CASE REPORT

Contribution to Histology of Paneth Cell Metaplasia from a Feline (*Felis catus domesticus)* With Colon Adenomatous

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With 6 figures **Received August, accepted for publication September 2020**

Abstract

The colon embryology in vertebrates recognizes the endoderm layer of the middle and caudal intestine as source of epithelium, and the surrounding lateral splanchnic mesoderm as a source of lamina propria, submucosa, muscular propria and adventitial or serosa tunic. The enterocytes, enteroendocrine cells and goblet cells of distal colon are generated by cranio-caudal expression of homeotic genes Hoxa13 and Hoxd13 both in the mesoderm and endoderm layer reported, and also spatially and temporally, everything under regulation of homeotic genes Cdx2. Concerning the Paneth Cell or PC, the endoderm layer is a source by sponsoring mesodermal layer related with lower expressions to Cdx2, leading to high expressions Hoxa13 and Hoxd13; and PC absence by high Cdx-2 expressions lead to lack of co-expression of same Hoxa genes. This case report contributes to the histology of PC from a domestic feline with a diagnosis of colon adenomatous, a species that does not normally express PC in the colon, reasonably proposing that PC metaplasia in feline adenomatous may imply expressive disabilities of Cdx-2 and / or Hoxa13 and Hoxd13 expressions

Keyword: Paneth cells, metaplasia, adenoma colon

Introduction

The colon embryology in vertebrates recognizes endoderm layer

of the middle and caudal intestine as source of epithelium, and the surrounding lateral splanchnic mesoderm as source of lamina propria, submucosa, muscular propria and adventitial or serosa tunic (Kiefer, 2003) (Thomason et al, 2012); In this developmental process, the enterocytes, enteroendocrine cells and goblet cells of distal colon are generated by cranio-caudal expression of homeotic genes Hoxa13 and Hoxd13 both in the mesoderm and endoderm layer reported, and also spatially and temporally (De Santa Barbara et al. 2003), everything under regulation of homeotic genes Cdx2 (Grainger et al. 2010) (Guiu & Jensen, 2015) (Gao, et al. 2009).

About Paneth Cell or PC in the colon when they are normally present, developmental biology explains this by the sponsorship of low Cdx2 expression, which leads to high expression of Hoxa13 and Hoxd13 and which in turn allows the expression of stem cells during the formation of colic glands with Lgr5 receptors or 5 receptors coupled to G proteins and with repeats rich in leucine (Bialecka 2011) (Múnera et al., 2017) so that they are bound by mesodermal Wingles Int or Wnt molecules of the Wnt2b type (Van et al, 2019 and Spit et al, 2018). On the other hand, when there is a normal absence of CP in the distal colon, developmental biology explains it by the lack of co-expression of homeotic genes Hoxa13 and Hoxd13 (De Santa Barbara et al. 2003) by a high expression of Cdx- 2 (Grainger et al. (2010), Guiu and Jensen, (2015), Gao et al., (2009), Crissey et al, (2011), Noah et al. (2011), and Silberg et al. (2000).

In retrospect, the first to notice Paneth cells (PC) absence in the colon was Joseph Paneth, (Paneth, 1887) and later Hally (1958) and Sandow & Whitehead (1979). They described Paneth cells (PC) with electron microscopy (EM), only in the small intestine of mice, as having dense granules of 0.75 to 1.5 microns, with peri-granular halo in vacuoles of 1 to 2 microns in diameter.

Histo-functional studies confirmed Paneth cells (PC) in the small intestine of the same species, Troughton & Trier (1969), coinciding with that reported with ME only in the human small intestine (Trier, 1963), Lagomorphs (Pit'ha, 1968) and

Echidnas (Krause, 1971). All descriptions with pancreatic acinar granular morphological similarity of lysozymal proteolytic content or LIS (Riecken & Pearse, 1966).

After, Peeters and Vantrappen (1975) using LIS as an immunomarker, showed again that PC exists only in the small intestine of mice; and already in the second century, with the polymerase chain reaction with reverse transcriptase or TR-PCR (Ayabe et al. 2001) together with other histo-functional studies with Western Blot, they again identified LIS in PC located only in crypts of Lieberkühn from the small intestine of mice but not from the colon (Salzman et al. 2003).

