Histomorphological Studies on Prenatal Development of Ileal Peyer’s Patches in Buffalo

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ABSTRACT

The histomorphological studies on prenatal development of Peyer’s patches in ileum of 20 buffalo foetuses ranging from 11.5 cm CVRL (80 days) to 100 cm CVRL (full term) were conducted. The foetuses were categorized into three groups based on their curved crown rump length (CVRL). No typical Peyer’s patches were observed in ileum from 11.5 cm CVRL (80 days) to 33 cm CVRL (148 days). However, at 54 cm CVRL (195 days), the first primordial lymphoid follicle and dome was observed. At 70 cm CVRL (231 days), a number of developing lymphoid follicles were encountered. The interfollicular spaces began to develop at this stage. At 80 cm CVRL (254 days), dome of the follicles was prominent with subepithelial dome region (SED) or corona. Germinal centre (GC) was first observed at this stage. At 100 cm CVRL (full term), multiple layers of completely developed follicles were present with eminent dome, sub epithelial dome, diffuse interfollicular tissue and high endothelial venules.

Key words: Buffalo, Histomorphology, Ileum, Peyer’s patch, Prenatal

Where Y is age in days and X is curved crown rump length (CVRL) in cm.

The foetii were divided into three groups based on their curved crown rump length (CVRL) viz; group I (CVRL between 0-20 cm, 0-118 days), group II (CVRL >20-40 cm, 119-163 days) and group III (CVRL >40 cm, 164-310 days). Tissue pieces of ileum were fixed in 10 per cent neutral buffered formalin for routine processing. Sections of 5-6 µ thickness were stained with haematoxylin and eosin for routine histology (Luna, 1968), Masson’s trichome for collagen fibres (Luna, 1968), Gridley’s for reticular fibres and Verhoeff’s for elastic fibres (Sheehan and Hrapchak, 1973).

RESULTS AND DISCUSSION

Typical Peyer’s patches (PP) were not observed from 11.5 cm CVRL (80 days) to 33 cm CVRL (148 days). However, aggregates of 2-3 lymphocytes arranged linearly in submucosa of ileum were observed (Fig. 1) at 33 cm CVRL (148 days). Nicander et al. (1991) observed small groups of lymphocytes in subepithelial tissue of ileum from 90 days of gestational age in sheep foetus. Among the lymphocytic accumulation, few lightly stained mesenchymal cells with processes formed a meshwork around the lymphocytes. Similar observations were made in the terminal
ileum of the foetal pig (Binns and Licence, 1985). At 35 cm CVRL (152 days), accumulations of darkly stained lymphocytes were observed at scattered locations and at 48 cm CVRL (181 days), the number of lymphocytes accumulations increased in size and were arranged in two layers in submucosa. Asari et al. (1987) reported the first primordial ileal PP in bovine foetus at about 5-6 months of gestational age. Elongated to oval lymphoid aggregates were observed at 54 cm CVRL (195 days) that formed primordia of lymphoid follicle, surrounding blood capillaries at most places. However, the first primordial developing dome like structure was also encountered with presence of darkly stained lymphocyte aggregates within the intestinal villi at this stage (Fig. 2) as reported by Beyaz and Asti (2004) in bovine foetuses.

With the increase in gestational age, at 70 cm CVRL (231 days), several layers of round to elliptical compact lymphoid follicles were observed on anti-mesenteric side and occupied 90% of the mucosal folds. The apex of few elliptical follicles in the upper layer protruded into epithelium by disintegrating lamina muscularis mucosae and formed dome (Fig. 3). Dome protruded in between the bases of villi and was surrounded by arcs formed by it. Similarly, Raju et al. (2012) also observed in sheep foetus at 4 months of foetal age that muscularis mucosae was not continuous throughout and were absent in the follicle having domes. Shukla and Singh (1996) made similar observations in dogs. Dome of the invading follicles were lined by a specialised developing lymphoepithelium or follicle associated epithelium that was more eosinophilic as compared to adjacent villi. The interfollicular spaces, however, began to develop at this stage with diffuse lymphoid tissue and numerous high endothelial venules (HEVs) (Fig. 3). Beyaz and Asti (2004) observed HEVs lying between adjacent lymphoid follicles in interfollicular area at 227 days of gestation in bovines. Thin collagen capsule from deep submucosa began to encapsulate the follicles from base towards apex.

