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Histological Aspects of Small Intestine of the Capybara *Hidrochoerus hydrochaeris* Linnaeus, 1766 (Mammalia, Rodentia, Hydrochaeridae)

Aspectos Histológicos do Intestino Delgado da Capivara *Hydrochoerus hydrochaeris* Linnaeus, 1766 (Mammalia, Rodentia, Hydrochaeridae)

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Abstract

The histological and morphometric study of the small intestine of adult capybaras *Hydrochoerushydrochaeris*was developed in this work, emphasizing particularities of the duodenum, jejunum, and ileum. Fragments of the cranial, medial, and caudal portions of each intestinal segment were collected, submitted to the routine histological processing, and stained by the techniques of Hematoxylin-Eosin, Alcian Blue, and Periodic Acid Schiff. The small intestine of the capybara, regarding its histological structure, is similar to most mammals, composedofthe mucosal, submucosal, muscular, and serosal layers. There was no significant difference in the thickness of those layers among the intestinal segments. Thick and ramified folds were found along the small intestine, being more developed in the duodenum. Finger-shaped villus and with other forms, besides ramified villus, and a thick brush border were observed. Brünner's glands were seen in the cranial portion of the duodenum, distributed in the submucosal and the basal area of the mucosal layer, as well as numerous goblet cells along the small intestine, both presenting acid, and neutral glycoconjugates. Several defense cells were found in the connective tissue of the mucosal and submucosal layers, mainly lymphocytes, diffuse or forming lymphoid nodules, which aggregate to form Payer´s patches in the caudal portion of the ileum. Paneth´s cells and enteroendocrine cells were also detected in the intestinal epithelium.

Keywords: Digestive Tract. Histometry. Morphology. Rodents.

Resumo

O estudo histológico e morfométrico do intestino delgado de capivaras adultas Hydrochoerus hydrochaeris foi desenvolvido neste trabalho, enfatizando particularidades do duodeno, jejuno e íleo. Foram coletados fragmentos das porções cranial, medial e caudal de cada segmento intestinal, submetidos ao processamento histológico de rotina e corados pelas técnicas de Hematoxilina-Eosina e Alcian Blue-Ácido Periódico de Schiff. O intestino delgado da capivara é semelhante ao da maioria dos mamíferos em relação à sua estrutura histológica, sendo constituído pelas camadas mucosa, submucosa, muscular e serosa. Não houve diferença significativa na espessura dessas camadas entre os segmentos intestinais. Pregas espessas e ramificadas foram encontradas ao longo do intestino delgado, sendo mais desenvolvidas no duodeno. Observaram-se vilosidades digitiformes e com outras formas, além de vilosidades ramificadas, e uma espessa borda em escova. Glândulas de Brünner foram observadas na porção cranial do duodeno, distribuídas na camada submucosa e na área basal da camada mucosa, além de numerosas células caliciformes ao longo do intestino delgado, ambas apresentando glicoconjugados ácidos e neutros. Várias células de defesa foram encontradas no tecido conjuntivo das camadas mucosa e submucosa, principalmente linfócitos difusos ou constituindo nódulos linfóides que se associam para formar as placas de Peyer na porção caudal do íleo. Células de Paneth e células enteroendócrinas também foram detectadas no epitélio intestinal.

Palavras-chave: *Roedores. Morfologia. Histometria. Trato Digestivo.*

1 Introduction

The small intestine is the organ chiefly responsible for food digestion and nutrient absorption, and inmammals, it consists of the duodenum, jejunum, and ileum segments. Along withthese segments, the small intestine has adaptations that increase its surface, at a macroscopic level by its length and internal folds, and at a microscopic level by villi and microvilli (BARRET, 2006).

Although there are many common features to the duodenum, jejunum, and ileum segments, they may present some particular aspects, such as the predominance of Brünner's glands in the duodenum and Peyer's patches in the ileum (BANKS, 1992; GEORGE; ALVES; CASTRO, 1998). The morphological differences are probably associated with the physiology of each intestinal region: the duodenum receives the acid chyme and the pancreatic and biliary juices, being an important pH neutralization site (CUNNINGHAM, 1993; SWENSON; REECE, 1996).

Previous studies on the small intestine of mammals allow the development of applied research in the nutrition and gastrointestinal pathology field, and, about the wild mammals, comparative morphology and ecomorphologyare relevant.

Given the zootechnical relevance of capybara in meat and leather production (NOGUEIRA-FILHO; NOGUEIRA,

2013, 2018), and since it is a reservoir of parasites with zoonotic potential (CATROXO *et al*., 2010, 2014; CUETO, 2013; SOUZA *et al*., 2015; VALADAS *et al.*, 2010), there are relatively several studies on capybara digestive system (BRESSAN *et al.,* 2004, 2005; CARRASCAL; ORTIZ; PETRO, 2016; FREITAS *et al.,* 2008; GONZÁLEZ-JIMÉNEZ, 1995; KIANI *et al*., 2019; MORAES *et al*., 2002; RODRIGUES *et al.,* 2006; SARTORI *et al*., 2018; VÁZQUEZ; SENOS; PÉREZ, 2012; VELÁSQUEZ *et al.,* 2002, 2003), although none of them involves small intestinehistometry.

Thus, this work aimed to describe histological and histometric aspects of the small intestine of capybara *Hydrochoerushydrochaeris* and to verify possible differences between duodenum, jejunum, and ileum, regarding the qualitative and quantitative constitution of its wall.

