Histomorphological and Histochemical Study of Esophagus and Stomach in Adult Guinea Pigs (Cavia porcellus)

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**ABSTRACT:** This study was performed to compare the esophagus and stomach histomorphological and histochemical characteristics during adulthood to clarify micromorphological alterations. In addition to the importance of this animal species as a model in many scientific experiments and due to some of its unique anatomical and physiological features. To conduct such a project, ten healthy adult guinea pigs at 2 months age and weighing 461.8±8.3g were gathered from the public spinning market. The esophagus is a musculo-membranous tube structure extended from the pharynx to the stomach, measured about 10.54±0.14 cm in length and 0.35±0.01 g in weight, and segmented according to its position in the body into three sections: the cervical, thoracic, and abdominal. The stomach is a simple unilocular transverse grayish white sac-like structure of the digestive system; it connects the esophagus to the intestine; its mean length was 4.39±0.11 cm; its capacity was 17.01±0.22 ml; and its weight was 7.07±0.05 gm. It lacks an outward demarcation-limiting ridge between its divisions and appears relatively smooth. Histologically, the esophagus is devoid of glands, while the stomach's three interior sections are dotted with glands, and both organs consist of the same four basic histological layers.

**Keywords:** histomorphological study, epithelia, esophagus, stomach.

1. **INTRODUCTION**

One of the most common rodents in South America is the progenitor of the guinea pig (Cavia porcellus), due to their popularity as pets and food sources, they are widely spread, belong to the order Rodentia, suborder Hystricomorpha, and the Family Caviinae [1, 2]. The animals mostly consume complex and insoluble feed materials. Therefore, the digestion process aims to gradually transform the feed materials into soluble, simple forms that are suitable for consumption and absorption [3]. Because of adaptation brought about by long-term natural selection processes, the gastrointestinal tract (GIT) has a tremendously diversified physical structure and function. To adapt to these environmental changes, rodent GITs have extremely specialized morphological, histological, and functional characteristics that have evolved throughout time. That includes the esophagus and stomach, among its most important vital parts [4, 5]. The esophagus is a cramped, muscular tube that is folded and connects the pharynx to the stomach's cardio in a straight line. It passes through the cervical area and thoracic cavity before passing through the diaphragm and entering the stomach through the cardiac sphincter. Starting at the inferior margin of the cricopharyngeal muscle and ending at the lower esophageal sphincter. As a result, the esophagus has been identified as three distinct macroscopic portions the cervical, thoracic, and abdominal [6, 7, 8, 9]. The stomach, which makes up the majority of the gastrointestinal tract, stores, digests, and churns food into a semi-liquid termed a chyme before sending it to the duodenum. Vertically within the abdominal cavity, situated between the esophagus and the small intestine [10]. It occupies most of the left side of the median plane, lying under the ribs and behind the diaphragm in the left half of the abdomen. Greater curvature (major curvature ventriculi), convex posterior surface, and less curvature (minor curvature ventriculi) and concave anterior surface are the anatomical properties of the stomach [6, 8]. There are four components to a simple stomach: the cardia, fundus, body, and pylorus. The esophagus reaches the fundus portion of the stomach through the cardiac section, which is the Histologically, a thick, stratified squamous epithelium is lining the esophagus. Food and species diversity also affected the keratinization of the epithelium,
whilst the stomach's lining was glandular simple columnar epithelium [6]. Because of the significance of these two digestive system organs, which are essential for turning food into the energy that the body needs to function. To shed light on the qualitative changes that occur in these organs at adulthood and to highlight the importance of this crucial laboratory animal in biomedical model research this study examined the histomorphological characteristics of the adult guinea pig’s esophagus and stomach.

