



ISSN : 2348-1935

RESEARCH ARTICLE

Annals of Experimental Biology
2015, 3 (4):27-35

Anatomical and Histochemical Studies of the Large Intestine of the African Giant rat (*Cricetomysgambianus*-Water house)- I

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ABSTRACT

Forty African giant rats (AGRs), (*Cricetomysgambianus*-Waterhouse) were used for morphometric, morphologic, histologic and histochemical studies. The rats were sacrificed according to the method of Adeyemo and Oke (1990) and the different segments of the large intestine (caecum, colon and rectum) were weighed, measured and photographs taken. Transverse sections of the different segments of the large intestine were stained with Haematoxylin and Eosin for normal histological studies. For Histochemical studies, the transverse sections were stained with Alcian Blue (AB), Periodic Acid Schiff (PAS) and Alcian Blue-Periodic Acid Schiff (AB-PAS) to determine their nature of secretions. The large intestine was observed to have a mean weight and length of 19.98 ± 0.39 g and 75.57 ± 1.00 cm respectively. The large intestine was observed to be made up of the caecum, colon and rectum, and all had similar basic histological structures, none of them had villi. The mucosal surfaces were smooth with goblet cells. Histochemical studies of the large intestine showed a positive response to AB, PAS, and AB-PAS. The thickening of the tunica muscularis of the colon and rectum was correlated with the temporary storage and expulsion of fecal materials. The presence of large number of mucous secreting cells provided a mucous layer around the fecal pellets facilitating its release and protecting the epithelium.

Keywords: Anatomical, Histochemical, Large intestine and African giant rat (*Cricetomysgambianus*-Waterhouse).

INTRODUCTION

The African giant rat (AGR) is found in the rain forest zone where they are restricted to farmlands, grasslands and human habitations. They are frequently seen at night crossing roads, running along drains and in house compounds. They are social animals and as such several individuals live together in a burrow. They walk and run on their four legs, usually with their tails raised and are good climbers and jumpers. In Nigeria, these rodents are often incorrectly called “rabbits” or “Nigerian rabbits” [1].

In view of their abundance and size, the rodents are often eaten by the people and considered a delicacy. Their smoked carcasses are often seen in village markets. Attempts have been made to breed and rear the animal in captivity for food [2]. The rodent has a good potential for use as a laboratory animal [3] and has been shown to be a good host for the laboratory passage of *Schistosoma mansoni* and *Trypanosoma evansi* [4]. Recently, the rodent has been used to detect tuberculosis patients and to sniff out landmines in Mozambique [5]. The excessive and uncontrolled consumption of this animal poses a threat to the ultimate survival of this species and [6] a researcher had attempted to study its biology and domestication. As a contribution towards this pioneering effort, several

attempts at characterization of the reproductive organs have been made [7-11]. Other works on the AGR include those on the brain [12-15]. The dietary requirement and feeding habit of the African giant rat cannot be properly understood without a detailed knowledge of the digestive system. It is also a well-known fact that the subfamily, *Cricetomyinae*, exhibits food storing behavior [16] and it comprises pouched rats that obtain their common name from large storage pouches inside their cheeks. This animal collects seeds, fruits, roots and other foods on the ground pack them into the pouches and carry them to special food stores in their burrows, but all these behavioral acts appear to have been based on feeding habit in the wild. An understanding of the digestive system will aid in maintenance, domestication and preservation of wild and endangered species [16].

The morphology of the gastrointestinal tract (GIT) has been reported in a number of other animals such as the cattle, sheep, pig, horse, dogs [17], man [18], laboratory rats [19-20] and birds [21]. Despite the features of this rat that have been studied, the large intestine is yet to be fully investigated. The only work done on the digestive system of this rodent in this part of the country is on the morphometry, morphology, histology and histochemistry of some aspect of the digestive system [22-25] and not the large intestine.

Basic information on the morphometry, morphology, histology and the histochemical features of the large intestine of the African giant rat is not fully available. In addition there are no reports on the histochemical studies in the AGR which will help ascertain the nature of their secretion which is a common feature of the digestive tract.

