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Ultrastructure of gut associated lymphoid tissues in Kadaknath fowl

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Lymphoid organs constitute the main structural category of the avian immune system. The secondary lymphoid organs, characterized by aggregates of lymphocytes and antigen presenting cells, are scattered throughout the body. Concomitant with the development of digestive structures and functions, a rapid development of the GALT occurs. The development of the bursa and other GALT starts during late embryogenesis (Kajiwara *et al.* 2003). Bar Shira *et al.* (2005) showed that development of GALT in the foregut was only slightly and temporarily impeded by feed withholding (for the first 72 h after hatch) in newly hatched broiler chicks.

The present study was aimed to investigate the ultrastructural observations on GALT in the Kadaknath fowl.

Birds were obtained from Instructional Poultry Farm (IPF) of the University. The birds were sacrificed by decapitation and tissue samples, viz. segments from duodenum, jejunum and ileum, were collected for transmission electron microscopy and fixed in 2.5% gluteraldehyde and processed for TEM as per the manual of AIIMS, New Delhi. Ultrathin (70-80 nm) sections were stained with uranyl acetate and lead citrate, according to Reynolds (1963) and examined in an electron microscope.

Lymphocytes were the main cell type seen aggregated beneath the epithelium of GALT either as germinal centres or as diffuse lymphoid tissue. The nuclear diameter profiles of these cells ranged from 1.5 to 5.2 mm. The small lymphocytes had a thin rim of relatively electron dense cytoplasm and more densely clumped chromatin than the large lymphocytes (Fig. 1). Chromatin was often seen along the inner membrane of the nucleus. A nucleolus was sometimes present. Few cytoplasmic organelles were seen in the small lymphocytes whereas the large lymphocytes had a few mitochondria, granular endoplasmic reticulum and occasionally a moderately developed Golgi Complex.

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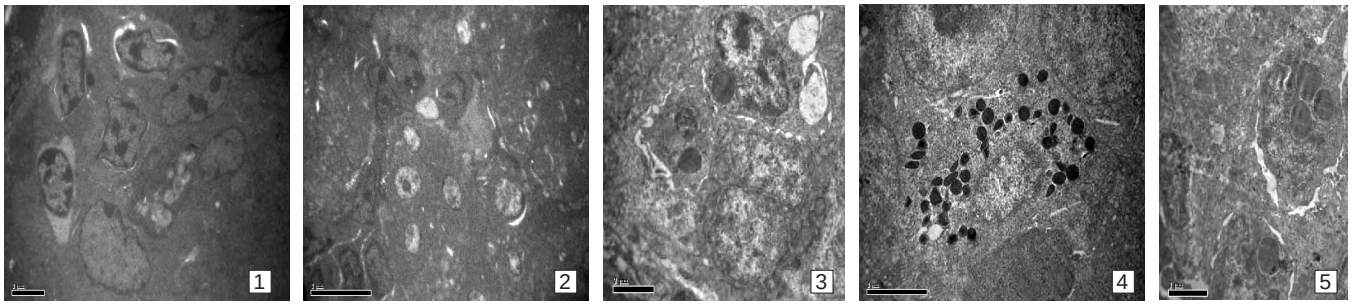
Lymphoblasts were common and could be distinguished by their large size, relatively large amount of cytoplasm, small amount of nuclear chromatin and by the presence of more cytoplasmic organelles than were seen in mature lymphocytes (Fig. 2). Vacuoles were observed sometimes in the cytoplasm of the lymphoblasts.

Plasma cells with dilated endoplasmic reticulum were seen among the lymphocytes and lymphoblasts (Fig. 3). It had the typical "clockface" arrangement of nuclear chromatin and was larger than other lymphoid cells. The division of the lymphoid tissue into germinal centres and diffuse lymphoid tissue (Befus *et al.* 1980, Burns 1982) was not always apparent at the ultrastructural level in fowl and nor was the presence of a dome area of lymphoid tissue (Abe and Ito 1978). However, the principle features of a well developed secondary lymphoid organ–small and large lymphocytes, plasma cells–macrophages and mast cells, were all present in Peyer's patches from fowls and turkeys (Burns and Maxwell1986).

Macrophages in moderate numbers were present throughout the lymphoid tissue. They often contained phagocytosed material; dead and dying cells could be seen within vacuoles in the cytoplasm. The nuclear membrane was often indented by phagocytosed material. A similar location for macrophages was reported in mammalian gut-associated lymphoid tissue (Crabb and Kelsall 1940, Abe and Ito1978, Lause and Backman 1981). Cunningham (1978) stated that macrophages are the main antigen-presenting cells in immune responses. Macrophages are considered to be the professional antigen presenting cells in generation of immune response (Kindt *et al.* 2007).

Mast cells were located in the lamina propria just beneath the epithelium, but never populated intestinal epithelium (Fig. 4). The granules were more varied in size and number than the globules of the globule leucocytes. The granules were moderately electron-dense and were bound by a single membrane (Fig. 4). The nuclei were usually regular in shape being either round or oval.

Globule leucocytes and lymphocytes were sandwiched between the epithelial cells throughout the intestine. Almost



Figs 1–5. Transmission electron microgrpah of GALT of the Kadaknath fowl showing : **1.** small (SL) and large lymphocyte (LL) × 10600; **2.** lymphoblast (LB) × 7960; **3.** plasma cell with dilated endoplasmic reticulum (Pc) × 10800; **4.** mast cell (MC), the granules of which are moderately electron dense (Gn) and are bound by a single membrane × 19900; **5.** globule leucocyte (GL). The granules (Gn) are generally homogenous and densely stained × 10600.

all the globule leucocytes (GL) were spherical and possessed thick cytoplasmic protrusions prolonged between the epithelial cells. The pattern of the chromatin and the size of the nucleus were similar to small or medium sized lymphocytes in the same area, but the cytoplasm of GL had a slightly lower electron density. Therefore, the cytoplasm was easily distinguished from that of the neighbouring epithelial cells. The globules varied in size and appearance within different leucocytes. They were generally homogenous and densely stained (Fig. 5) though sometimes vacuoles were seen within them. Gregory and Nolan (1981) recorded a direct association between globule leucocytes and lymphoid tissue (in Peyer's patches from lambs infected with coccidia) and also reported mast cells and globule leucocytes in Peyer's patches from uninfected control lambs.

SUMMARY

The present study conducted on 112 day-old Kadaknath fowl of both sexes revealed that the GALT were similar in many respects to those found in mammals. They were overlaid by a lymphoepithelium containing undifferentiated intestinal epithelial cells with a well developed microvillous border, lymphocytes and plasma cells. The organ was fully developed and the lymphoid aggregates of GALT comprised of small and large lymphocytes, lymphoblasts and plasma cells. Macrophages and globule leucocytes were seen among the lymphoid cells.

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