Recently, by means of Northern Blot, LIS mRNA was identified for the first time in PCs from Lieberkühn's crypts of the normal human small intestine, but not in the colon (Wehkamp et al. 2006). Thus, PC only in the Lieberkühn crypts of the small intestine are accepted as normal and LIS as its immunomarker (Elphick and Mahida, 2005)

Today, PCs are taught circumscribed to the epithelium of the thin intestinal mucosa at the base of the Lieberkühn crypts, as evidenced by contemporary university histological texts (Ross and Pawilina, 2007; Stevens and Lowe, 2006; Kierszenbaum, 2012; Eroschenko Fiore, 2013; and Mescher, 2013) and normally absent in murine distal colon (Piper et al, 2017, pg223) canine and feline (Gelberg, 2014) (Washabau & Day, 2013, pg54), (Ergün et al 2003), (Sandow and Whitehead 1979) (Creamer, 1967).

Already in a pathological context to the human distal colon, the expression of CP can be demonstrated histologically, when findings of CP colic metaplasia are configured (Lewin, 1969 and Gassler et al, 2017); for example, cases such as human chronic colitis (Tanaka et al, 2001), human ulcerative colitis (Bedini, et al, 2014 and Tanaka et al, 2001) and human Crhon disease (Stappenbeck & McGovern, 2017 and Tanaka et al., 2001) and even in frank human colic adenomas and adenocarcinomas (Mahon, et al., 2016).

The objective of this case report is to contribute to the histology of a CP meaplasia from a feline colon adenomatous, a species that does not normally express CP in the colon.

Case Description and Pathological Findings

A 5-year-old male creole feline (*Felis catus domesticus*), with a chronic history of diarrhea refractory to symptomatic treatment, consults at a Private Veterinary Clinic in the city of Bogotá- Colombia-South America, where after a 36-hour fast, a colonoscopy, observing a moderately distensible colon, whose mucosa in the first transverse and distal descending third is seen in a diffuse and generalized way with slight non-pedunculated granulomatous macrophoci, covered with a large amount of mucus, (Fig 1A, and Fig 1B.)

Five biopsies of the granulomatous masses are performed, fixing in 10% Formol for conventional histological processing Histokinette © in paraffin and 3-micron sections made with American Optical © microtome and H&E histochemistry at the commercial laboratory Base Médica SAS -Bogotá- Colombia.

The slides obtained were examined under the Olympus BX43 optical microscope showed a 4X magnification colic mucosa with hyperplastic foci at the expense of the colic glands (Fig 2); At 10X magnification, the colic glandular luminal surface is essentially covered by a large amount of goblet cells, in turn at 40X magnification diffuse distribution alternated with cells of eosinophilic granulated cytoplasm and nucleus when visible basal spindle-shaped heterochromatic, compatible with CP (Fig 3 and Fig 4). The location of the described PCs was found at the base, body and isthmus of the hyperplastic colic glands.

Regarding the underlying lamina propria, a slight increase in lymphoplasmacytic infiltrate was diffusely observed. To confirm CP immunophenotyping, immunohistochemical staining was performed based on the streptavidin-biotin system (LSAB). New sections of 3-micron thick and previous deparaffinization with xylol and gradual rehydration with PBS, were incubated for 24 hours with antibodies monoclonal Anti-lysozyme ABCAM Rabbit monoclonal EPR2994 dilution 1: 100. Pancreas sections were used as control; where the characteristic intense cytoplasmic brown color of confirmatory PC (Fig. 5).

The tissue morphological finding configured the concept of PC metaplasia in the distal colon of the male adult cat with colic adenomatous.

Discussion

Felines are within the group of mammals among which there is no normal expression of PC in microfolds of the colic mucosa (Washabau & Day, 2013, pg54) (Creamer, 1967) (Sandow & Whitehead 1979), (Ergün et al 2003) , (Gelberg, 2014). With H&E histochemistry PC has been reported formally in the normal duodenum (Da Silva et al. 2010), and with Masson's Trichrome histochemistry in the normal duodenum of young felines (Al-Saffar, F. J. & Al-Zuhairy, 2016).
This case report describes, for the first time, the presence of PC in the feline distal colon in a case diagnosed as colic adenomatous.

Considering the developmental biology, it is proposed that the metaplasia of CP in the feline adenomatous derived from colitis, may imply expressive disabilities of the Cdx-2 genes favoring Hoxa13 and Hoxd13 expressions as reported in the literature with a focus on the metaplastic development of PC in this case of colitis. Interestingly “in vivo”, intestinal metaplasia of PC has been confirmed in murine models (Hrynuik et al, 2012), humans (Liu et al, 2007) and even and beyond, in human colon adenomas and adenocarcinomas (Hinoi, et al 2002) and recently also demonstrated “in vitro” (Danielsen et al, 2018) by Hoxa13 and Hoxd13 expressions in neoplastic colon (Alfredo, 2014) of both colic adenomas and adenocarcinomas (Tatangelo et al., 2018).