MATERIALS AND METHODS

Animal Source

Forty adult African giant rats, (AGR) *Cricetomysgambianus*, of both sexes were captured alive in the wild around Samaru and Bomo villages in Zaria, Kaduna State, Nigeria from January to April 2009 using metal cage traps. They were transferred into standard laboratory rat cages in the Department of Veterinary Anatomy, Ahmadu Bello University, Zaria and fed with commercial feed for a while before sacrifice. Water was given *ad libitum* during the period. The captured African giant rats were used for morphometric, morphologic, histologic and histochemical studies

Morphometric studies

Forty of these rats were used for the morphometric study and each was weighed alive using a mettler balance (Model P1421) with a sensitivity of 0.1gm and the weight recorded in grams. Sex difference was not taken into consideration. Each rat was placed on a lateral recumbency and sacrificed according to [26]. An incision was made from the first cervical vertebrae up to the level of the pelvic region with the rat lying on a dorsal recumbency to show the esophagus and the content of the GIT. The entire GIT was exteriorized and the large intestine and its segments (caecum, colon and rectum), were weighed and their lengths measured and recorded. All these measurement were done using a Fatzun balance (model P141 with a sensitivity of 0.01 gm), a ruler and thread (in cm), respectively.

Morphologic studies

Photographs of the live AGR were taken before sacrificing [26]. The entire GIT was exteriorized and stretched out to full length. The positions of the pancreas, gall bladder and the lobes of the liver were noted. Photographs of all the relevant segments were taken at appropriate times *insitu* and when dissected out.

Histological studies

After sacrificing the rats [26], they were opened up and tissues were collected from the large intestinal segments. The tissues were immediately fixed by complete immersion in 10% normal formalin and labeled and kept for two days. They were dehydrated through a series of graded alcohol (70%, 80%, 90%, 95% and 100%). They were later cleared in xylene and infiltrated with molten paraffin wax. Transverse sections of 5µ thick were cut from the embedded tissues using disposable microtome knives. These sections were mounted on grease free clean glass slides and stained at room temperature using Haematoxylin and Eosin (H and E) method for routine histological studies.

Histochemical studies

For histochemical studies transverse sections from the embedded tissues were cut and stained with Alcian blue (AB), for acidic mucin identification, Periodic Acid-Schiff (PAS), for identification of neutral mucin and Alcian blue together with Periodic Acid-Schiff (AB-PAS) for the identification of both the neutral and acidic mucins.

The slides were studied using light microscope (Olympus binocular microscope) at x40 x100 and x250 magnifications. Photographs of the prepared slides mounted on the binocular microscope were taken using a digital camera. The pictures taken were transferred to a computer and detailed studies were carried on them. Relevant areas and structures were labeled and printed.

Statistical Analysis

Recorded weights and lengths studied were express as mean \pm standard error of mean (M \pm SEM) using statistical package for social sciences (SPSS) version 17.

RESULTS

Morphometry.

The mean weight of the AGR was observed to be 1045.0 \pm 28.06 g. The mean weight of the gastrointestinal tract was 58.18 \pm 0.89 g which accounted for 5.56% of the total weight of the animal. The large intestine and its segments(caecum, colon and rectum) were 19.98 \pm 0.39 g, 9.51 \pm 0.21 g, 8.83 \pm 0.21 g and 1.84 \pm 0.07 g, accounting for 1.91%, 0.85%, 0.88% and 0.18% of the total weight of the animal, respectively. The caecum had the highest mean weight followed by the colon, the rectum had the least mean weight (Table 1).