2. MATERIALS AND METHODS

Ten healthy adult guinea pigs measuring 26.44±0.23cm in length and weighing 461.8±8.3g were collected from the public spinning market. Following the standard clinical exam, they seemed normal on the outside, and there was no indication of a systemic illness. The animals were first given an intramuscular injection of overdose 85-95mg/kg. BW ketamine HCl to anesthetized and sacrificed [11,12, 13, 14]. Then applied the classic dissection method to the morphological examinations in a way that allowed the best conditions of the esophagus and stomach to be highlighted in situ and determined the relationships between each organ and the other structures [15, 16, 17]. The study involved the collection of the esophageal and stomach specimens for histological and histochemical analyses [18, 19]. Hematoxylin Eosin and Masson's trichrome stains were used on the specimens to determine micromorphometric parameters which have been calculated by T test program, get a general histological characteristic, and visualize connective tissues, particularly collagen, in tissue sections. The esophageal and stomach glands and cells were stained and identified using PAS and Alcian blue [20]. The statistical analysis data underwent statistical analysis using the T-Test statistics version (2024). The numerical results have been reported as the mean values with the standard error (SE). The mean values were analyzed for statistical significance using one-way analysis of variance (ANOVA). The statistical significance level was defined at P < 0.05.

3. RESULTS AND DISCUSSION

Esophagus: Morphologically, the esophagus is a tubular laminated musculomembranous organ that forms a direct connection from the pharynx to the stomach's cardio. It measures about 10.54±0.14 cm in length, 39.01±1.1 cm relative esophagus length, 0.35±0.01 gm in weight, 0.08±0.001 gm relative esophagus weight (Table 1-A, B) where there is a difference in the relative weight scale only. Its color ranges from white to pink. It can be divided into three sections based on where it is located in the body: the cervical, thoracic, and abdominal (Fig 1-A, B). The cervical segment extends the full length of the neck; it starts at the caudal end of the pharynx esophagus and ends where the first set of ribs and the right thoracic inlet meet. It weighs 0.10±0.002 gm and has a length of roughly 3.18±0.03 cm that is significant (Table 1-A, B). The omohyoid and sternohyoid muscles laterally, the cervical vertebra dorsally, the left and right common carotid arteries ventrolaterally, and the trachea ventrally encircle this portion. These results agree with [9, 21]. The length of the thoracic portion of the esophagus was 5.02±0.05 cm, and its weight was 0.16±0.002 gm which are not significant (Table 1-A, B). The thoracic portion of esophagus extends from the thoracic inlet dorsally connected to the caudal vena cava and it passes dorsally to the heart via the lung lobes' mediastinum before entering the esophageal hiatus through the diaphragm where is the vagus trunk gathers ventrolaterally with the thoracic portion of the esophagus (Fig 1-A, B). With the exception of measurement data, this observation is comparable to that of [17], who reported that the cat's esophagus was situated ventrally to the ventral cervical muscles and dorsally to the trachea. The short abdominal segment of the esophagus was located on the posterior surface of the left lobe of the liver's esophageal groove, measuring 2.19±0.07 cm in length and 0.07±0.003 gm in weight not significant (Table 1-A, B). It extended from the esophageal hiatus of the diaphragm to the gastroesophageal at the stomach cardiac orifice. The base of the esophagus is surrounded by a thicker region of muscle fibers making a circular muscle known as the esophageal sphincter (Fig 1-B). These results agree with [8, 22] stated that the esophagus abdominal part begins after the diaphragm and ends in the stomach cardiac. The histological findings demonstrated that the mucosa, submucosa, muscularis, and adventitia or serosa were the four tunics that made up the esophagus in all three locations (cervical, thoracic, and abdominal). The esophageal tunica mucosa was folded creating a star-shaped, wide lumen (Fig 2-1). This is comparable to the observation published by [16], who observed that the rabbit esophageal lumen looks star-shaped in cross-section. In the cranial, intermediate, and caudal esophageal areas, the thickness of the mucosa was increased gradually as the follow measurements: 197.4, 224.9, and 319.8 um (Table- 3) which are significant. The mucosa is composed of the epithelium, lamina propia, and muscularis mucosae. Along the esophagus length, the epithelia were keratinized stratified squamous epithelium. It was interdigitated with well-developed lamina propria which is composed of relatively dense connective tissue, sporadic lymphocytes, and vascular structure (Fig 2-3). In agreement with [23], who stated that the lamina propria looked to be interdigitated with the keratinized stratified squamous epithelium that lined the camel's esophagus. The muscularis mucosae were composed of thin smooth muscle cell layers in the cranial part, becoming thicker in the middle and caudal portion of the esophagus (Fig 2-3, 4). However, these results disagree with [9] who states that the lamina muscularis in grey mongooses was thin and consisted of scattered interrupted skeletal muscle bundles in the cervical part. Loose connective tissue rich in collagen fibers and a small number of elastic fibers, along with some blood vessels and lymphocytes, comprise the non-glandular...
submucosa (Fig 2-1, 2, 4). The thickness of this layer varies along the esophagus, as in the following measurements, according to the sequence of parts 23.8, 21.7, and 47.4um (Table 2). This observation, which included some elastic and collagen fibers but no obvious submucosal glands, was in step with the findings reported by [23]. The cranial region of the esophagus had a tunica muscularis made up of striated muscle fibers that gradually changed into mixed fibers in the intermediate region and smooth fibers in the caudal region. It was composed of three layers that went from outside to inside: the thinner layer on the outside, formed by muscle fibers oriented longitudinally; the thicker layer in the middle, muscle fibers disposed circularly; and the internal layer, which had a longitudinal arrangement of muscle fibers (Fig 2-2). But it may also consist of only two layers in some parts, its thickness varies among the three parts: the abdominal part is the thicker at 430.5um, and 190.2, 259.4 um subsequently in the cervical and thoracic parts (Table -2) which are show significant. Concurred with [16] who stated that muscularis externa thickness varies along the esophagus. The adventitia, the outermost layer of the esophagus, is composed of loose connective tissue that is highly vascularized and deeply innervated in the pre-diaphragmatic segment, and the serosa, or simple squamous epithelium, in the post-diaphragmatic segment (Fig 2-1). Is similar [9, 16, 24].