Conflict of interests

The authors do not declare any conflict of interest

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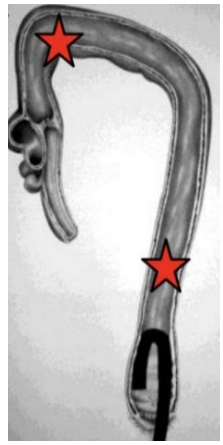

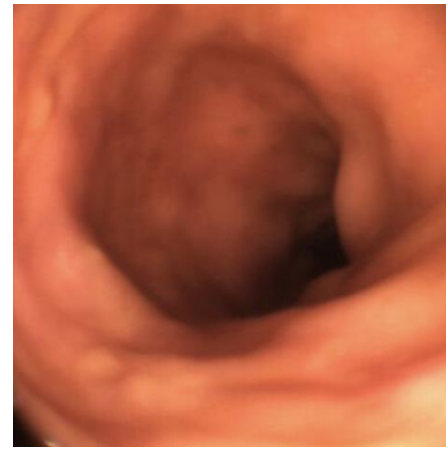

1A **1B**

Fig (1A): Anatomical diagram of the feline colon, indicating the beginning of the transverse and distal descending colon where the biopsies were taken for histological analysis.

Fig (1B): Mild granulomatous morphological appearance of the transverse and distal descending colic mucosa.

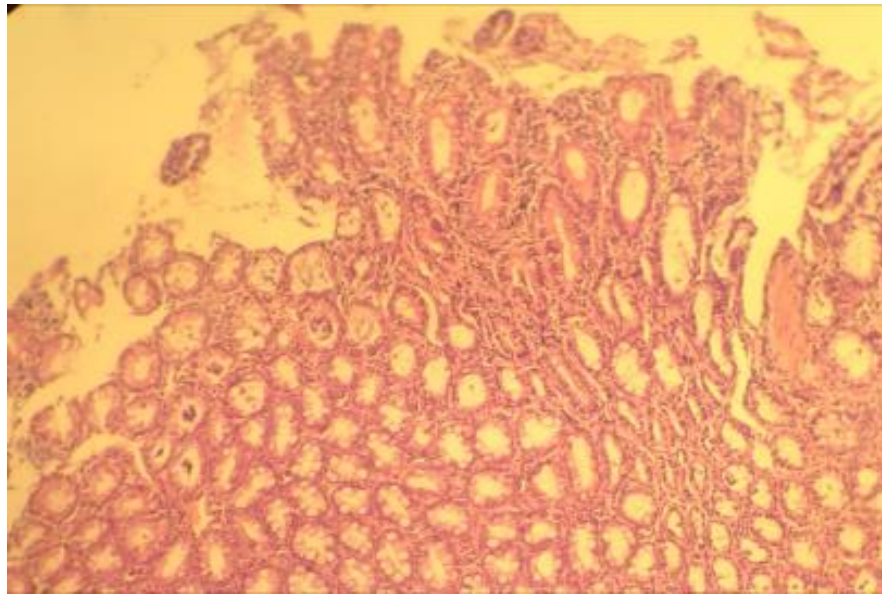

Fig (2): Distal colic mucosa with hyperplastic glands in distal colon of feline with an adenoma. (H&E. 4X.)