2 Material and Methods

Nine adult capybara, six males, and three females, with an average weight of 38,0±8,0 kg, and an average length of 1,15±0,11m ("snout-rump length", measured from the end of the snout to the end of the tail, along the back of the animal) came from and were euthanized at the Cachoeirinha Farm of the Federal University of Viçosa, in Viçosa – MG, under authorization from Brazilian Institute of Environment and Renewable Natural Resources (IBAMA) (Authorization Number: 060/04-NUFAS/MG), and following the ethical principles for the use of animals in experimentation procedures(Brazilian College of Animal Experimentation – COBEA, 1991).After the euthanasia, the animals were weighed, measured, and sexed, and the abdominal cavity was exposed to remove the small intestine, whose segments (duodenum, jejunum and ileum) were identified and delimited "*in situ*".

A 2cm^2 fragment was collected from the cranial, middle, and caudal portions of the duodenum, jejunum, and ileum. The fragments were fixed in 10% aqueous buffered formalin solution for 24 hours at room temperature. After fixation, the fragments were dehydrated in a serial increasing concentration of ethyl alcohol, cleared in xylene, embedded in histological paraffin (Bancroft Stevens, 1996), and sectioned (5µm thick) in a manual rotating microtome (Spencer, American Optical). The obtained slices were stained with Hematoxylin-Eosin (HE) (BANCROFT; STEVENS, 1996) for histological and histometric analysis, and with the conjugate technique of Alcian Blue pH2.5 (AB) and Periodic Acid Schiff (PAS) (MC MANUS; MOWRY, 1960), to identify cells containing acidic and neutral glycoconjugates, respectively. In the AB-PAS technique, counterstaining with hematoxylin was performed to visualize the cell nuclei.

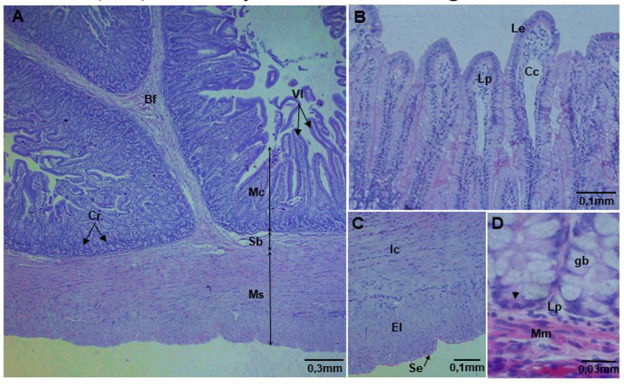
The subsequent analyzes were carried out with a microscope (Leitz Wetzlar), and, by using a micrometric eyepiece (Leitz Wetzlar), the height of the wall and the intestinal mucosa, the submucosal and muscular layers, the muscular sublayers (internal circular and external longitudinal), and of the villi and the crypts, were measured in each portion of each intestinal segment. These measurements were made in nine random fields of the histological sections, obtaining an average number for each parameter analyzed in the different intestinal portions (n = 81).

The illustrative photos shown in this work were taken using a camera (Fujix HC-300Zi) coupled to a microscope (Nikon Eclipse E600) and a computer to capture the images.

3 Results and Discussion

The small intestine wall of *H. hydrochaeris* capybara is formed by four well-defined layers: mucosa, submucosa, muscular, and serosa (Figure 1). About the thickness of the intestinal wall, there was no significant difference between the cranial, middle, and caudal portions of each segment, not even between the segments, duodenum, jejunum, and ileum (Table 1).

Figure 1 - Histological sections of the small intestine of *H. hydrochaeris* capybara. A-Constitution of the intestinal wall, showing a large branched fold (Bf) and the parietal layers - mucosa (Mu) with villi (Vl) and crypts (C), submucosa (Sb), and muscular (Ms). B- Intestinal villi covered by the epithelium (Le) and filled by the lamina propria (Lp) with a central cheliferous vessel (Cc). C- Muscle layers, inner circular (Ic) and outer longitudinal (El), and serous layer (Se). D- Intestinal crypts surrounded by the lamina propria (Lp), showing goblet cells (gb) and Paneth cells (arrowhead), in addition to the muscularis mucosae (Mm).Hematoxylin and eosin staining



Source: The authors.

Table 1 - Wall thickness in different portions and segments of the small intestine of the capybara *H. hydrochaeris* (data expressed as mean ± standard derivation, in mm)