At 80 cm CVRL (254 days) prominent dome was observed in follicles that invaded the epithelium and gave it an eroded appearance because of lack of villi over the dome (Fig. 4). Dome was pyramidal to conical in

Figs. 1-3. 1. Photomicrograph of ileum at 33 cm CVRL (148 days) showing group of lymphocytes (encircled) in submucosa (SM), tunica muscularis (TM) and villi (V). H. & E. × 400. [Inset: showing circled area as group of lymphocytes]; 2. Photomicrograph at the age of 54 cm CVRL (195 days) showing lymphoid aggregates (arrows) in submucosa (SM) around blood capillaries and tunica muscularis (TM). H. & E. × 100 [Inset: Showing primordial dome (D) with infiltration of lymphocytes (arrows) in villi at the age of 54cm CVRL in another section of ileum. H&E X400]; 3. Photomicrograph of 70 cm CVRL (231 days) buffalo foetus showing dome (D) of lymphoid follicles (LF) invading villi (V), disintegration of lamina muscularis mucosae (arrow), diffuse lymphoid tissue (yellow arrow) and high endothelial venules (H) in developing interfollicular space (dotted arrow). H. & E. × 100

Figs. 4-6. 4. Photomicrograph showing prominent dome (D) in between arcs of villi (V) and germinal centre (arrow) of lymphoid follicle (LF) in submucosa (SM) of buffalo foetus at 80cm CVRL (254 days). H. & E. × 100. [Inset: Showing enlarged view of dotted area as lymphoid follicle with prominent sub epithelial dome (SED) and germinal centre (GC)]; 5. Photomicrograph of 100cm CVRL (full term) buffalo foetus showing absence of capsule (arrow) towards apex in follicles having dome (D), thick capsule (Ca) around follicles towards serosal side, diffuse lymphoid tissue forming arcs (dotted arrow) and high endothelial venules (H) in wide interfollicular spaces. Masson’s trichome × 40. [Inset: enlarged view of dotted area showing presence of reticular fibres in diffuse lymphoid tissue, Gridley’s × 400]; 6. Photomicrograph showing dome (D) lined by follicle associated epithelium (FAE), group of lymphocytes (arrow) in few epithelial cells (M cells) of FAE in pockets, sub epithelial dome region (SED) and villi forming arc (V) in 100cm CVRL (full term) buffalo foetus.
appearance at apical part and follicle was oval to square below. The area just below the dome formed subepithelial dome region (SED) or corona, containing developing lymphocytes (small to medium-sized lymphocytes and few lymphoblasts), macrophages and follicular dendritic cells (FDC). Presence of lighter area in the centre that formed germinal centre (GC) was first observed at this stage (Fig. 4). Further, the differentiation into a light central zone and a darker periphery was observed in sheep foetus at 130-135 days of gestation (Nicander et al., 1991) and in bovine foetus at 271 days of gestation (Beyaz and Asti, 2004).

At 100 cm CVRL (full term), the follicles of different shapes (oval to elongated, elliptical) were arranged in multiple layers. The interfollicular spaces became wide, occupied by lymph vessels with some evidently visible lymphocytes and abundant HEV. Narrow, triangular diffused lymphoid tissue was present in interfollicular region and formed arches with the concavity facing towards upper layer of multiple lymphoid follicles (Fig. 5) as reported by Beyaz and Asti (2004) in bovine foetus, Lalitha (2000) in buffaloes. The diffuse lymphoid tissue and the centre of the follicles was composed of abundant reticular fibres that formed connective tissue framework for lymphoid cells. The capsule of developed follicles was highly vascularised. Apical portion of such follicles was devoid of collagen fibres capsule and lined by a thin layer only on the sides whereas follicles at the base were lined by thick capsule (Fig. 5). Nicander et al. (1991) found that continuous argyrophilic capsule was less distinct in the region of follicle neck around each follicle in sheep foetus by 130-135 days of gestation. The number of follicular domes protruding into the epithelium by disintegration of lamina muscularis mucosae increased with more evident SED region, outer dark cortex and lighter central area i.e., medulla or germinal centre at this stage. The dome was clearly identified by its prominent FAE containing a group of lymphocytes in pockets within it. Epithelial cells in FAE containing a group of lymphocytes were likely to be M-cells (Fig. 6).

Lymphocytic aggregations started at 152 days of gestation during the present study and at full term the fully differentiated follicles were observed.

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