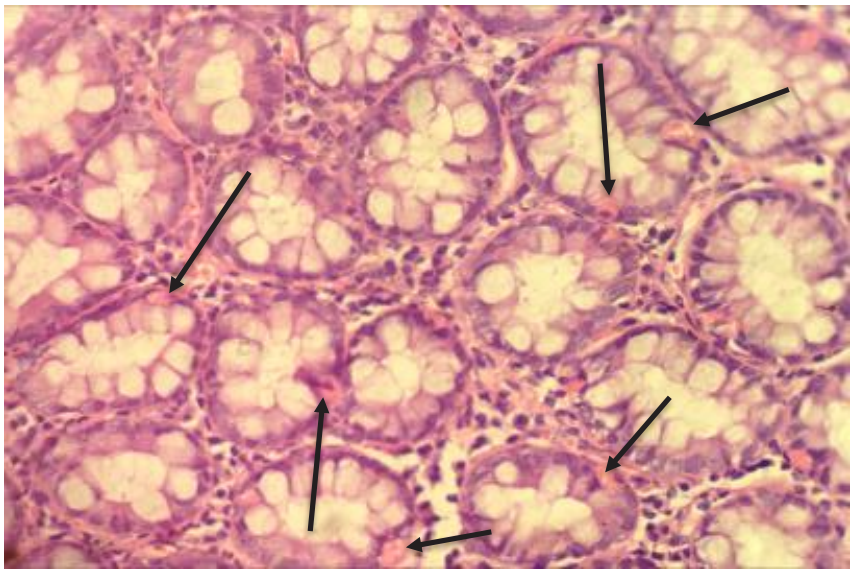

Fig (3): Cross sections of hyperplastic colic glands in feline distal with adenomatous colon and arrows showing Paneth Cells. (H&E. 10X)

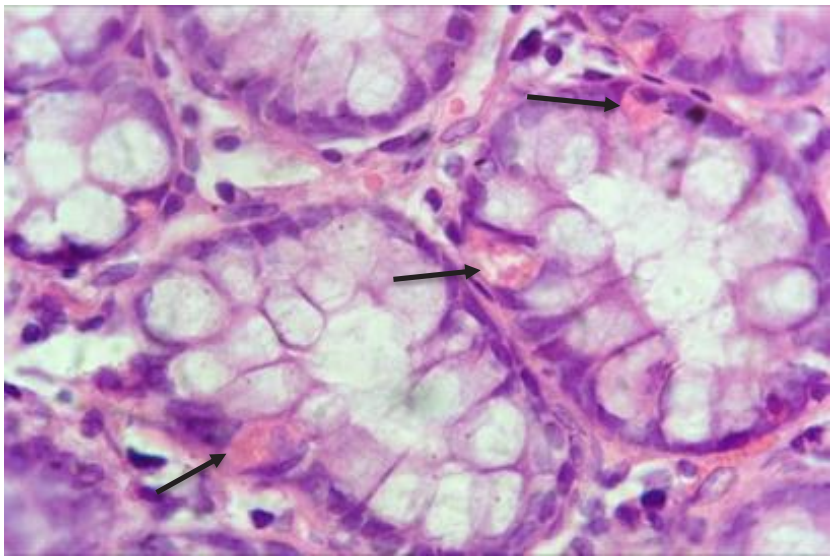

Fig (4): Colic glands surface close-up with arrows showing occasional Paneth cells in feline distal adenomatous colon. (H&E. 40X)

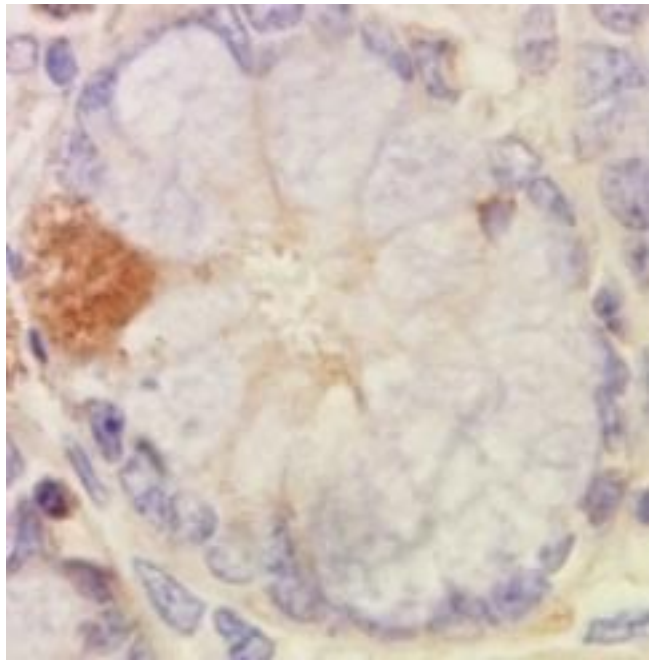

Fig (5): H&E at 40X with additional 2X digital zoom with feline distal colic gland with where a Paneth cell positively labeled with monoclonal and polyclonal antibodies is observed in the middle of goblet cells Anti-lysozyme - Anti LIS- (ABCAM Rabbit monoclonal EPR2994 dilution 1: 100).