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The Histological Structure of the Ileal Peyer's Patches of the Egyptian Water Buffalo *(Bos Bubalus)*

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ABSTRACT

The water buffalo represents an important source of animal products in Egypt and characterized by high resistance to several diseases and parasites. The present study carried out on the Egyptian water buffaloes of different ages (from 50 days to 8 years old buffaloes) to illustrate the structure of ileal Payer's patches with age. The specimens of ileum were fixed and processed for light and electrone microscopy The Payer's patches (PPs) formed of elongated dome regions flanked by intestinal villi. These domes were short, wide villi. The domes were consisted of lymphoid follicles covered with a typical dome- associated epithelium of enterocytes and M cells without any goblet cells in young and adult ages. The M cells showed variable appearance depending on the functional status. The lymphoid follicles had a clear germinal center. In conclusion Payer's patches (PPs) were still active until the age of 5 years old and then begin to be inactive at 8 years old we think there is a long period of activity which express the tolerance to many diseases.

1. INTRODUCTION

The water buffalo represents an important source of meat, milk and leather in Egypt, as the estimated herd number exceeds 3.6 million heads (FAO, 2002). Gut- associated lymphoid tissues form a central part of the inductive site of the mucosal immune system (Brandtzaeg and pabst 2004). Payer's patches (PPs) are sub-epithelial aggregation of lymphoid tissue located along the anti-mesenteric side of the small intestine (Owen and Jones, 1974: Owen, 1977). The number, structure, size and distribution of Payer's patches (PPs) vary according to species (Liebler-Tenorio and Pabst, 2006; Abe and Ito, 1977).

Payer's patches (PPs) is formed of groups of lymphoid follicles with clear germinal centers arranged in more than layer located in submucosa of the ileum. These follicles supported by reticular fibers. The follicle associated epithelium (FAE) overling the Payer's patches are formed of enterocytes and M cells among them. The morphology of M cells varies greatly amongst different animal species, and within anatomic sites of a species. For instance, the microfold structure is present only in human M cells (Owen & Jones 1974), also human M cells lack microvilli. In contrast, the microvilli are present on the surface of murine M cells, but these are short and irregular in contrast to the microvilli on the M cells of rabbit caecal lymphoid patches which were longer than the neighboring enterocytes (Owen 1999). The most important feature of the human M cell under light or electron microscopy was determined the absence of surface microvilli which were characteristic of the intestinal epithelial cells. Instead, the apical membrane of the M cell has a micro fold. Like other epithelial cells, M cells form tight junctions to maintain a barrier function, (Bockman, Cooper 1973& Kato, Owen 1999). The basolateral membrane of M cells is invaginated, and forms many “pockets”, which harbor infiltrating lymphocytes (Regoli et al 1995). The formation of these “pockets” greatly reduces the intracellular distance that antigens have to travel and allows M cells to rapidly transport (within 10 to 15 min) antigenic materials to the basolateral membrane (Owen 1977).

2. MATERIAL AND METHODS

The ilea of 30 clinically healthy Buffaloes from 40 days to 10 years old buffaloes were obtained from the abattoir of the Faculty of Agriculture, Alexandria University, Alexandria, Egypt. The specimens were collected and examined macroscopically then used for the following studies.

2.1. Light microscopy

The specimens were fixed in 10% phosphate-buffered formaldehyde and processed for paraffin sectioning. Serial sections (4 µm) were prepared and stained using Mayer's hematoxylin and eosin (H&E) stain (Mayer, 1903), Gomori`s silver impregnation stain (Gomori, 1937). And Crossman trichrome stain (Crossmon, 1937).