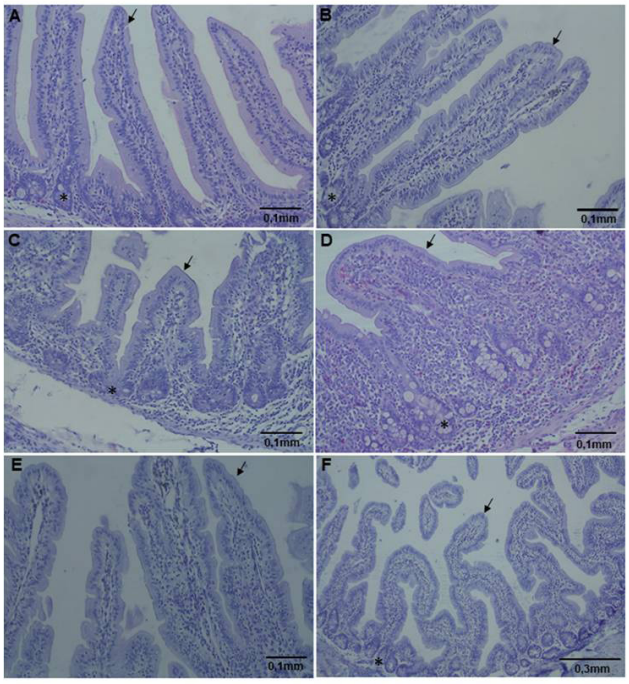
Portion	Wall thickness	Segment	Wall thickness
Cranial duodenum	2,08 ±0,24 ^{A}		
Middle duodenum	1,98 ±0,23 ^{A}	Duodenum	1,96 ±0,22 ^{A}
Caudal duodenum	1,82 ±0,17 ^{A}		
Cranial jejunum	1,90 ±0,19 ^{A}		
Middle jejunum	1,89 ±0,19 ^{A}	Jejunum	1,93 ±0,20 ^{A}
Caudal jejunum	2,00 ±0,21 ^{A}		
Cranial ileum	1,88 ±0,18 ^{A}		
Middle ileum	1,86 ±0,16 ^{A}	Ileum	1,93 ±0,18 ^{A}
Caudal ileum	2,06 ±0,22 ^{A}		

Means followed by equal letters, between intestinal portions and segments, do not differ from each other by the Tukey test at a 5% significance level.
Source: Research data.

In different parts of the small intestine, there are prominent folds, parallel to each other, in a spiral arrangement along the digestive tract, with smaller folds between them, arranged in a network pattern. The prominent folds can be seen by the naked eye, being branched, especially those in the duodenum (Figure1A).

The mucosa layer is made up of villi (Figures 1B and 2A), crypts or intestinal glands (Figures 1B, 2B), and a thin muscular layer: the muscularis mucosae (Figures 1B and 2B). The villi are characterized by being mucosal projections, lined with epithelium, and filled with the connective tissue lamina propria. On the other hand, the crypts are tubular invaginations lined with epithelium and surrounded by the lamina propria (Figures 2A and 2B). Regarding the thickness of the mucosa layer, there was no significant difference between the portions of each intestinal segment, not even between the segments (Table 2).

Figure 2 - Histological sections of the small intestine of *H. hydrochaeris* capybara, showing villi with different shapes (arrows). A-Finger-shaped villi. B-Branched finger-shaped villi. C-Pyramidal villus. D-Bell-shaped villi. E-Tongue-shaped villi. F-Leaf-shaped, sinuous, and branched villi. Asterisks - crypts. Hematoxylin and eosin staining



Source: Research data.

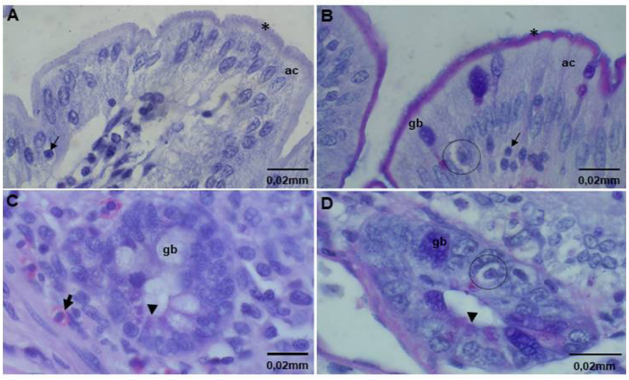
Table 2 - Mucosa layer thickness in portions and segments of the small intestine of the capybara *H. hydrochaeris* (data expressed as mean ±standard derivation, in mm)

Portions	Mucous Layer Thickness	Segment	Mucous layer thickness
Cranial duodenum	0,74±0,14 ^{A}		
Middle duodenum	0,67±0,20 ^{A}	Duodenum	0,67±0,15 ^{A}
Caudal duodenum	0,61±0,11 ^{A}		
Cranial jejunum	0,60 ±0,09 ^{A}		
Middle jejunum	0,56 ±0,10 ^{A}	Jejunum	0,55 ±0,10 ^{A}
Caudal jejunum	0,49 ±0,11 ^{A}		
Cranial ileum	0,54 ±0,11 ^{A}		
Middle ileum	0,51 ±0,10 ^{A}	Ileum	0,54 ±0,12 ^{A}
Caudal ielum caudal	0,57 ±0,14 ^{A}		

Means followed by equal letters, between intestinal portions andsegments, do not differ from each other by the Tukey test at a 5% significance level.
Source: Research data.

Most villi are finger-shaped being simple or branched (Figures 3A and 3B). However, villi with other shapesare also seen, particularly in the jejunum and ileum: pyramidal (Figure 3C), tongue-shaped (Figure3D), or leaf-shaped (Figures 3E and 3F) villi. Many of the villi show recesses on their luminal face (Figures 3A - 3E), and some of them, especially the leaf-shaped ones, are quite sinuous and are not fully registered in the histological sections (Figures 3F). There was no significant variation in the height of the villi in the intestinal portions or segments (Table 3). On the other hand, the crypts are significantly deeper in the duodenum compared to the other segments (Table 4).

Figure 3 - Histological sections of the small intestine of *H. hydrochaeris* capybara, evidencing the lining epithelium (A, B) and crypts (C, D). ac – absorptive cells; arrowheads – Paneth cells; asterisks – brush border; circles – enteroendocrine cell; gb – goblet cells; thin arrows –intraepithelial lymphocyte; thick arrow – mast cell. Hematoxylin and eosin staining (A and C); Periodic Acid Schiff and Alcian Blue staining (B and D)



Source: Research data.