Table 1. A: Morphometric esophagus results show the mean

<table>
<thead>
<tr>
<th>Mean</th>
<th>Body length</th>
<th>Esophagus length</th>
<th>Relative Esophagus length</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>26.44±0.23</td>
<td>10.54±0.14</td>
<td>39.01±1.1</td>
</tr>
<tr>
<td>3.18±0.03</td>
<td>5.02±0.05</td>
<td>2.19±0.07</td>
<td></td>
</tr>
</tbody>
</table>

The different letters refer to significant differences at (P≤ 0.05)

Table 1. B: Morphometric esophagus results show the mean

<table>
<thead>
<tr>
<th>Mean</th>
<th>Body weight</th>
<th>Esophagus weight</th>
<th>Relative Esophagus weight</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>461.8±8.3</td>
<td>0.35±0.01</td>
<td>0.08±0.001</td>
</tr>
<tr>
<td>0.10±0.002</td>
<td>0.16±0.002</td>
<td>0.07±0.003</td>
<td></td>
</tr>
</tbody>
</table>

The different letters refer to significant differences at (P≤ 0.05)

Table 2. Histometric esophagus results show the mean

<table>
<thead>
<tr>
<th>Mean</th>
<th>Lumen</th>
<th>Keratin</th>
<th>T.mucosa</th>
<th>T.submucosa</th>
<th>T.muscularis</th>
<th>T.adventitia or T.serosa</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cervical part</td>
<td>620.6</td>
<td>81.6</td>
<td>197.4</td>
<td>23.8</td>
<td>190.2</td>
<td>17.0</td>
</tr>
<tr>
<td>Thoracic part</td>
<td>800.1</td>
<td>50.9</td>
<td>224.9</td>
<td>21.7</td>
<td>259.4</td>
<td>10.6</td>
</tr>
<tr>
<td>Abdominal part</td>
<td>995.7</td>
<td>69.5</td>
<td>319.8</td>
<td>47.4</td>
<td>430.5</td>
<td>15.5</td>
</tr>
</tbody>
</table>

The different letters refer to significant differences at (P≤ 0.05)