Table1: Morphometric Features of the Digestive System of the African giant rat (AGR) (g).

| Parameters | Minimum Value (g) | Maximum Value (g) | Mean \pm SEM | % body wt |
|---------------------------|-------------------|-------------------|--------------------|-----------|
| Body weight | 800 | 1400 | 1045.0 \pm 28.06 | 100 |
| Weight of GIT | 47.70 | 69.20 | 58.18 \pm 0.89 | 5.56 |
| Weight of large intestine | 15.40 | 25.70 | 19.98 \pm 0.39 | 1.91 |
| Weight of caecum | 5.90 | 11.40 | 9.51 \pm 0.21 | 0.85 |
| Weight of colon | 5.00 | 10.47 | 8.83 \pm 0.21 | 0.88 |
| Weight of rectum | 1.20 | 2.70 | 1.84 \pm 0.07 | 0.18 |

n = 40, SEM = Standard Error of Mean.

The mean length of the gastrointestinal tract was found to be 213.32 \pm 3.88 cm. The mean length of the large intestine and its segments(caecum, colon and rectum) were found to be 75.78 \pm 1.78 cm, 12.70 \pm 0.37 cm, 57.30 \pm 1.58 cm and 5.68 \pm 0.26 cm, respectively accounting for 35.50%, 6.0%, 26.90% and 2.60% of the total length of the GIT, respectively. The colon had the longest mean length followed by the caecum and finally the rectum (Table 2)

Table 2: Morphometric Features of the Digestive System of the African giant rat (cm).

| Parameter | Minimum Value(cm) | Maximum Value(cm) | Mean \pm SEM | % body wt |
|---------------------------|-------------------|-------------------|-------------------|-----------|
| Length of GIT | 169.0 | 264.50 | 213.32 \pm 3.88 | 100.00 |
| Length of large intestine | 54.00 | 95.00 | 75.78 \pm 1.78 | 35.50 |
| Length of caecum | 9.00 | 17.00 | 12.70 \pm 0.37 | 6.00 |
| Length of colon | 39.00 | 74.00 | 57.30 \pm 1.58 | 26.90 |
| Length of rectum | 3.00 | 9.00 | 5.65 \pm 0.26 | 2.60 |

n = 40, SEM = Standard Error of Mean.

Morphology

The large intestine was made up of the caecum, colon, and rectum. The caecum was the largest segment of the intestine. It was large, a cone-shaped, blind-ended sac situated at the ileo-cecal junction (Plate I). The colon was wrinkled in appearance with a wide lumen which contained fecal balls and had sacculations with a thick wall compared to the caecum (Plate I). The rectum was observed to be the caudal continuation of the colon and was observed not to be wrinkled but had some fecal pellets inside (Plate I). The anal opening formed the caudal end of the gastrointestinal tract (Plate I).

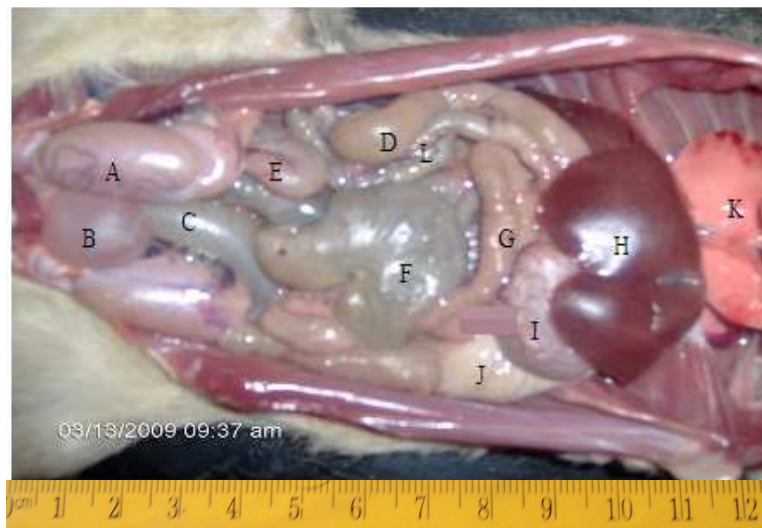


Plate I: Abdominal viscera of the AGR showing the testes (A), the urinary bladder (B), the rectum (C), the duodenum (D), the jejunum (E), the caecum (F), the transverse colon (G), the liver (H), the glandular stomach (I), the non glandular stomach (J), the lungs (K) and descending colon (L).