2.2.Transmission Electron Microscopy:

Fresh five specimens, approximately 1 mm in size, were obtained from the region ileal Payer's patches (PPs) and fixed in 4F1G (McDowell EM, Trump F. 1976). After post-fixation in 1% solution of phosphate buffered osmium tetroxide at 48C, specimens were dehydrated in ascending grades of ethanol and embedded in Epon (Serva, Heidelberg, Germany) according to standard protocols. Semithin sections (1 mm) were prepared, stained with toluidine blue and examined by light microscopy. Suitable areas for electron microscopic examination were selected. From these areas ultrathin sections (50–70 nm) were cut by a glass knife and stained with urinyl acetate followed by lead citrate. The sections were observed with a Geol electron microscope (Japan) at 80 kV.

3. RESULTS

Macroscopical examination of the ileum did not reveal any prominent appearance of Peyer" patches. Microscopically the Payer's patches were located as a discontinuous patches in the submucosa of the anti-mesenteric side of the ileum. In general, the Payer's patches were a lymphoid aggregation ranged from single to several secondary lymphoid nodules of variable shapes and sizes (Figs.1&2).

The patches were supported by a reticular network of reticular fibers with clear germinal centers (Fig.3). The lymphoid nodules pulged into the mucosa forming dome area covered with short villus. The interfollicular zone that consisted of diffusely arranged lymphocytes (Fig.4).

The epithelium covering the domes of Peyer's Patches consisted of enterocytes without goblet cells (Fig.5). The epithelial cells covering the dome are more columnar and eosinophilic than other epithelial cells and lacks goblet cells. Transmission electron microscopic investigation revealed that the follicular associated epithelium includes M cells with numerous microfolds on the luminal surfaces with several enpocketing lymphocytes. (Fig.6). in the young ages the lymphoid nodules forming typical dome area (Fig,7) In the middle ages (from 1-3 years) the ileal Payer's patches were found in the submucosa (Fig.8) the lymphoid nodules became smaller and few in numbers in the old ages (Fig.9).

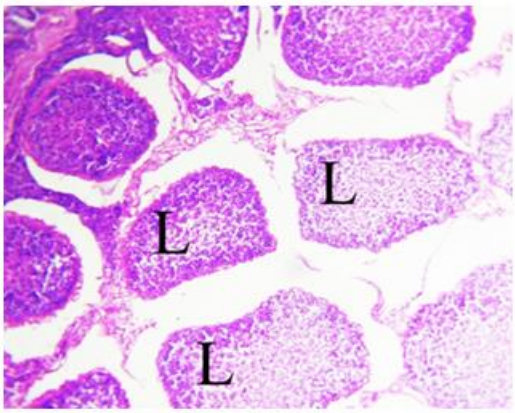

Fig. (1): Light photomicrograph of one-month buffalo ileum s howing submucosal aggregations. H&E stain.X 100.

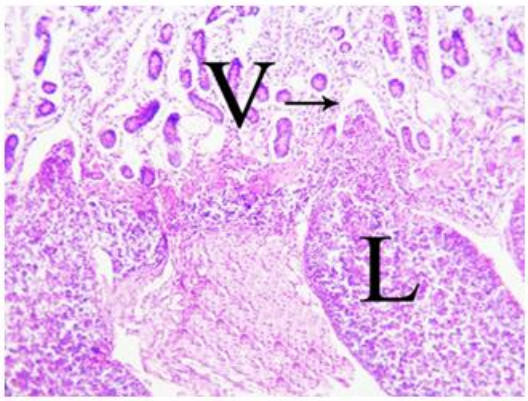

Fig. (2): Light photomicrograph of 2-month buffalo ileum showing PPs formed of elongated secondary lymphoid nodules(L), dome area (arrow) flanked with intestinal villi (V). H&E stain. X 100.

