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By N. K. B. Raju, Geetha Ramesh, Sabiha Hayath Basha, S. Ushakumary
& R. Suresh kumar

Madras Veterinary College, India

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Histochemical Studies On the Peyer's Patches of Sheep (*Ovis aries*)

N. K. B. Raju^α, Geetha Ramesh^σ, Sabiha Hayath Basha^ρ, S. Ushakumary^ω & R. Suresh kumar[¥]

Abstract - Tissue pieces of jejunum and ileum from different prenatal and postnatal age groups of sheep were collected from Corporation slaughter house, Perambur, Chennai. The goblet cells in the villous epithelium were positive for Periodic acid Schiff (PAS) but the negative reaction was observed in the follicle-associated epithelium as the goblet cells were absent. The follicle-associated epithelium showed positive activity for the alkaline phosphatase. The activity was more intense over the follicle domes in all the prenatal and postnatal age groups. An intense acid phosphatase activity was observed in the follicle-associated epithelium and dome areas of Peyer's patches. A linear activity of the adenosine triphosphatase was observed in the capsule of the ileal follicles and a mild enzyme activity was noticed in the interfollicular region. A reticular pattern of 5'nucleotidase enzyme activity was observed in the follicles of ileal Peyer's patch.

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I. INTRODUCTION

The health of the digestive tract is important to receive the food material and for proper digestion and absorption. About 70 per cent of the body immune system is found in the digestive tract. This immune system often referred to as gut-associated lymphoid tissue and works to protect the body from invading pathogens (Ma et al., 2007). Peyer's patches are the aggregations of lymphatic nodules in the mucosa and submucosa of the jejunum and ileum of mice (Rowinski et al., 1984). Peyer's patches are immunocompetent lymphoid organs primarily engaged in immune responses to antigens presented from the intestinal lumen in guinea pig (Jurg et al., 1975).

The secretions from the loops containing Peyer's patches exhibit a stronger early Ig A response to bacteria than the secretions from loops lacking Peyer's patches. The large number of lymphocytes in Peyer's patches increases the probability of an antigen encountering an immunocompetent cell (Keren et al., 1978).

Author α : Assistant Professor, Dept. of Veterinary Anatomy, NTR. College of Veterinary Science, Gannavaram – 521 102, Krishna (Dt), Andhra Pradesh, India. E-mail : nkbraju@gmail.com

Author σ : Professor and Head.

Author ρ : Associate Professor, Dept. of Veterinary Anatomy, Madras Veterinary College, Chennai – 600 007, India.

Author ω : Associate Professor, Dept. of Veterinary Anatomy, Madras Veterinary College, Chennai – 600 007, India.

Author ¥ : Professor and Head, Dept. of Veterinary Surgery and Radiology, Madras Veterinary College, Chennai – 600 007, India.

A thorough knowledge of the histological changes in gut-associated lymphoid tissue (GALT) is very essential to gain a comprehensive knowledge on the gut immunology and to form a basis for the interpretation of various pathological conditions of the gut. Hence, the present work has been undertaken to explore the histochemistry of the GALT in sheep.

II. MATERIALS AND METHODS

Tissue pieces from the terminal part of jejunum and parts of ileum were collected from sheep. The tissues from six animals each from different age groups viz. three months, four months and five months in prenatal and neonatal (0-2 months), young (3-9 months) and adult (10 months- 2 years) in postnatal groups were procured from the Corporation slaughter house, Perambur, Chennai. The determination of age ascertained as described by Richardson et al. (1976) in prenatal and Noden and de Lahunta (1985) in postnatal age groups.

The routine paraffin sections of 3-5μm thickness were used for carbohydrates. Frozen sections of tissues fixed in chilled formol calcium (4°C) were used for localization of alkaline phosphatase (Singh and Sulochana, 1996), acid phosphatase (Singh and Sulochana, 1996) and 5'Nucleotidase (Bancroft and Gamble, 2003). Frozen sections of fresh unfixed tissues were also used for localization of adenosine triphosphatase (Bancroft and Gamble, 2003). All the frozen sections were cut at 15- 20μm thickness by cryostat.

III. RESULTS AND DISCUSSION

a) Carbohydrates

The goblet cells of the villous epithelium were positive for PAS reaction (Fig.1). But, these cells were absent in the follicle-associated epithelium of the ileal Peyer's patches in all the age groups of sheep which is in accordance with Befus et al. (1980) in chicken.