Table 3 - Villi height in different portions and segments of the small intestine of the capybara *H. hydrochaeris*(data expressed as mean ± standard derivation, in mm)

Portions	Villi height	Segment	Villi height
Cranial duodenum	0,49 ±0,06 ^{A}		
Middle duodenum	0,45 ±0,06 ^{A}	Duodenum	0,46 ±0,05 ^{A}
Caudal duodenum	0,43 ±0,06 ^{A}		
Cranial jejunum	0,43 ±0,05 ^{A}		
Middle jejunum	0,38 ±0,07 ^{A}	Jejunum	0,40 ±0,05 ^{A}
Caudal jejunum	0,39 ±0,03 ^{A}		
Cranial ileum	0,39 ±0,07 ^{A}		
Middle ileum	0,39 ±0,06 ^{A}	Ileum	0,40 ±0,07 ^{A}
Caudal ileum	0,41 ±0,07 ^{A}		

Means followed by equal letters, between intestinal portions and segments, do not differ from each other by the Tukey test at a 5% significance level.
Source: Research data.

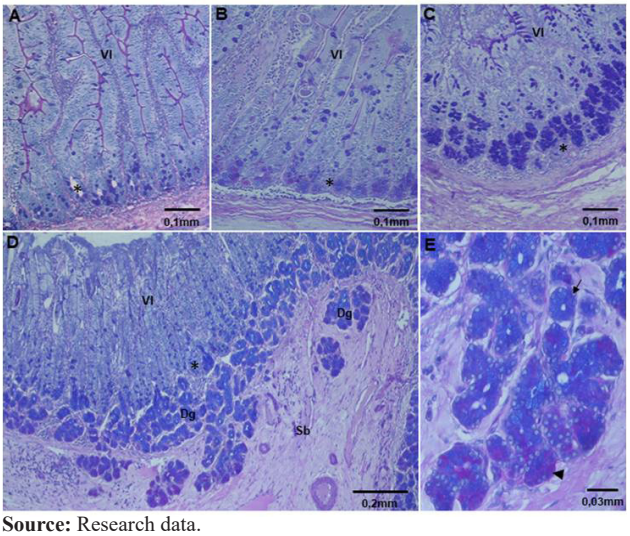
Table 4 - Crypts depth in portions and segments of the small intestine of the capybara *H. hydrochaeris*(data expressed as mean ± standard derivation, in mm)

Portions	Crypts depth	Segment	Crypts depth
Cranial duodenum	0,18 ±0,06 A		
Middle duodenum	0,16 ±0,05 A	Duodenum	0,16 ±0,05 A
Caudal duodenum	0,15 ±0,06 A		
Cranial jejunum	0,14 ±0,05 A		
Middle jejunum	0,11 ±0,07 A	Jejunum	0,11 ±0,05 B
Caudal jejunum	0,09 ±0,03 B		
Cranial ileum	0,09 ±0,02 A		
Middle ileum	0,10 ±0,02 A	Ileum	0,10 ± 0,02 B
Caudal ileum	0,11 ±0,02 A		

Means followed by equal letters, between intestinal portions and segments, do not differ from each other by the Tukey test at a 5% significance level.
Source: Research data.

The intestinal epithelial tissue consists ofa simple layer, composed of prismatic absorptive cells, mucus-producing goblet cells, Paneth cells, and endocrine cells (Figure 4). Absorbent cells are predominant in the epithelium lining the villi; goblet and endocrine cells are predominant in the crypt region; while Paneth's were found only in the basal portion of the crypts (Figure 2). The absorptive cells (Figure 4A and 4B) have a prismatic shape, oval nucleus and the apical region shows a well-developed brush border, strongly marked in the PAS technique.

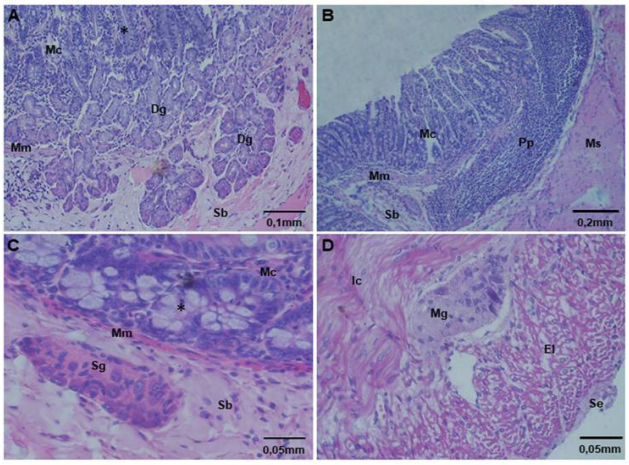
Figure 4 - Histological sections of the small intestine of *H. hydrochaeris* capybara, evidencing goblet cells in the duodenum (A), jejunum (B), and ileum (C), and the duodenal glands (D, E). arrow -positive AB cells; arrowhead -positive PAS cell. Asterisk -crypts; Dg - duodenal glands; Sb - submucosa; Vl - villi. Periodic Acid Schiff and Alcian Blue staining



Source: Research data.

The found goblet cells presented acid (AB-positive), neutral (PAS-positive), or both glycoconjugates (AB-PAS-positive), being the AB-PS positive cells in greater number (Fig. 4B and 4D). In thelongest axis section, the goblet cellspresent a typical chalice shape, a voluminous cytoplasm, and a nucleus displaced towards the basal portion. In crypts, the goblet cells tend to form clusters, often being difficult to differentiate the cell boundaries, while in villi they are in a dispersed arrangement. The amount of goblet cells seems to be greater in the ileum, followed by the jejunum and duodenum (Figure 5).