Plate II: The reflected part of the GIT of the AGR showing the pancreas (A), the caecum (B), the duodenum (C), the glandular stomach (D), the nonglandular stomach (E) and the kidney (F).

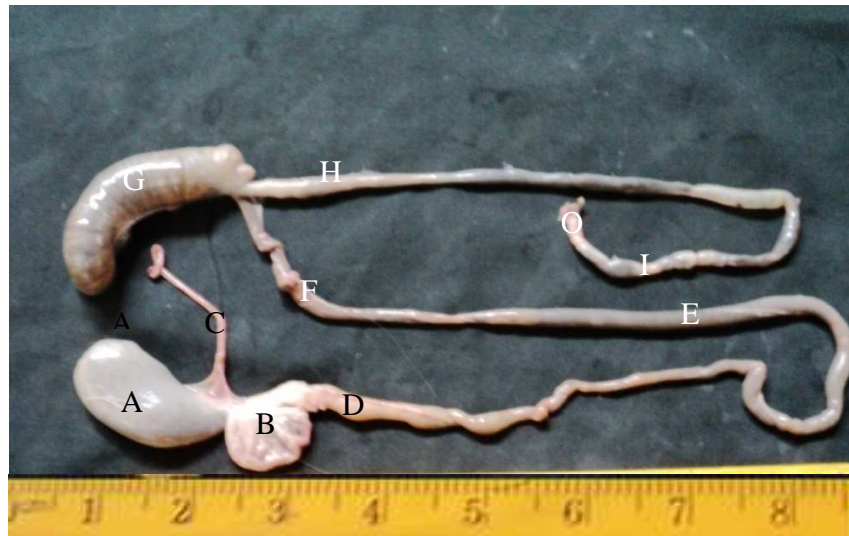


Plate III: The GIT of the AGR showing the non glandular stomach (A) , the glandular stomach (B), the esophagus (C), the duodenum (D), the jejunum(E), the ileum (F), the caecum (G), the colon (H), the rectum (I) and the anal opening (O).

Histology

Intestinal glands were observed to be prominent and the goblet cells were numerous (Plates IV,V and VI).The caecum was observed to have numerous goblet cells and intestinal glands. Adipose tissues were found in the submucosa and the tunica muscularis was made up of longitudinal muscle (Plate IV).The colon was observed to have Simple columnar cells with abundant goblet cells. The muscularis externa presented thick band of longitudinal muscles (Plate V).Like the colon and caecum, the rectum was observed to be made up of the goblet cells and intestinal glands which, were found to be more numerous compared to those of the caecum and colon (plate VI). All other features were observed to be similar to those of the colon and caecum.

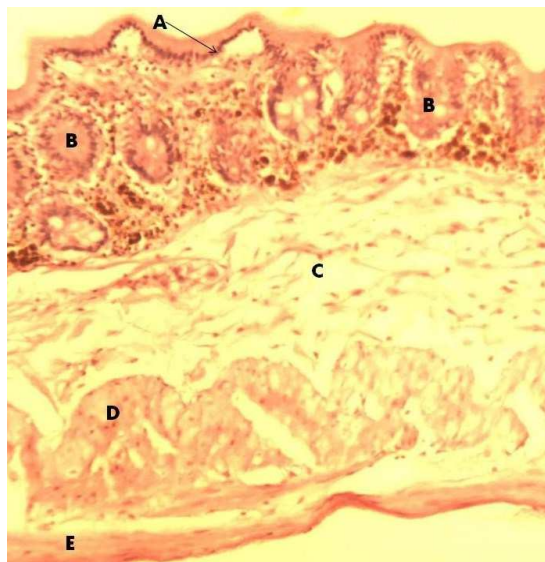


Plate IV: Transverse section of the caecum showing simple columnar epithelium (A),intestinal gland and goblet cell (B), submucosa(C) adipose tissue (D) and the tunica muscularis (E).H&E x100.