When the combined PAS and alcian blue technique was applied, a blue reaction was observed in the follicle-associated epithelium of the ileal Peyer's patches. However, Burns (1982) observed that when the alcian blue was followed by PAS a purple reaction was observed in the epithelium of Peyer's patches of the domestic fowl. Laliitha (1991) found that, the PAS positive mast cells were observed in the peripheral

cortex of the lymphatic nodule of Peyer's patches in buffaloes.

b) Enzymes

i. Alkaline phosphatase

The follicle-associated epithelium showed positive activity for the alkaline phosphatase. The activity was more intense over the follicle domes (Fig.2) which is in agreement with Landsverk (1981) in calves, Bjerknes and Cheng (1981) in rat, Burns (1982) in domestic fowl and Owen and Bhalla (1983) in rat. However, Schmedtje (1965) noted that, the lymphoid follicle domes in rabbit appendix revealed negligible alkaline phosphatase activity over the cytoplasmic band separating intraepithelial lymphocytes from the lumen. Nordstrom et al. (1968), Weiser (1973) reported that the alkaline phosphatase activity increased as maturing epithelial cells migrated upto the villi in rats and human foetuses respectively. Ropke et al. (1972) noted in mouse that the high endothelial postcapillary venules through which lymphocytes entered the lymph nodes and Peyer's patches expressed a weak or no reaction for alkaline phosphatase in contrast to other endothelia. Ono (1975) recorded the alkaline phosphatase activity in tubules and vacuoles of enterocytes overlying the follicle in neonatal rats but not in adult animals. Further, Halleraker et al. (1990) observed in ruminant that, the alkaline phosphatase enzyme activity was shown in the follicle capsule.

A reticular pattern of staining was found in the centre of the follicle, interfollicular area and in the corona. A weak reaction was seen in the dome. Muscularis mucosae was positive for the enzyme. In lamb, the reticular reaction in the dome of the ileal Peyer's patches was absent. Nicander et al. (1991) reported that in sheep and goat foetuses by 90 days of gestation, a positive band of alkaline phosphatase activity was seen at the base of the villi in the follicular area. Weaker staining was observed in stromal elements of subepithelial tissues in both villi and primordial domes.

ii. Acid phosphatase

An intense enzyme activity was observed in the follicle associated epithelium and dome areas of ileal Peyer's patches (Fig.3) which is in accordance with Owen et al. (1986) in rat. Halleraker et al. (1990) stated that the cells stained for acid phosphatase were interpreted as macrophages in the follicle and dome in ruminants.

A strong activity in the interfollicular region and a mild activity of the enzyme in the capsule was observed in the ileal Peyer's patches. A pattern of enzyme activity was noticed in the follicles. However, Halleraker et al. (1990) stated that, the smooth muscle cells and reticular cells of the interfollicular tissue reacted strongly. A linear reaction outlined the follicle capsule.

iii. Adenosine triphosphatase

A linear activity of the enzyme was observed in the capsule of the ileal follicle (Fig.4). A mild enzyme activity was noticed in the interfollicular region and a weak positive reaction for the enzyme was observed in the follicle. However, Nicander et al. (1991) noticed a stronger activity of enzyme within the follicle and more pronounced activity within the centre of the follicle by 128 days in sheep foetuses. Halleraker et al. (1990) found that in lambs, the reticular cells stained weakly for this enzyme in the T-cell area and in the centre of the follicle. The follicle capsule had a weak, linear and discontinuous staining.

iv. 5'nucleotidase

A reticular pattern of enzyme activity was observed in the follicles of ileal Peyer's patches (Fig.5). A linear activity of the enzyme was outlined in the capsule of the follicle which is in accordance with Halleraker et al. (1990) in the follicles of jejunal Peyer's patch in ruminants. Nicander et al. (1991) also observed a positive reaction of the enzyme in the capsule by 128 days in sheep foetuses.

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LEGENDS TO FIGURES

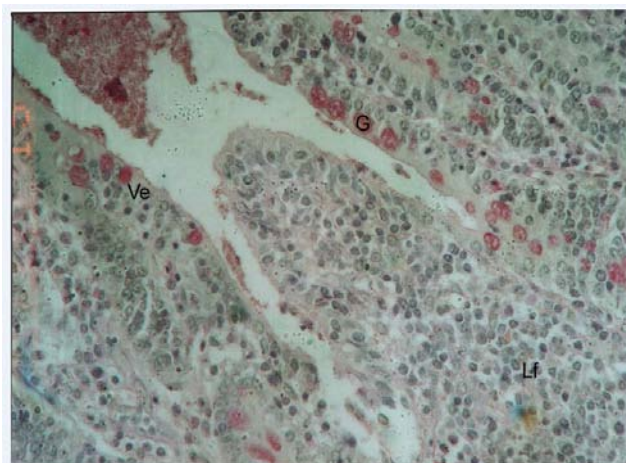


Fig. 1 : Photomicrograph of ileal Peyer's patch of two month-old sheep showing PAS negative reaction in lymphoid follicles and positive reaction in goblet cells of the villous epithelium.