Stomach: morphologically the stomach in the current study is very simple sac-like structure that is uniformly shaped, grayish white, and has two surfaces (parietal and visceral), two openings (cardiac and pyloric), two curvatures (lesser and greater), and two extremities. It is situated transversally in the abdominal cavity, to the left of the median plane, between the small intestine and the esophagus, retro-diaphragm, and retro-hepatic (Fig 1-A, B, C). This observation is similar to [10] who stated that the stomach of rats is a huge vertical organ lying in the abdominal cavity while is (J) shaped sac in rabbits, bulky sac-like with distinct ridges in in African rodents it was U-shaped, and in laboratory mice with oval features [21, 24.25]. The stomach of the adult guinea pig was unicocular, exhibiting a smooth surface and lacking any external demarcations between its sections. The lesser curvature was significantly shorter and linked to the small intestine through the omentum, whereas the bigger curvature was notably wider and associated with the spleen, which extends considerably dorsally (Fig 1-B, C). Compared to other rodents such as grey mongoose, rabbits, and southern African rodents, the
stomach of this particular rodent can be described as either a compound unilocular stomach, an unspecialized unilocular hemiglandular stomach, a bilocular discoglandular stomach with a visible limiting ridge between non-glandular and glandular areas, or a multichambered stomach [9, 24, 25]. The internal surface of the stomach is composed of three main sites: the cardiac region, is connected to the stomach's superior opening which is thicker and rougher than others, the fundic region which is made up of a larger portion of the stomach due to including the fundus and the body, finally the pyloric region which has a pyloric antrum that is funnel-shaped connected to the duodenum by a pyloric canal (Fig 1-C). In contrast to [9] findings, which indicate that the stomach of grey mongooses has become narrower in its central region. In the internal surface of the stomach, there are numerous folds (rugae) starting in the fundic region, which gradually decrease towards cardiac and increase towards pyloric. These folds work to increase the inner surface of the stomach and thus increase the digestion process and its efficiency (Fig 1-C). This result resembles the principle of the grey mongoose, whose stomach contains longitudinal folds to increase the efficiency of digestion. While the stomachs of some animals, such as hamsters and rabbits, are entirely flat [21, 25]. The stomach's mean length was 4.39±0.11 cm, 16.66±0.51 relative stomach length, capacity was 17.01±0.22 ml, weight was 7.07±0.05 gm, and 1.53±0.03 gm relative stomach weight (Table 3) shows significant. The morphometric results are different from those seen in other rodents [6, 24]. The stomach histologically is simple glandular; its wall has all four basic layers: mucosa, submucosa, muscularis, and serosa, in the three regions (cardiac, fundic, and pyloric). The mucosa consists of three layers: epithelia, lamina propria, and muscularis mucosa which measured 252.8, 305.0, 245.2um subsequently in the cardiac, fundic, and pyloric regions (Fig 3-A, B, D) (Table 4). The simple columnar epithelium is the mucosal surface of the stomach, which invaginates deeply into the lamina propria, forming the gastric pits that are short in the cardiac region and long in both the fundic and pyloric (Fig 3-A, D). As mentioned in the study [26] for rabbits. The lamina propria was home to a complex network of blood vessels, neurons, and small glands, which are simple tubular, branching, or coiled glands made up of mucous-secreting cells that are cuboidal in shape with foamy cytoplasm and a basally situated nucleus, parietal cells with a central nucleus, spherical chief cells with a pale granule-filled top area and a darkly pigmented basal nucleus, enteroendocrine cells, and undifferentiated cells (Fig 3-B, C). The glands shape and cells type distribution differ through the stomach regions. The same was observed by [24, 27] in Chinchil lalaniger and African rope squirrel. Smooth muscle fibers arranged in a circle make up the thin muscularis mucosa, followed by thin tunica submucosa of 46.7, 89.5, 81.1um subsequently in stomach three parts that revealed that is thicker in the middle part (Table 4) which are not significant. Submucosa consists of lymphocytes, blood vessels, and loose connective tissue. subsequent by the thick tunica muscularis, composed of circular muscle fibers in the medial and longitudinal muscle fibers on the outside and inside with 236.5, 199.7, 214.6um measurement in the cardiac, fundic, and pyloric (Table 4) that are significant. Like that observation by [28]. Finally, the outermost layer of the stomach is the tunica serosa, which is measured subsequently at 22.5, 24.2, 21.8 um (Table 4) which are significant. It consists of blood vessels, nerves, and areolar adipose connective tissue. As mentioned in the study [26] for rabbits. The histochemical results showed a positive for both the PAS and Alcian blue reactions, which suggests they synthesize both neutral and acidic mucins. The gastric glands appear magenta for PAS, and blue for Alcian blue (Fig 3-E, F). The same was reported by [20].

