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HISTOLOGICAL AND HISTOCHEMICAL STUDY OF THE DIGESTIVE TRACT OF THE WORM-LIKE REPTILE*, *DIPLOMETOPON ZARUDNYI* (SQUAMATA)

By

AISHA S. AL-THANI** AND G. EL-SHERIF
Zoology Department, Faculty of Science, University of Qatar, P. O. Box 2713, Doha, Qatar

دراسة التركيب النسيجي وكيمياء النسيج للقناة الهضمية للزاحف شبيه الديدان «دبلوميتوبون زارودينى» (الحرشفيات)

عائشة سعود آل ثاني و جمال الشريف

يعتبر «دبلوميتوبون زارودينى» من الزواحف شبيهة الديدان المعروفة في البيئة القطرية باسم «النادوس»، وقد أجريت دراسة بالمجهر الضوئي للتركيب النسيجي وكذلك تفاعل حمض البيرأيوديك وكاشف شيف . وقد لوحظ أن مخاطية المرئ تتكون من طلائية عمودية مصففة بينما مخاطية المعدة خلايا طلائية عمودية مخاطية . كما وجدت الخلايا الامتصاصية ذات الحواف الفرجونية والغدد الكأسية في مخاطية الأمعاء الدقيقة ، وفي الأمعاء الغليظة لوحظ وجود مخاطية هرمية الشكل وعدد كبير من الغدد الكأسية مع طبقة محددة بوضوح من المواد المخاطية . كما أظهرت خلايا المرئ والمعدة تفاعل إيجابي بصورة موحدة مع تفاعل البيرأيوديك وكاشف شيف بينما تميزت الغدد الكأسية بكميات كبيرة من عديدات التسكر المخاطية شديدة التفاعل . تراوحت الطبقة العضلية بين الألياف الطولية فقط في المرئ والطولية والعرضية الرقيقة في المعدة ، بينما تميزت الأمعاء الغليظة بوجود طبقتين سميكتين من الألياف الطولية والعرضية .

Key Words: Histology and Histochemistry, Digestive tract, Diplometopon Zarundnyi

ABSTRACT

The structure of the digestive tract of *Diplometopon zarudnyi* was studied by means of light microscopic histology and PAS reaction. The oesophagus is lined with stratified columnar epithelium while the stomach is lined with mucous columnar epithelium. The small intestine is lined with brush-border absorptive columnar cells and goblet cells, while the large intestine is lined with mucous pyramidal cells and larger goblet cells with an extreme layer of mucin lining. The oesophageal and gastric mucosae are uniformly PAS positive . In both the large and small intestine, goblet cells are normal with large amounts of mucin granules. The muscular layer varies in structure being only longitudinal in the oesophagus, thin longitudinal and circular in stomach and more thick in the large intestine.

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**Author to whom all correspondence should be addressed.

INTRODUCTION

The reptilian fauna of Qatar was briefly reported few years ago [1] . Reptiles of Kuwait were also recorded [2] .However no previous studies have been made on the histology and histochemistry of reptiles recorded in Qatar.

Diplometopon zarudnyi is a worm-like burrowing reptile classified at one time with the lizards but now generally given a subordinal rank within the Squamata in parallel with lizards and snakes since it has some characters typical of snakes and lizards [3] . It is limbless, long and cylindrical with blunt head, short pointed tail and very small eyes. It feeds on small invertebrates, ants and termites and lives in burrows in sandy areas. Its dorsal side has a spotted brown colour while the ventral side is whitish with smooth scales, It is locally known as "Nadus". It is known in El-Karaana area, south-west of Doha and Qatari western desert near Umm-Bab.

Few studies are known regarding histology of the digestive tract of certain reptilian species [4,5]. Other investigations concerned with histochemical and ultrastructural observations on the digestive tract of reptiles are also reported [6,7,8,9,10].

The main objective of this work was to investigate the histological structure of the digestive tract of *D.zarudnyi* with special reference to the polysaccharides in the tissues of the digestive system .