Lf- Lymphoid follicle

G- Goblet cells

Ve- Villous epithelium

PAS x 400

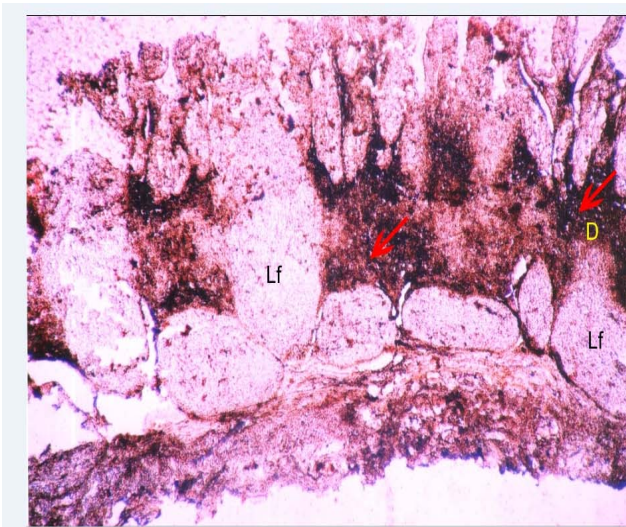


Fig. 2 : Photomicrograph of ileal Peyer's patch of one year-old sheep showing positive alkaline phosphatase activity (arrows) in the domes.

Lf- Lymphoid follicle

D- Dome

Alkaline phosphatase x 40

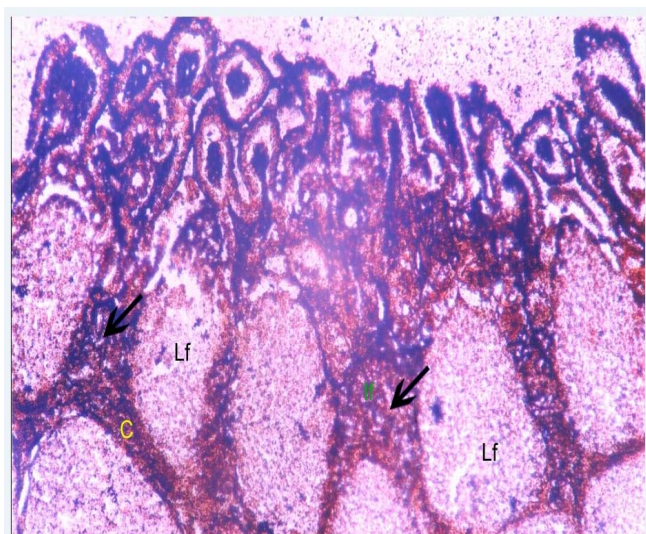


Fig.3 : Photomicrograph of ileal Peyer's patch of five month-old foetus of sheep showing an intense acid phosphatase activity (arrows) in the internodular and a linear activity in the capsule.

Lf- Lymphoid follicle

C- Capsule

If- Interfollicular area

Acid phosphatase x 40

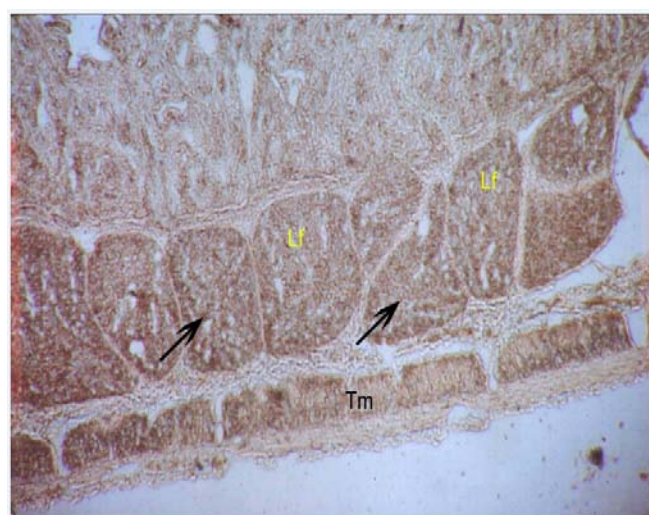


Fig.5 : Photomicrograph of ileal Peyer's patch of eighteen month-old sheep showing a pattern of 5'nucleotidase activity (arrows) in the follicles.

Lf- Lymphoid follicle

Tm- Tunica muscularis

5'nucleotidase x 40



Fig.4 : Photomicrograph of ileal Peyer's patch of six month-old sheep showing a linear activity of the adenosine triphosphatase (arrows) in the capsule and a mild activity in the interfollicular region.

C- Capsule

If- Interfollicular area

Adenosine triphosphatase x 40