Figure 5 - Histological sections of the small intestine of *H. hydrochaeris* capybara, showing in A) the duodenal glands- Dg; in B) the Peyer's plaque - Pp; in C) a submucosal ganglion - Sg; and in D) a myenteric ganglion – Mg; asterisks - crypts; El - external longitudinal muscle; Ic - internal circular muscle; Mc - mucosa; Mm – muscularis mucosae; Ms - muscle; Sb - Submucosa. Hematoxylin and eosin staining



Source: Research data.

Paneth cells, presenting a pyramidal shape, are present in different segments of the small intestine. By the HE technique, the Paneth cells showlarge acidophilic granules in the upper portion of the cytoplasm (Figure 4C), and by the glycoconjugatestechnique, they present PAS-positive granules (Figure4D).

Endocrine cells were also observedthroughout the small intestine in a dispersed and sparse pattern. Despite being difficult to identify, the endocrine cells show a clear cytoplasm and a little basophilic nucleus (Figures 4Band 4D).

The lamina propria (Figures 2, 3, and 4), found just below the epithelium, consists of loose connective tissue with small blood vessels and a prominent lymph vessel located at the villi central region (Figures 2A). There are typical connective cells, fibroblasts and fibrocytes (Figure 4A), smooth muscle cells (Figure 2B), and defense cells (Figures 4A, 4B, and 4C): lymphocytes, macrophages, and mast cells. The mast cells are seen in a great number, especially in the jejunum and ileum, showing a highly acidophilic cytoplasm.

The muscularis mucosae (Figures 1B and 2B) is made up of two thin layers of smooth muscle fibers. The inner layer has a circular arrangement, while the outer layer is positioned in the longitudinal plane. In some parts of the organ, the muscularis mucosaehavea single layer, and in the cranial duodenum,a portionis invaded byBrünner's glands, resulting in a discontinuous appearance.The same occurs in the caudal ileum portion, due to Peyer's patches occurrence.

The submucosal layer (Figures 1A, and 5A-5C) is

composed ofdense connective tissue and medium-sized blood and lymphatic vessels. This layer may also present glands, lymphoid nodules, and nervous ganglia. Regarding the thickness of the submucosal layer, there was no significant variation between the intestinal portions and segments (Table 5).

Table 5 - Submucosal layer thickness in portions and segments of the small intestine of the capybara *H. hydrochaeris* (data expressed as mean ± standard derivation, in mm)

Portions	Submucosal Layer Thickness	Segment	Submucosal Layer Thickness
Cranial duodenum	0,21±0,06 ^{A}		
Middle duodenum	0,17 ±0,05 ^{A}	Duodenum	0,19 ±0,05 ^{A}
Caudal duodenum	0,18 ±0,06 ^{A}		
Cranial jejunum	0,17 ±0,05 ^{A}		
Middle jejunum	0,17 ±0,07 ^{A}	Jejunum	0,15 ±0,05 ^{B}
Caudal jejunum	0,12 ±0,03 ^{B}		
Cranial ileum	0,13 ±0,03 ^{A}		
Middle ileum	0,13 ±0,04 ^{A}	Ileum	0,14 ±0,04 ^{B}
Caudal ileum	0,16 ±0,04 ^{A}		

Means followed by equal letters, between intestinal portions and segments, do not differ from each other by the Tukey test at a 5% significance level.
Source: Research data.

The duodenal (or Brünner) glands (Figure5A) are present in the submucosa layer of the duodenum'scranial and middle portions, beingmore abundant in the first one. These glands do not occur exclusively in the submucosa: they penetrate the mucous layer, reaching the crypts. These glands are convoluted and tubule acinoustype, presenting mucous and serous acini, being the first the most abundant (Figure 5A). The acinarcells are (AB-PAS) positive type, the serous and mucous cell appearing to be more PAS-positive and more AB positive, respectively (Figure 5A).

Lymphocytes can be seen dispersed in the lamina propria, interspersed between epithelial cells (intraepithelial lymphocytes), or forming lymphoid nodules. The lymphoid nodules are more numerous and have a higher size when found in the ileum, in which the caudal portionsare associated to form Peyer's patches (Figure 5B).

Submucosal nerve ganglia (Figure 5C) are found throughout the small intestine, being larger in the folds internal region. These ganglia contain many neurons cell bodies, which are large and present a markedly basophilic cytoplasm and a clear nucleus with an evident nucleolus.

The muscular layer (Figures 1A, 1C, and 5D) is subdivided into an inner layer made of muscle bundles in a circular arrangement, and an outer layer having bundles in a longitudinal orientation. Between these layers, well-developed nerve ganglions (Figure 7)occurin greater numbers and are often larger thanthose found in the submucosa. The inner circular layer is often thicker than the outer longitudinal one. About the thickness of the muscle layer and its constituent parts, there was no significant variation between the intestinal portions and segments (Tables 6, 7, and 8).