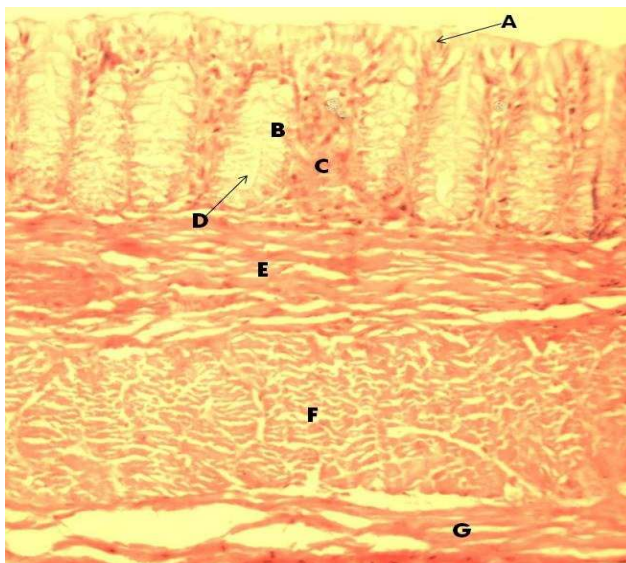


Plate V: Transverse section of the colon showing the epithelial lining (A), goblet cells (B), muscularismucosa (C), intestinal glands (D), submucosa (E),tunica muscularis (F) and tunica serosa (G). H&E x 100.



Plate VI: Transverse section of the rectum showing the absorptive columnar cells (A), goblet cells (B),intestinal glands (C), laminar Propria (D), muscularis mucosa (E), blood vessels (F), submucosa (G). H&E x 250.

Table4.3: Histochemical Reaction of the gastrointestinal tract and the Salivary Glands of the African giant rat (*Cricetomysgambianus* – Waterhouse).

| Organs | Histochemical Test. | | |
|--------|---------------------|-----|--------|
| | AB | PAS | AB-PAS |
| Caecum | + | + | ++ |
| Colon | + | + | ++ |
| Rectum | + | + | ++ |

+=Positive; - =negative.

Histochemical Features

Table 4.3 shows the Histochemical reactions of the large intestine of the AGR, using Alcian Blue (AB), Periodic Acid Schiff (PAS) and Alcian blue combined with periodic Acid Schiff (AB-PAS). Positive reactions were seen

when there is a color change to blue and pink for Alcian Blue and Periodic Acid Schiff and Magenta or blue and pink for Alcian Blue combined with Periodic Acid Schiff. The goblet cells and the intestinal glands of the large intestine were all observed to be AB, PAS and AB-PAS positive, respectively (Plates VII, VIII and IX).

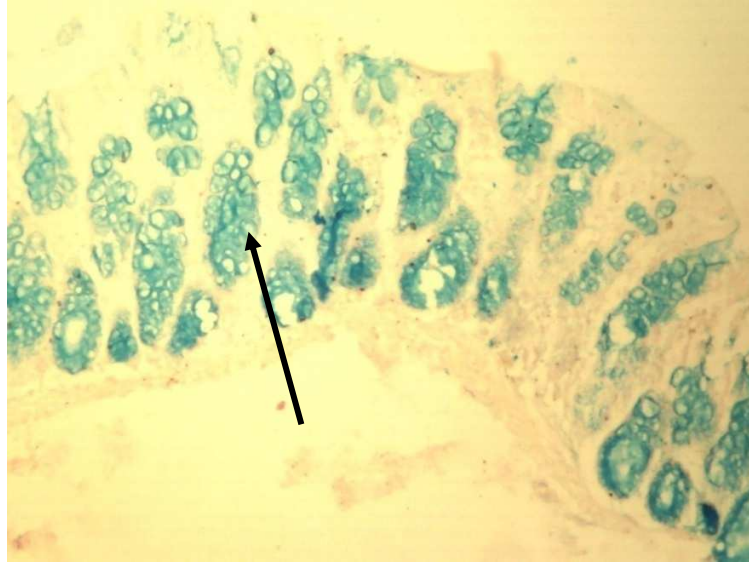


Plate VII: Transverse section of the intestinal glands and the goblet cells of the colon showing AB positive (arrow) x250.