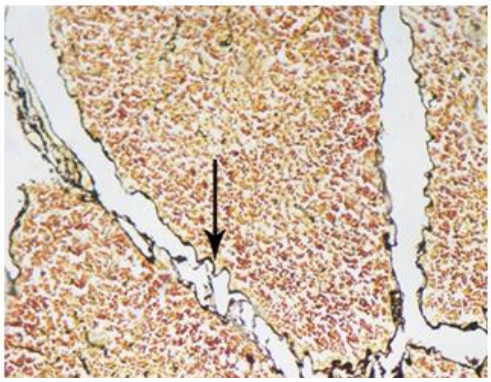

Fig. (3): Light Photomicrograph of buffalo calve ileum depecting elliptical lymphoid nodules supported by reticular fibers (arrow). Reticulin stain X400.

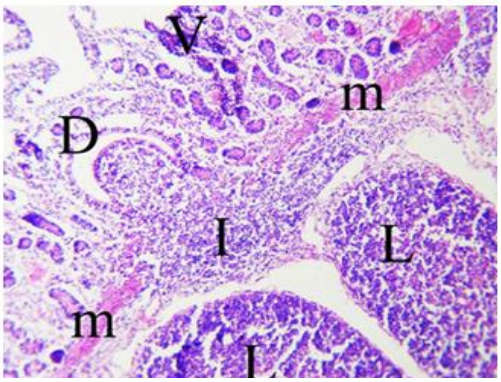

Fig. (4): Light Photomicrograph of buffalo calf ileum showing the interfollicular region (I) lymphoid nodules (L). Note the lymphoid nodul that form the dome region (D) in the ileal villi(V) and interrupted lamina muscularis (m) H&E X100

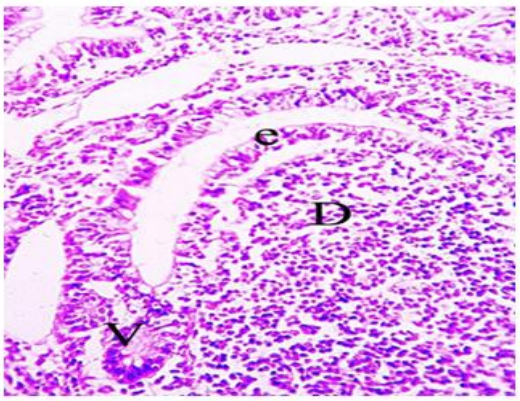

Fig. (5) Light photomicrograph of 6 years buffalo ileum showing Peyer"S patchs formed from lymphoid aggregation associated with short villi forming dome area (D), the associated epithelium (e) is highly infiltrated with lymphocytes forming lymphoepithelium,, intestinal villi (V)is lined with enterocytes and goblet cells.

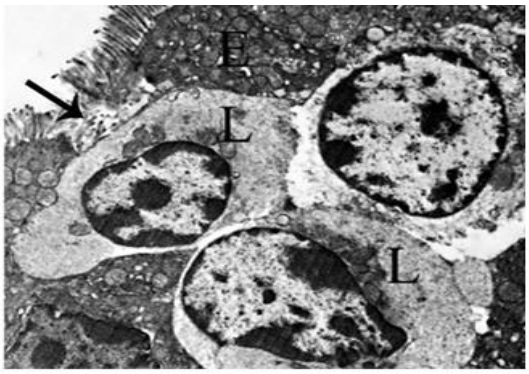

Fig. (6) Transmission Electrone micrograph of adult buffalo ileum showing 3 lymphocytes (L) with clear dark nucleus (N) that enpocketing M cell sometimes it reachs to the luminal border. Mag.X2500.

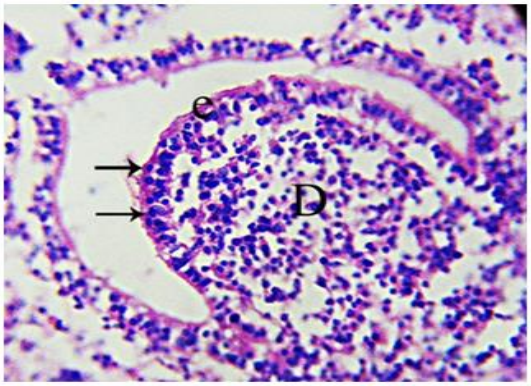

Fig. (7): Light photomicrograph of buffalo ileum showing migrating lymphoid nodule forming dome (D) that consists of lymphoid aggregation. Note the lymphocytic infiltration (arrows) through the associated epithelial cell (e)H&E stain. X 1000.