Table 6 - Muscular layer thickness in portions and segments of the small intestine of the capybara *H. hydrochaeris* (data expressed as mean ± standard derivation, in mm)

Portions	Muscular Layer Thickness	Segment	Muscular Layer Thickness
Cranial duodenum	1,04 ±0,06 ^{A}		
Middle duodenum	1,03 ±0,05 ^{A}	Duodenum	1,02 ±0,05 ^{A}
Caudal duodenum	0,99 ±0,06 ^{A}		
Cranial jejunum	0,89 ±0,05 ^{A}		
Middle jejunum	1,02 ±0,07 ^{A}	Jejunum	0,93 ±0,05 ^{A}
Caudal jejunum	0,88 ±0,03 ^{A}		
Cranial ileum	0,98 ±0,07 ^{A}		
Middle ileum	0,96 ±0,09 ^{A}	Ileum	0,96 ±0,09 ^{A}
Caudal ileum	0,93 ±0,11 ^{A}		

Means followed by equal letters, between intestinal portions and segments, do not differ from each other by the Tukey test at a 5% significance level.
Source: Research data.

Table 7 - Thickness of the inner circular muscle sublayer in portions and segments of the small intestine of the capybara *H. Hydrochaeris* (data expressed as mean ± standard derivation, in mm)

Portions	Thickness of the Inner Circular Muscle	Segment	Thickness of the Inner Circular Muscle
Cranial duodenum	0,62 ±0,06 ^{A}		
Middle duodenum	0,71 ±0,05 ^{A}	Duodenum	0,67 ±0,05 ^{A}
Caudal duodenum	0,69 ±0,06 ^{A}		
Cranial jejunum	0,64 ±0,05^{A}		
Middle jejunum	0,79 ±0,07 ^{A}	Jejuno	0,70 ±0,05 ^{A}
Caudal jejunum	0,67 ±0,03 ^{A}		
Cranial ileum	0,67 ±0,07 ^{A}		
Middle ileum	0,65 ±0,08 ^{A}	Ileum	0,66 ±0,08 ^{A}
Caudal ileum	0,66 ±0,10 ^{A}		

Means followed by equal letters, between intestinal portions and segments, do not differ from each other by the Tukey test at a 5% significance level.
Source: Research data.

Table 8 - Thickness of the outer longitudinal muscle sublayer in portions and segments of the small intestine of the capybara *H. Hydrochaeris* (data expressed as mean ± standard derivation, in mm)

Portions	Thickness of the Outer Longitudinal Muscle	Segment	Thickness of the Outer Longitudinal Muscle
Cranial duodenum	0,37 ±0,06 ^{A}		
Middle duodenum	0,31 ±0,05 ^{A}	Duodenum	0,31 ±0,05 ^{A}
Caudal duodenum	0,25 ±0,06 ^{A}		
Cranial jejunum	0,20 ±0,05 ^{A}		
Middle jejunum	0,22 ±0,07 ^{A}	Jejunum	0,22 ±0,05 ^{A}
Caudal jejunum	0,23 ±0,03 ^{A}		
Cranial ileum	0,28 ±0,07 ^{A}		
Middle ileum	0,29 ±0,04 ^{A}	Ileum	0,30 ±0,05 ^{A}
Caudal ileum	0,32 ±0,04 ^{A}		

Means followed by equal letters, between intestinal portions and segments, do not differ from each other by the Tukey test at a 5% significance level.
Source: Research data.

The serosa layer (Figure 1A) consists of a thin connective membrane covered by simple squamous mesothelium, having some small blood vessels.

The small intestine wall of capybara *H. hydrochaeris* is similar to that of other mammals concerning its constitution: the mucosa, submucosa, muscular and serous layers are present. According to Velásquez *et al*. (2003), the thickness of the intestinal wall of *H. hydrochaeris* capybara is variable along its length, being greater in the duodenum. In the presentstudy, however, no statistically significant variation in wall thickness was observed along the small intestine of capybara, despite the apparent variation as observed by Velásquez *et al*. (2003).

This work findings include the presence of folds in the inner lining of the small intestine of capybara, presenting a spiral path, as reported for other mammals, with a spiral, circular or semilunar path (BANKS 1992; GEORGE *et al*., 1998), being also observed smaller folds, in a network arrangement. George et al. (1998) reported that the folds do not occur in the initial region of the duodenum, are higher in the jejunum, and are much less prominent in the ileum as they reach the colon. Unlike George *et al*. (1998) findings, the folds are present in the cranial portion of the duodenum of the capybara, being even apparently higher and branched in the duodenum than in the jejunum and ileum. A similar result was found by Velásquez *et al*. (2003), who observed in the duodenum of capybara more developed circular folds that gradually decrease in number towards the ileum.

According to Andrew and Hickman (1974), finger-shaped villi are the only structures in relief on the inner surface of the intestine of mice, while in man and many other large mammals circular folds also occur, which include mucosal and submucosal projections. Although capybara belongs to the Rodentia order, theyproved to be different from mice and similar to man and other large mammals concerning the presence of permanent intestinal folds. The existence of these folds represents another form of adaptation of the small intestine to increase digestion and absorption surface.

As seen in other mammals (BANKS, 1992; GEORGE *et al*., 1998; HENRIKSON, KAYE; MAZURKIEWICZ, 1999), the mucosa of the small intestine of capybara has villi and crypts, composed ofa lining epithelium, a lamina propria, and the muscularis mucosae. Finger-shaped villi are found in the small intestine of most mammals (BANKS, 1992; GEORGE *et al*., 1998), but villi can show species-specific and regional variations, such as the different patterns presented in the intestinal segments of capybara.