Plate VIII: Transverse section of the intestinal glands and the goblet cells of the colon showing PAS positive (arrow) x250.

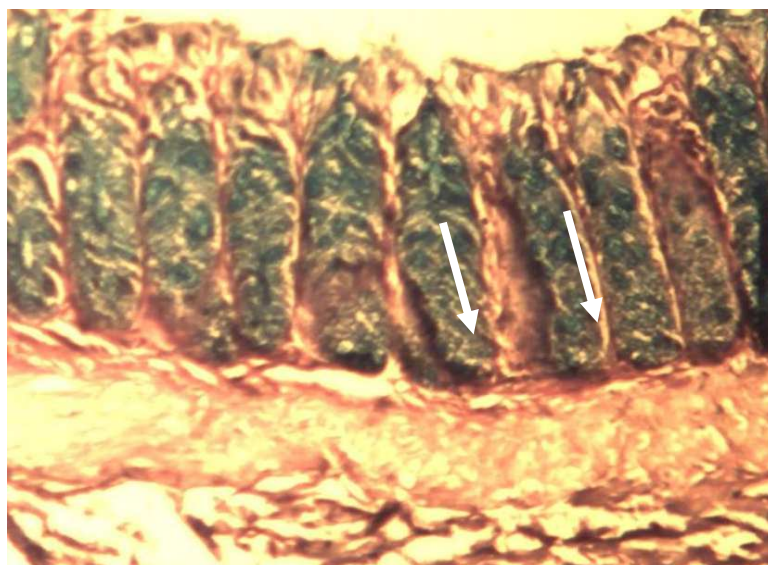


Plate IX: Transverse section of the intestinal glands and the goblet cells of the colon showing AB-PAS positive (arrows) x250.

DISCUSSION

The large intestine of the AGR was observed to be made up of caecum, colon and rectum. The caecum was the largest segment in the abdominal cavity and serves as a principal site for microbial fermentation. Vitamins and fatty acids were absorbed in the colon, water was also said to be absorbed in the colon resulting in fecal ball formation. The fecal balls which are the undigested and mostly indigestible portion of what was fed will then pass through the rectum to the anus. The histology of the large intestine of the AGR was characterized by the absence of villi and a gradual increase in the number of goblet cells from the caecum to the rectum. The intestinal glands were numerous in the caecum compared to the colon and rectum, while the goblet cells were more abundant in the colon and rectum than the caecum. The epithelium lining this region was observed to be simple columnar with long irregular microvilli, suggesting an absorptive function, particularly in the colon. The gross and histological studies of the large intestine in this study showed that its major functions are water absorption and formation of fecal mass in addition to the production of mucous to lubricate the intestinal surface. The observations of the large intestine in this study agreed with the findings of [27]. They reported that the caecum in the grasscutter is the largest internal organ, within the abdomen cavity. In the caecum, fermentation of the intestinal contents occurs and periodically the caecum contracts and the fermented ingesta are propelled into the colon and then out of the anus. The thickening of the muscularis has been correlated with the temporary storage and expulsion of fecal materials from this area. These fecal materials are coated with mucus which acts as a barrier to the acidic pH of the jejunum and the ileum. In the present study, the pattern of staining for acidic and neutral mucous substance with PAS at the mucosal surface and AB throughout the colon and rectum suggested the presence of acidic mucins in all these segments. The presence of neutral mucous substance may indicate absorptive functions. An increase in number of goblet cells in the rectum increases, the mucosa protection and lubrication for fecal expulsion. The presence of large number of mucous secreting cells provides a mucous layer around the fecal pellets, facilitating its release and protecting the epithelium as was described by [28].

CONCLUSION

The caecum was observed to be the largest organ found within the abdominal cavity and was observed to be a blind sac. Histological and histochemical study of the caecum, colon and rectum revealed numerous goblet cells and intestinal glands which were AB, PAS and AB-PAS positive.

The presence of large number of mucous secreting cells provides a mucous layer around the fecal pellets facilitating its release and the protection of the epithelium.

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