**Table 3. Morphometric stomach results show the mean**

<table>
<thead>
<tr>
<th>Mean</th>
<th>Stomach length</th>
<th>Relative Stomach length</th>
<th>Stomach capacity</th>
<th>Stomach weight</th>
<th>Relative Stomach weight</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>4.39±0.11</td>
<td>16.66±0.51</td>
<td>17.01±0.22</td>
<td>7.07±0.05</td>
<td>1.53±0.03</td>
</tr>
</tbody>
</table>

The different letters refer to significant differences at (P≤ 0.05)

**Table 4. Histometric Stomach results show the mean**

<table>
<thead>
<tr>
<th>Mean</th>
<th>T.mucosa</th>
<th>T.submucosa</th>
<th>T.muscularis</th>
<th>T.serosa</th>
</tr>
</thead>
<tbody>
<tr>
<td>cardiac</td>
<td>252.8</td>
<td>46.7</td>
<td>236.5</td>
<td>22.5</td>
</tr>
<tr>
<td>fundic</td>
<td>305.0</td>
<td>89.5</td>
<td>199.7</td>
<td>24.2</td>
</tr>
<tr>
<td>Pyloric</td>
<td>245.2</td>
<td>81.1</td>
<td>214.6</td>
<td>21.8</td>
</tr>
</tbody>
</table>

The different letters refer to significant differences at (P≤ 0.05)
Figure 1. **Topographic photograph in adult guinea pig shows:**

(A) Esophagus (E), stomach (S), pharynx (Ph), trachea (T), lung (Lu), diaphragm (D), liver (L), small Intestine (Si), and large Intestine (Li).

(B) Cervical section (Ce), thoracic section (Th), abdominal section (Ab), esophageal hiatus (H), esophageal sphincter muscle (Sm), spleen (Sp), and stomach (S).

(C) Gastroesophageal junction (Gj), pyloric antrum (Pa), lesser curvature (Lc), greater curvature (Gc), cardiac region (Ca), fundic region (Fu), pyloric region (Py), cardiac opening (Co), pyloric opening (Po), and rugae (R).
Figure 2. Histological cross section of the esophagus in adult guinea pigs shows:
(1) Lumen (L), keratin (K), mucosa (M), submucosa (Sm), muscularis (Me), and adventitia (Ad). X4 H&E stain.
(2) Keratin (K), stratified squamous epithelium (E), submucosa (Sm), and muscularis (Me). X10 H&E stain.
(3) Keratin (K), lamina propria (Lp), and muscularis mucosa (Mm). X40 H&E stain.
(4) Collagen fibers (Cf), lamina propria (Lp), and muscularis mucosa (Mm). X100 Masson's trichrome stain.
(5, 6) No esophageal glands. X10, Alcian blue and PAS stains.

Figure 3. Histological cross section of the stomach in adult guinea pigs shows:
(A) Mucosa (M), Simple columnar epithelium (E), gastric pits (Gp), Submucosa (Sm), Muscularis externa (Me), blood vessel (Bv), and serosa (S). X10 H&E stain.
(B) Mucosal glands (Mg), submucosa (Sm), muscularis mucosa (Mm), and muscularis externa (Me). X40 H&E stain.
(C) Parietal cells (P), chief cells (C), and mucus neck cells (Mn). X100 H&E stain.
(D) Collagen fiber (Cf), gastric pits (Gp), lamina propria (Lp), and muscularis mucosa (Mm). X10 Masson's trichrome stain.
(E, F) Positive reaction for Alcian blue and PAS stain X100.
4. CONCLUSION
- The esophageal devoid of glands, While the stomach's three interior sections are dotted with glands.
- The inner surface of the stomach contains longitudinal and transverse folds (ruge) localized in the fundus and decrease towards the cardiac and pyloric in adults.
- The basic layers that comprise the digestive system are the same that comprise both organs.

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