In Xenarthra Order species, finger-shaped villi were observed in armadillos (*Dasypusnovemcinctus*) and sloths (*Bradypustorquatus*), while in anteaters (*Myrmecophagatridactyla*) the villi are leaf-shaped (CARVALHO *et al*., 2014). In short-tailed monkeys (*Macacaspeciosa*) villi are tongue-shaped in the duodenum and finger-shaped in the jejunum and ileum (BURKE; HOLLAND, 1976), the same occurring in hamsters and dogs (TAYLOR; ANDERSON, 1972). In rats, villi show ridge shapes, but present differences in theirdimensions and configurations between the duodenum, jejunum, and ileum regions (HOSOYMADA; SAKAI, 2005). The bat *Desmodusrotundus*has pyramidal villi throughout the small intestine, while *Sturniralilium* showed pyramidal villi in the duodenal region and finger-shaped villi in the other regions (GADELHA-ALVES; ROZENSZTRANCH; ROCHA-BARBOSA, 2008). Wiese, Simon and Weyrauch (2003) verified the presence of finger-shaped, leaf-shaped and tongue-shaped villi in the small intestine of weaned piglets, being the latter in great number. Ramifications and lateral dilations in the villi of these animals were also described, similar to what was observed in some villi of capybara, even forming networks in certain regions, especially in the ileum.

The villi of the small intestine of capybara have an average height close to the lowest value indicated by Junqueira and Carneiro (2004) for mammals (0.5 to 1.5 mm). Despite this relatively low value, it is relevant to note that many of the villi present in the small intestine of capybara are wide and/or branched, leading to an enlargement of the surface area.

According to Brennan, McCullough and Carr (1999), the villi height in the small intestine of mice decreases towards the tail region. A villi sizedecreasewas also observed in crab-eating foxes (*Cerdocyon thous*), in which duodenal villi are larger than those in other regions (HELENO *et al.,* 2011). Hosoyamada and Sakai (2005) found that villi and crypts in rats are greater in the duodenum.

Although there was no significant difference in these parameters between the intestinal segments of the capybara, the height of the villi and crypts was greater in the duodenum, an important segment for digestion, by receiving pancreatic and biliary juices.

The lining of the small intestine of a capybara is a simple prismatic epithelium, similar to that described for mammals, composed ofabsorptive, goblet, Paneth and enteroendocrine cells, presenting their typical characteristics (BANKS, 1992; GEORGE *et al*., 1998; HENRIKSON *et al.,* 1999; JOHNSON, 2007; LLANOS, 1996; SWENSON; REECE, 1996). The absorptive cell's brush border appeared thicker in the jejunum and especially in the ileum, whichrepresents an adaptation to increase the absorptive surfaceof these segments. Regarding to the ileum, this is an important factor for the assimilation of nutrients that return from the cecum by retroperistalsis (GONZÁLEZ-JIMÉNEZ, 1995; SWENSON; REECE, 1996), since capybara is an herbivore that performs cecal fermentation, therefore having a well-developed cecum (BRESSAN *et al*., 2005; GONZÁLEZ-JIMÉNEEZ, 1995; KIANI *et al*., 2019).

According to reportedfindings ofmammals (BANKS, 1992; GEORGE *et al*., 1998; ROSS; ROMRELL, 1993; VELÁSQUEZ *et al*., 2003), goblet cells are less frequent in the capybara duodenum and progressively increase towards

the ileum. These mucus-producing cells'main function is to lubricate and protect the intestinal epithelium. Although goblet cells are less frequent in the duodenum, there are duodenal glands (Brünner's), in which mucus and bicarbonate production occurs. These glandular products have a protective role, especially against the gastric acid released in this segment. According to Banks (1992), the Brünner'sglands can be mucous, as seen in man, in ruminants and dogs; seromucous as reported to cats and capybara (present work); or serous, as found in horses and pigs.Concerning Brünner's glands distribution in humans, carnivores, small ruminants, the cane rat (*Thryonomys swinderianus)*,and rodents like capybara (ALOGNINOUWA; AGBA; KPODEKON, 1996), it is known that they occur exclusively between the initialand medium duodenal regions; in rabbits, they extend throughout all the duodenal segments (AINSWORTH *et al*., 1995); and in horses, pigs and large ruminants they reach the jejunum (GÓMEZ-ÁLVAREZ; SERRANO, 1983; NICKEL; SCHUMMER; SEIFERLE, 1973). On the other hand, González-Jiménez (1995) and Velásquez *et al*. (2003) reported the absence of Brünner's glands in the duodenum of capybara. However, the analyzedduodenal fragments were obtainedfrom the caudal portion, whichled to the wrong conclusion about duodenal glands in this species.

Although Brünner's glands are known to produce an alkaline secretion (GEORGE *et al*., 1998; JUNQUEIRA; CARNEIRO, 2004), they present, in the duodenum of capybara, a large number of mixed cells, rich in acidic and neutral mucins, probably have a slightly lower pH than that reported for other mammalian species. However, the low duodenal pH can be fixed by the pancreatic juice that, according to Reece (1996), is made of a large amount of buffering bicarbonate solution, especially in non-ruminant herbivores.

Additionally, the report by Ainsworth *et al*. (1995) revealed that the highest rates of bicarbonate secretion in the proximal duodenum are independent of Brünner's glands, in rats and rabbits. Bicarbonate, in addition to protecting the intestinal mucosa against acidic gastric juice, is also responsible for maintaining the optimal pH levels for pancreatic enzymes action. Concerning the non-ruminant herbivores, the large volume of buffering fluid secreted by the pancreas is important for the microbial digestion occurring in the cecum and colon. Thus, the pancreatic juice performs a role similar to that of ruminant's saliva (REECE, 1996).

