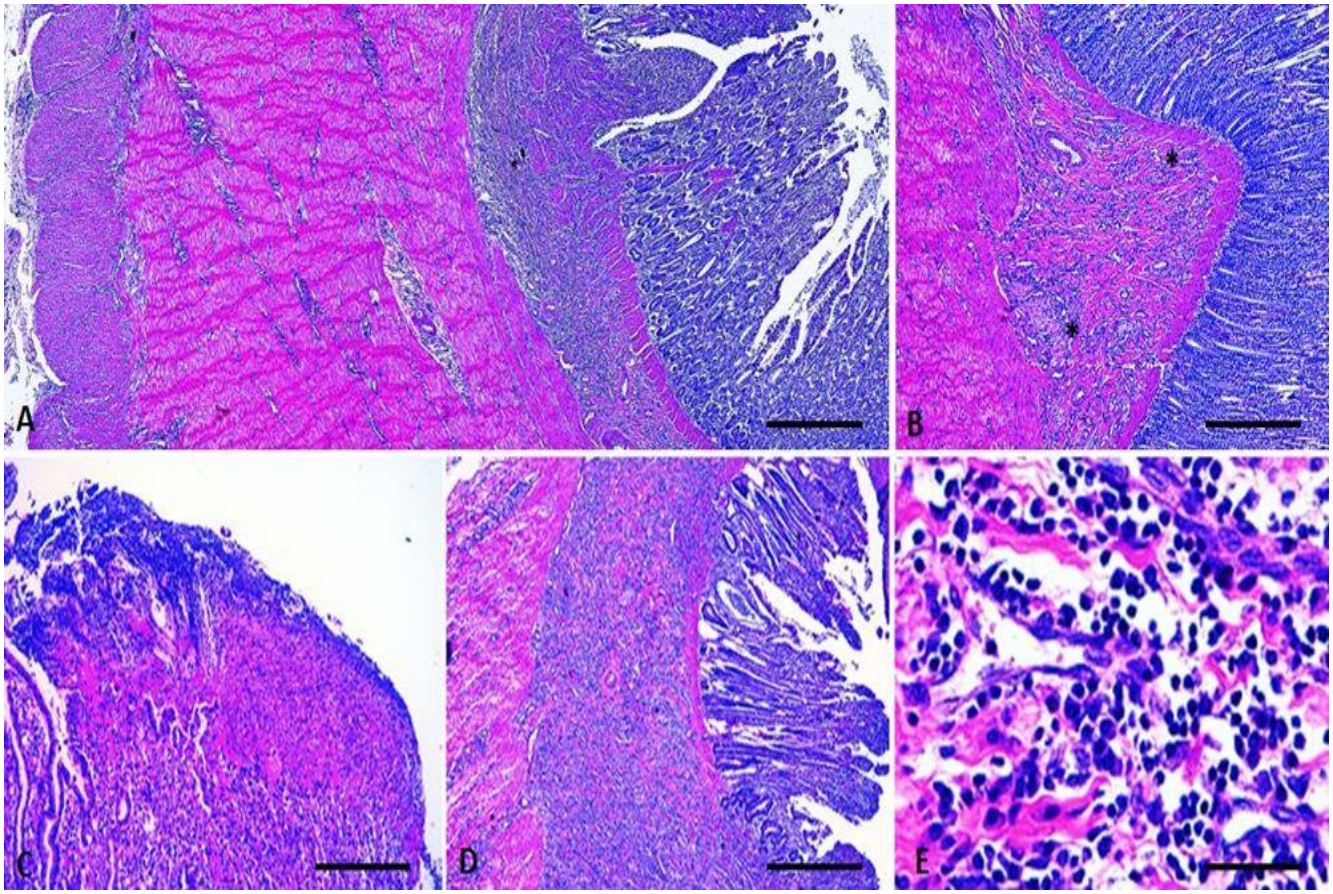

Figure 2. Idiopathic muscle hypertrophy of the colon in a cat. A) Colon with severe thickening of the inner circular and outer longitudinal muscle layers. B) Smooth muscle fibers of the inner circular layer and muscularis mucosa (asterisk) misdirected and interposed. C-E) Moderate multifocal to coalescing lymphocytic inflammatory infiltrate in mucosa (C) and muscular (D-E). (Hematoxylin and eosin staining; bar = 360 µm (A-D); 56 µm (E)).