MATERIALS AND METHODS

Animals were collected in November 1995, identified and some individuals from both sexes were injected with formalin and preserved in jars containing 10% formalin. Eight other animals, were dissected under chloroform anaesthesia, the digestive tract was distinguished and then injected with 10% formalin with 1% CaCl2 in phosphate buffer saline (PBS). Transverse strips (5 mm wide) were cut at different parts of the digestive tract: the oesophagus,the stomach, the small intestine and the large intestine. Samples were fixed by extra immersion in buffered 10% formalin with 1% CaCl2 overnight then washed and routinely processed for paraffin embedding and 5 µm sections were prepared. These were investigated by H & E technique for general histological studies and by PAS for polysaccharide histochemistry.

RESULTS

Different organs of the digestive tract of *D. zarudnyi* exhibited histological and histochemical variations which can be summarized as follows:

OESOPHAGUS

The mucosa of the oesophagus showed many longitudinal folds. The muscosal epithelial lining is formed of stratified columnar epithelium with middle well-defined polyhedral cells (Fig. 1). Wide layer of longitudinal muscle fibres occurs underneath the distinct lamina propria.

After PAS reaction, the oesophageal mucosa exhibited a moderate regular distribution of polysaccharides on the lining epithelial cells (Fig. 2). However, the polysaccharide content was more intense in the cytoplasm of basal cells while surface polyhedral cells showed only PAS positive cell membranes. Lamina propria, submucosa and muscle layer were very poor in polysaccharides.

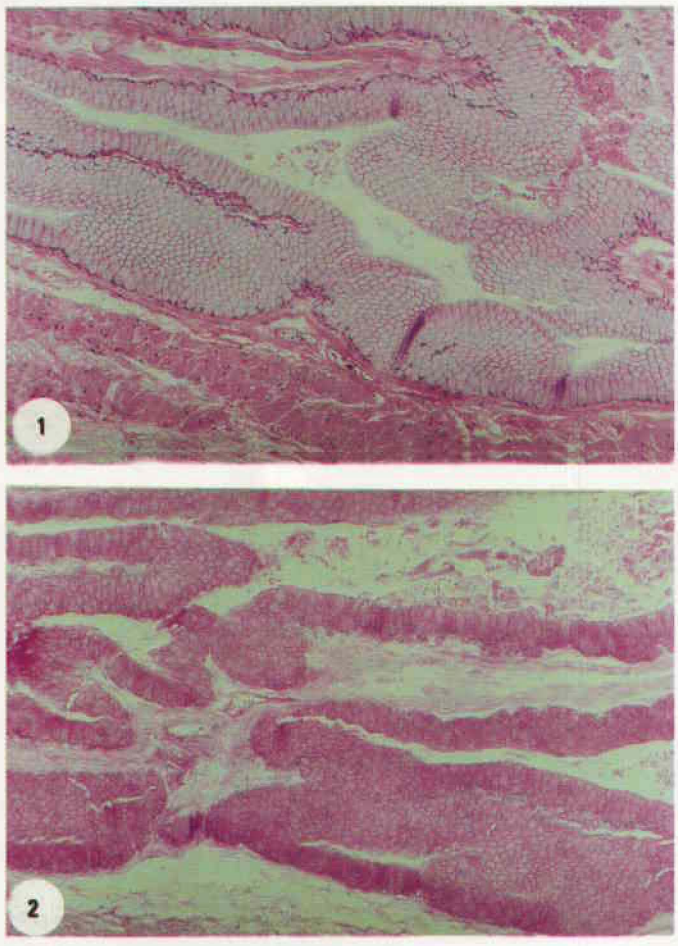


Fig. 1: Section of the oesophagus, showing mucosa lined by stratified columnar polyhedral epithelial cells with longitudinal muscle fibres underneath the lamina propria. H & E, x 100.

Fig. 2: Moderate regular distribution of polysaccharides in the oesophageal lining. Basal cells exhibit more intense reaction than the surface cells. PAS, x 100.

STOMACH

It shows prominent muscosal folds with a short simple gastric villi supported by a submucosal core. There was a thin layer of inner longitudinal and outer circular muscle fibres (Fig. 3). The gastric mucosa is built of mucous columnar cells with basal nuclei.The mucous columnar cells of the gastric mucosa revealed a uniformly polysaccharide distribution with PAS reaction (Fig. 4).

Fig. 3: Gastric mucosa of mucous columnar cells with basal nuclei and thin layer of outer circular and inner longitudinal muscle fibres. H & E, x 100.