The capybara *Hydrochoerushydrochaeris*, as reported by the present work, issimilar to most mammals regarding the presence of Paneth cells in the small intestine, with characteristics and location comparable to those described by Banks (1992). These cells are believed to be involved in controlling intestinal flora and defending against pathogens. This protective pathwaytakes place through lysozyme, an enzyme that breaks the bacteria cell wall, in addition to other antimicrobial products (BANKS, 1992; BEVINS, 2004; JOHNSON, 2007). Since acidophilic and PAS-positive serous cells are present in the duodenal glands of capybara (as well asPaneth cells), we believe that their serous secretion may contain antimicrobial products, which increasesintestinal protection, something especially important in an animal that performs cecotrophagia.

In the present work, enteroendocrine cells were observed along the small intestine of capybara, being predominant in the crypts, as reported for other mammals (BANKS, 1992; GARTNER; HIATT, 1993; GEORGE *et al*., 1998). Endocrine argyrophilic and argentophilic cells, predominantly pyramid-shaped ("open-type"), were identified in the small and large intestines of capybara, especially in the crypts (BRESSAN *et al*., 2004; SARTORI *et al*., 2018). Enteroendocrine cells produce many peptide hormones or amines, which act by controlling secretory and motor functions in the digestive tract. These chemical messengers can be identified by immunohistochemicaltechniques, which have already been done for serotonin in the intestine of capybara (BRESSAN *et al*., 2004; SARTORI *et al*., 2018).

The neural control of digestive events is performed by the nervous plexuses, mainly: the submucosal plexus, involved in transepithelial ion transport control, mucosal blood flow and secretory and motor functions; and the myenteric plexus, involved in intestinal motility control (HENRIKSON *et al*., 1999; HUDSON *et al*., 2000).

As reported by Velásquez *et al*. (2003) about the digestive tract of capybara, in the present work, submucosal and myenteric nerve ganglia with large bodies of neurons were observed. The ganglia were quantified and numerically correlated with enteroendocrine cells number in different segments of the small intestine of the capybara. Amorphophysiological correlation between neuroendocrine elements wasshown (SARTORI *et al*., 2018), especially in the ileum cecal transition, as well as in the cecum (BRESSAN *et al*., 2004, 2005).

The small intestine of capybara's lamina propria presents standard characteristics reported for other studied mammals: it is composed of a loose connective tissue with blood and lymph vessels, smooth muscle cells, connective and immune cells, especially lymphocytes (BANKS, 1992; GEORGE *et al*., 1998). The lymphocytes have a scattered distribution in the lamina propria and epithelium, and especially in the submucosa, they present a nodular arrangement, forming the Peyer's patches. Nickel *et al*. (1973) reported that Peyer's patches can have from a few centimeters to meters in length, reaching up to 3.5m in the intestine of pigs. These authors also showed that the number, size, and shape of lymphoid nodules, both in the isolated or associated form, vary with the age, intestinal region, the animal's diet, and species.

In capybara, lymphoid nodules were larger and the greatest number in the ileum, additionally, Peyer's plaques were found in the ileum caudal portion. Thus, the ileum represents a strategic segment for intestinal immune defense, especially in

animals that perform cecal fermentation.

The muscular is mucosae layer, responsible for villi contraction and movement, is similar to that described by Bacha and Bacha (2000): consists of smooth muscle bundles that, at certain points, penetrate the villi axis. And as reported for many mammals (BANKS, 1992; GEORGE *et al*., 1998), it usually presents a double layer of muscle bundles, being discontinuous in certain stretches, such as in the initial portions of the duodenum. This fact was also observed by Alogninouwa *et al*. (1996) with the cane rat.

The submucosal layer of the small intestine of the capybara is a dense connective tissue, as described by Henrikson *et al*. (1999) and in disagreement with that described by Banks (1992) as a loose connective tissue. The composition of the muscular layer, as well as the serous layer, is similar to that described for most mammals (BANKS, 1992; GEORGE *et al*., 1998). The muscle is arranged in an internal circular and an external longitudinal layer. According to Kent and Miller (1997), the circular layer, through contractions and relaxations of its fibers, constricts and dilates the intestine, while the longitudinal layer promotes the shortening of the intestine when contracting. Therefore, the coordinated action of these two muscle layers is responsible for intestinal movements of peristalsis and segmentation. In the present work, no significant variation for the thickness of these layers was observed.

4 Conclusion

The finding in this work allowsus to describe morphological features about the small intestine of the capybara *Hydrochoerus hydrochaeris*: the thickness of the wall and the constituent layers do not vary significantly along the intestine.

Thick and branched folds are present in the intestinal lining and are more developed in the duodenum. There are villi with different shapes and sizes, including the branches type and the brush border is thick, especially in the jejunum and ileum.

Brünner's glands are present in the cranial and middle portions of the duodenum and present seromucous secretion with a great amount of acidic and neutral glycoconjugates.

Goblet cells are numerous, increasing their density from the duodenum to the ileum, being concentrated in the crypt region, and producing a secretion of acidic and neutral glycoconjugates while Paneth cells and enteroendocrine cells are present throughout the intestine.

Lymphoid nodules are frequent and isolated in the mucosa and submucosa of different intestinal segments or associated forming Peyer's patches in the ileum caudal portion and the submucosal and myenteric ganglia are well developed and are present throughout the intestine.

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