(Ultra Vision Block; Lab Vision Corp., Fremont, CA, USA) and streptavidin peroxidase. Incubation with the secondary antibody (goat biotin; Lab Vision Corp., Fremont, CA, USA) was performed for 45 minutes. The chromogen used was diaminobenzidine (DAB substrate system; Lab Vision Corp., Fremont, CA, USA). The sections were counterstained with Harris haematoxylin (Harris Hematoxylin, Merck, Darmstadt, DE). A negative control was obtained by replacing the primary antibody with IgG. As positive control, we used the intestine fragment from the animal because crypt cells have CDC47 expression. Immunohistochemistry revealed no CDC47 expression in the inner and outer muscle layers of the small and large intestine, unlike that of the mucosa where strong CDC47 expression was observed in several crypt cells and in some epithelial and lamina propria cells (Fig. 3).

The toxicological examination of the animal confirmed that the death was due to poisoning by carbamate.

Discussion

The intestinal muscle hypertrophy observed in this case was classified as primary because it was not associated with any predisposing lesion, such as cancer, stenosis or cranial impaction to the area of muscle hypertrophy (2,12). Furthermore, no changes in the animal's feces were observed, and there were no clinical signs such as weight loss and tenesmus. In this case, the intestinal muscle hypertrophy was an accidental necropsy finding because death was due to poisoning by carbamate. A absence of clinical signs can be justified by the location of the injury that affected only the colon. In contrast, muscular hypertrophy in the small intestine can lead to death due to weight loss, anorexia, vomiting and diarrhoea, and in horses, cramps and intestinal rupture have also been observed (2,12).

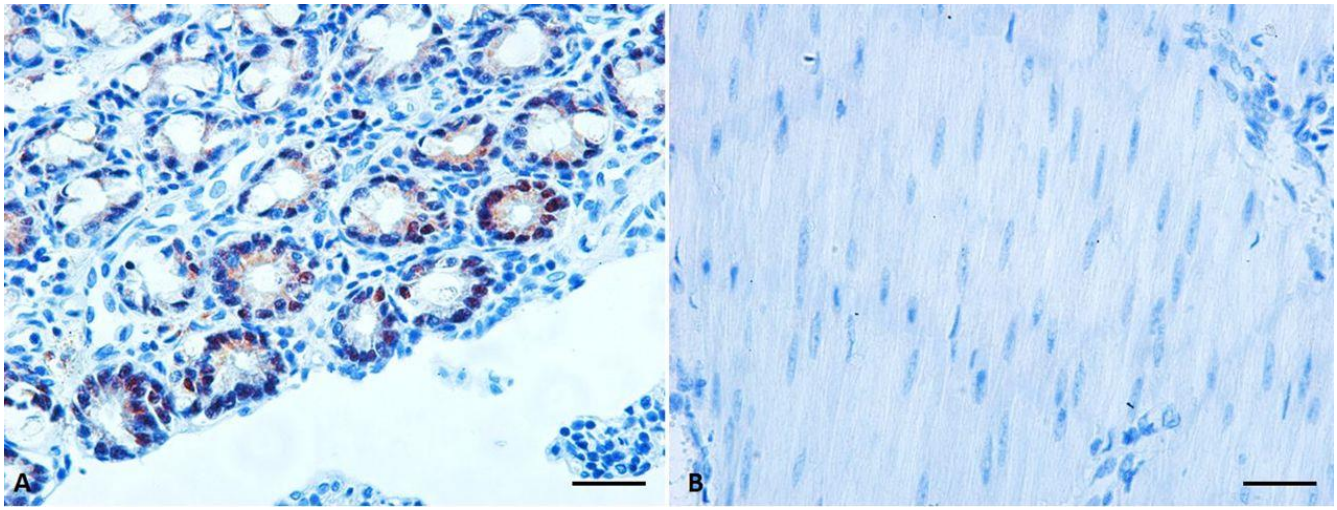

Figure 3. Idiopathic muscle hypertrophy of the colon in a cat. A and B) CDC47 immunohistochemistry expression in the colon. Mucosa (A) with strong CDC47 expression in crypt cells, unlike that of the inner and outer muscle layers (B) with no CDC47 expression. (Streptavidin-biotin-peroxidase method, Harris' hematoxylin counterstain, bar = 56 µm).