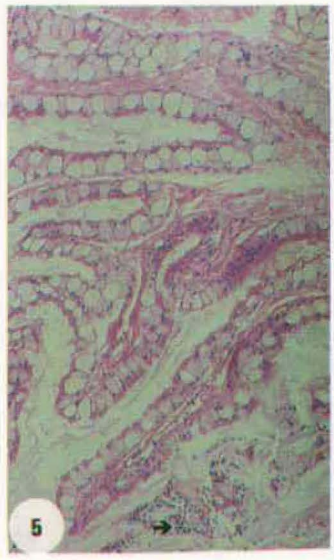
Fig. 4: PAS positive materials distributed along the gastric mucosal surface. PAS, x 100.

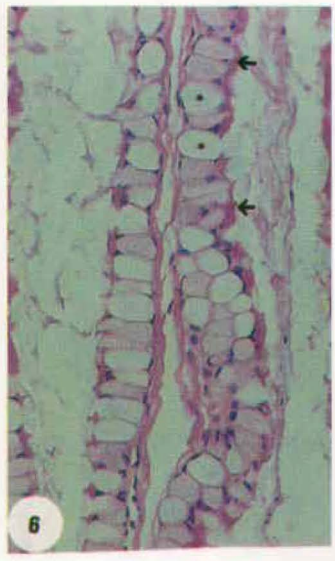
SMALL INTESTINE

It is characterised by long and narrow folds, with very poor musculature and submucosal lymph cell aggregations (Fig. 5) and well defined lamina propria with no special intestinal glands. The lining epithelium is composed of columnar brush-border absorptive cells with distinct microvilli and prominent large goblet cells (Fig 6).Intestinal lining epithelium is provided by goblet cells which are larger and more common than the absorptive cells. The former cells have abundant amounts of mucin granules (Fig. 7) and PAS positive lamina propria.

LARGE INTESTINE

The mucosa of the large intestine shows a characteristic pattern with short, branched but fewer folds than those of the small intestine. Submucosal cores and lymph aggregations are prominent. The large intestine is provided by thicker patches of inner longitudinal and outer circular muscle fibres than those of the small intestine ; vascularized submucosa is also observed (Fig. 8). The lining epithelium was formed of a single layer of mucous columnar cells incorporated with goblet cells (Fig. 9).





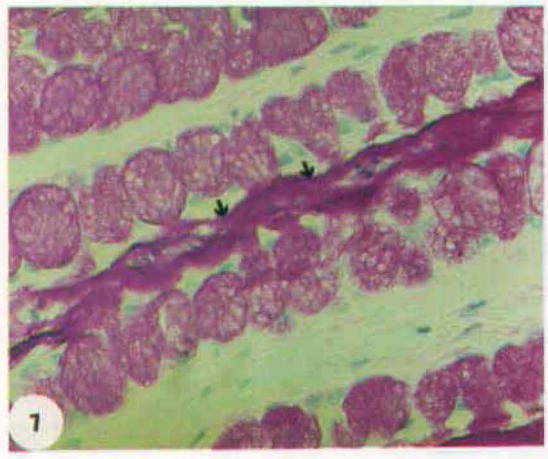
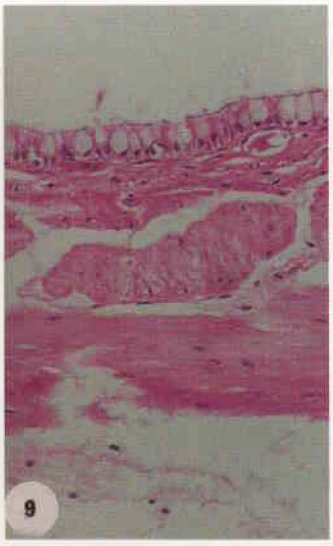


Fig. 5: Long and narrow folds of the small intestinal mucosa with scattered lymph aggregations (arrow). H & E, x 100.

Fig. 6: Mucosa of small intestine showing the columnar absorptive cells with brush-borders (arrows) and large goblet cells (asterisks). H & E. x 200.

Fig. 7: Large goblet cells of the small intestinal mucosa exhibited ideal PAS positive reaction with a distinct lamina propria (arrows) PAS, x 400

Large intestinal mucosal cells are covered by a layer of mucin with large number of goblet cells which are completely filled with larger amounts of mucin coming out of the goblet cells all over the mucosal surface (Fig. 10). Some columnar cells are binucleated.