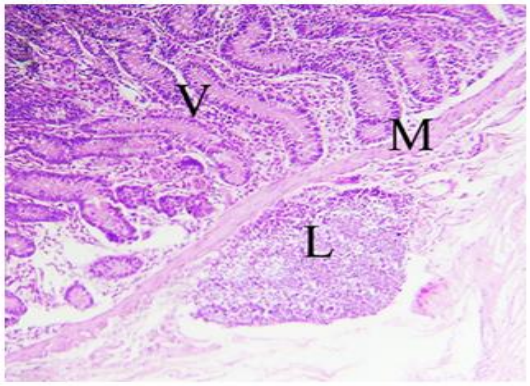

Fig. (8): Light photomicrograph of 2 years' buffalo ileum showing PPs formed of small lymphoid follicle (L) in the submucosa M- muscularis mucosae V - (intestinal villi).H&E stain. X 100

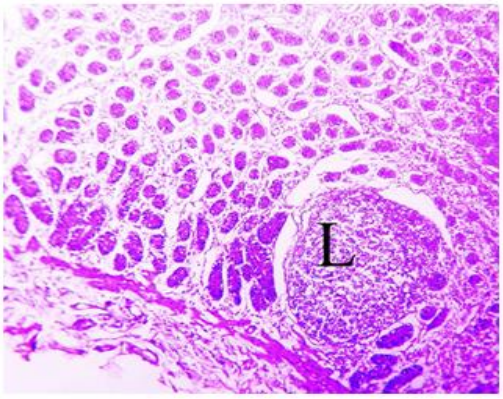

Fig. (9) Light photomicrograph of 8 years' buffalo showing PPs formed of small lymphoid nodules lymphoid nodules (L) H&E stain. X 100.

4. DISCUSSION

The present work showed that the ileal Peyer" patches of the water buffalo (*Bos bubalus*) were located in the anti-mesenteric side of the ileum as discontinuous patches and this disagree with the result of in agreement with (Kapoor and Singh, 2015) the Peyer`s patches formed from elongated, elliptical or spherical lymphoid nodules in the submucosa on the anti-mesenteric side in ilium of calves. In contrast to jejunal Peyer`s patches which were randomly distributed at mesenteric and anti-mesenteric regions (Hasanzadeh and Monazzah, 2011)

The submucosal lymphatic nodules of the Peyer`s patches were supported by a network of reticular fibers and follicular dendritic cells with clear germinal centers. (Jung et al. 2010) suggested that the arched appearance of the PP is due to the germinal centers forming the core of each follicle.

In agreement with Zidan and Pabst (2008) there were elevated regions overlying the lymphatic nodules forming a dome shape between the intestinal villi and looks like a short, broad villus. The epithelium covering the domes of Peyer's Patches consisted differed from the adjacent epithelium to the ordinary intestinal villi as it lacks goblet cells.

In agreement with Neutra et al., (1996) the basolateral side of M- cell a pocket-like invagination of the plasma membrane to house lymphocytes and antigen presenting cells. Similar finding was reported in the present work

Where the M cells of Peyer`s patches of buffalo play a role in the immunity as other species Kanaya et al., (2012) (Kraehenbubl and Neutra, 2000;

Neutra et al., 1996; Neutra et al., 2001), proved that, M cells have a high activity for phagocytosis and transcytosis, this functions make a fast transport of antigen into the underlying lymphoid tissues especially the antigen presenting cells. Then the antigen is presented to T cells which activate B cells generating immunoglobulin-A producing plasma cells. M cells mediated antigen transport is important for the initiation of mucosal immune responses,

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