The genesis of the primary or idiopathic intestinal muscle hypertrophy has been attributed to enteritis, increased peristalsis of independent origin, abnormal consistency of the digesta, hormonal influence and the action of chemical mediators produced by parasitism (2). In this report, the only change that was associated with hypertrophy was lymphoplasmacytic colitis, which has also been observed in cases of idiopathic muscular hypertrophy of the small intestine (2,4,5,7,8,12,14). It is postulated that chronic enteritis can harden the intestinal segment, forcing it to function more intensely, which would result in smooth muscle hypertrophy. Another explanation is that the inflammatory process can release mediators with mitogenic action on smooth muscle cells (8,15). Among these mediators are the platelet-derived growth factor, insulin-like growth factor and tumour necrosis factor (8,16). However, it is important to emphasize that, in this case, enteritis was also observed in the small intestine which was not affected by muscle hypertrophy.

In this case, the percentage of nuclei in the muscle layers of the colon and normal duodenum was similar and had no CDC47 expression in the smooth muscle cells of the small and large intestine, confirming that these cells were not proliferating. Furthermore, the thickness of the colon was 3.5X greater than the normal duodenum, besides presenting a higher percentage of muscular in relation to other intestinal layers. These findings reinforce the diagnosis of hypertrophy instead of hyperplasia. It has been shown that the increase in cell volume observed in the intestinal muscle hypertrophy may be due to increased smooth and rough endoplasmic reticulum, non-contractile intermediate filaments and actin (17-22). However, hyperplasia of smooth muscle cells may be associated with intestinal muscle hypertrophy (8,17,23). Bettini et al. (12) observed rare MIB-1 expression, another marker of cell proliferation, in the muscular layer of the small intestine of cats with idiopathic muscular hypertrophy, stating that hyperplasia was also present in those cases. However, it is important to note that, although microscopic features of hyperplasia have not been observed in the present study, the possibility that hyperplasia preceded hypertrophy cannot be excluded because experimental studies have shown that the process can begin with hyperplasia and be overlapped by muscle hypertrophy (23).

Conclusion

Based on macroscopic, microscopic, and immunohistochemical findings, the diagnosis of primary muscle hypertrophy of the colon was confirmed. This lesion must be considered in the differential diagnosis of gastrointestinal tract diseases that result in tenesmus, weight loss and/or diarrhea in cats, such as neoplastic processes in the large intestine, chronic colitis and stenosis of the rectum or anus, which could predispose to secondary muscle hypertrophy of the large intestine. The prognosis will depend of the length of the affected segment and it is poor if the small intestine is also affected.