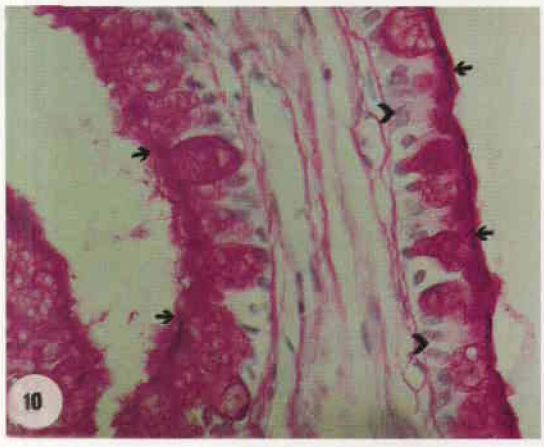


Fig. 8: Section in the large intestine showing short and branched villi with distinct inner longitudinal and outer circular muscles. H & E, x 100.

Fig. 9: Goblet cells and mucous columnar cells of the large intestinal mucosal lining. H & E, x 100.

Fig. 10: Mucosa of the large intestine with goblet cells completely filled by mucins forming a uniformal layer on the mucosal surface (arrows) and the binucleated columnar cells (arrowheads). PAS, x 400.

DISCUSSION

Previous studies showed that the epithelial lining of the oesophagus in reptiles consists of ciliated columnar type [11,12,13,14]. The oesophagus of a freshwater turtle was described as being lined by keratinized stratified squamous epithelium [15]. Unlike other reptiles, the oesophagus of *D. zarudnyi* is lined with stratified columnar epithelium.

The reptilian stomach was reported as having a gastric mucosa of 2 different cell types : mucous neck cells with their pyramidal shape and oxyntiopeptic cells [10,16 ,17]. On the other hand, the gastric mucosa of *D. zarudnyi* is found in the present work to be formed only of tall columnar oxynticopeptic cells with a distinct basal nuclei.

Goblet and absorptive cells of the small intestinal mucosa were described in many reptiles [4,5,10]. The small intestine of *D. zarudnyi* is lined by prominent large goblet cells and also by brush-border columnar absorptive cells which increase surface area for absorption. It was estimated that such brush-border columnar epithelium would increase the obsorptive surface area by about 600 folds [18].

In agreement with other findings in tortoises [10], the lining epithelium of large intestine of *D. zarudnyi* was composed of exclusively large goblet cells and mucous columnar cells.This confirms the observations of Madrid *et al* [9] in *Testudo graeca* where no absorptive cells were reported. The musculature of the digestive tract of *D. zarudnyi* exhibits different forms, being only longitudinal in the oesophagus where there is no need for transverse contractions of the circular muscles since the oesophagus is relatively short . Both longitudinal and circular muscle fibres form a thin layer which may help in the gastric contractions on both axes generating the peristaltic movements of the gastric wall during digestion. The poor muscular layer of the small intestine is probably enough for the passage of absorbed digested food to the large intestine. The latter has highly developed thick longitudinal and circular muscle layers which can produce two dimensional contractions to facilitate the process of defecation.

Lymph cell aggregations in the submucosa of the large and small intestines of *D. zarudnyi* may be considered as elements of the immune system in these animals.

Mucines exhibited moderate uniform distribution in the oesophageal and gastric lining epithelium especially the basal cells. This may protect the mucosa against mechanical injuries during engulfing and against chemical and bacterial attack during digestion. PAS positive mucins in the small intestinal mucosa may also be protective to the mucosa and allowing passage of wastes into the large intestine . The entire mucin coverage of the large intestinal mucosa confirms that the intestinal mucin partly play an important role in the protection of the mucosa against the physical and chemical damage [19] and also against bacterial invasion [20]. Such mucin lining facilitates the excretion of foeces, including hard wastes such as exoskeletons of invertebrates, ants and termites.

In conclusion, the histological features and PAS positive results in the digestive tract of *D. zarudnyi* common in Qatari fauna prove different from other reptiles in having oesophageal stratified columnar epithelium, gastric oxynticopeptic cells, small intestinal mucosa of brush-order columnar epithelium with goblet cells and a classical reptilian large intestinal mucosa.

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