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References

1. NIELSEN SW. Muscular hypertrophy of the ileum in relation to terminal ileitis in pigs; a preliminary report. **J. Am. Vet. Med. Assoc.**, 1955, 127, 437-441.
2. CHAFFIN MK., FUENTEABLA IC., SCHUMACHER J., WELCH RD., EDWARDS JF. Idiopathic muscular hypertrophy of the equine small intestine: 11 cases (1980-1991). **Equine Vet. J.**, 1992, 24, 372-378.
3. PRANTNER MM. Intestinal smooth muscle hyperplasia in a rhea (*Rhea americana*). **Avian Dis.**, 1995, 39, 197-200.
4. DE LARA FCM., HERVAS J., BAUTISTA MJ., PEREZ J., GÓMEZ-VILLAMANDOS JC., MARTÍN DE LAS MULAS J., CARRASCO L. Intestinal smooth muscle hyperplasia in a goat. **J. Vet. Diagn. Invest**., 1996, 8, 390-392.
5. KING JM. Duodenal ulceration with stenosis and smoothmuscle hypertrophy in a foal. **Vet. Med.**, 1996, 91, 520.
6. GEUNA S., CARDILLO S., GIACOBINI-ROBECCHI MG. Smooth muscle cell hypertrophy and hyperplasia in the partially obstructed gut of the rat: a quantitative evaluation. **Acta Anat.**, 1998, 163, 69-74.
7. SANTOS RL, COELHO MIM. Hipertrofia muscular intestinal idiopatica em cão: relato de caso. **Arq. Bras. Med. Vet. Zootec**., 1998, 50, 525-529.
8. BLENNERHASSETT MG., VIGNJEVIC P., VERMILLION DL., COLLINS SM. Inflammation causes hyperplasia and hypertrophy in smooth muscle of rat small intestine. **Am. J. Physiol.**, 1992, 262, G1041-G1046.
9. OWEN DA., KELLY JK. Large intestine and anus. DAMJANOW I., LINDER J. Eds. **Anderson's Pathology**, 10th ed., St. Louis: Mosby, 1996: 1741-1778.
10. SLACK WW. Bowel muscle in diverticular disease. **Gut**, 1966, 7, 668-670.
11. GABELLA G. Hypertrophy of intestinal smooth muscle. **Cell Tissue Res.**, 1975, 163, 199-214.
12. BETTINI G., MURACCHINI M., DELLA SALDA L., PREZIOSI R., MORINI M. GUGLIELMINI C., SANGUINETTI V., MARCATO PS. Hypertrophy of intestinal smooth muscle in cats. **Res. Vet. Sci.**, 2003, 75, 43-53.
13. DIANA A., PIETRA M., GUGLIELMINI C., BOARI A., BETTINI G. et al Ultrasonographic and pathologic features of intestinal smooth muscle hypertrophy in four cats. **Vet. Radiol. Ultrasound**, 2003, 44, 566-569.
14. LINDSAY WA., CONFER AW., OCHOA R. Ileal smooth muscle hypertrophy and rupture in a horse. **Equine Vet. J.**, 1981, 13, 66–67.
15. BLENNERHASSETT MG., BOVELL FM., LOURENSSEN S., MCHUGH KM. Characteristics of inflammation-induced hypertrophy of rat intestinal smooth muscle cell. **Dig. Dis. Sci.**, 1999, 44, 1265-1272.
16. OWENS MW., GRISHAM MB. Cytokines increase proliferation of human intestinal smooth muscle cells: possible role in inflammation- induced stricture formation. **Inflammation**, 1993, 17, 481-487.
17. GABELLA G. Hypertrophic smooth muscle. I. Size and shape of cells, occurrence of mitoses. **Cell Tissue Res.**, 1979, 201, 63-78.
18. GABELLA G. Hypertrophic smooth muscle. II. Sarcoplasmic reticulum, caveolae and mitochondria. **Cell Tissue Res.**, 1979, 201, 79-92.
19. GABELLA G. Hypertrophic smooth muscle. III. Increase in number and size of gap junctions. **Cell Tissue Res.**, 1979, 201, 263-276.
20. GABELLA G. Hypertrophic smooth muscle. IV. Myofilaments, intermediate filaments and some mechanical properties. **Cell Tissue Res.**, 1979, 201, 263-276.
21. SCOTT RB., SHEEHAN A., CHIN BC., TAN DT. Hyperplasia of the muscularis propria in response to massive intestinal resection in rat. **J. Pediatr. Gastr. Nutr.**, 1995, 21, 399-409.
22. SIEGMAN MJ., BUTLER TM., MOOERS SU., TRINKLE-MULCAHY L., NARAYAN S. et al. Hypertrophy of colonic smooth muscle: structural remodeling, chemical composition, and force output. **Am. J. Physiol.**, 1997, 272, G1560- G1570.
23. SRINATHAN SK., LANGER JC., BLENNERHASSETT MG., HARRISON MR., PELLETIER GJ. et al. Etiology of intestinal damage in gastroschisis. III: morphometric analysis of the smooth muscle and submucosa. **J. Pediatr. Surg.**, 1995, 30, 379-383.

