EPITHELIAL DEVELOPMENT IN THE FORESTOMACH OF PASTURE-FED LAMBS (BIRTH TO 8 WEEKS)

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ABSTRACT

The histology, mitotic index, ultrastructure and Na^+-K^+-ATPase cytochemistry of lambs reared on pasture were studied during the period of weaning.

Two embalmed mature non-pregnant Romney cross-bred ewes and thirty Romney-cross lambs reared on Massey University sheep farms pastures were used. Five lambs (three in 1981 and two in 1982) were taken from their dams on pasture at each of the following respective ages: within 24 hours of birth, and at 12, 23, 34, 45 and 56 days. Stomach-tissue samples from 1 adult and from the lambs reared during the 1981 lambing season were prepared for histology using Haematoxylin and Eosin, Masson's green trichrome, Periodic-acid-Schiff and Toluidine Blue stains, and for conventional transmission electron microscopy. Tissue samples from the rumens of lambs reared during the 1982 season were used for strontium-capture technique Na^+-K^+-ATPase cytochemistry.

Gross dissection of the stomach of one-day-old lambs confirmed that the largest compartment at birth is the abomasum, followed, in decreasing order of size, by the rumen, reticulum and omasum. Progressive development resulted in the forestomach compartments assuming their adult proportions by 56 days of age.

Preliminary histological studies of epithelium taken from the rumen, reticulum, omasum and reticular groove of the adult sheep confirmed it to be a stratified keratinizing epithelium. Five general cell layers were clearly seen: stratum basale, stratum spinosum, stratum granulosum, stratum transitionale and stratum corneum. (In previous studies, the stratum granulosum and the stratum transitionale have been considered as one layer.) Mucopolysaccharides were located in the intercellular spaces in the stratum corneum.

Examination of the forestomach mucosa revealed a number of changes between birth and 56 days of age: (1) an increase in papillary length; (2) starting from 23 days, the development of extensive papillary process-epithelial bulb interactions accompanied by proliferation of blood vessels in the papillary processes; (3) a decrease in epithelial thickness for the first 45 days of age; (4) the appearance of a complete layer of transitional cells by 45 days; (5) the disappearance of glycogen from the epithelium; (6) an increase in the amount of mucopolysaccharide in the intercellular spaces in the stratum corneum; (7) the increase in the number of non-keratinocytes in the basal layer; and (8) the appearance of apoptotic bodies due to single cell death in the basal layer of the epithelium from 45 days of age.

The mitotic index of the epithelium in developing lambs decreased from birth until 23 days of age, had increased at 34 days, but decreased again between 45 and 56 days.

Examination of the ultrastructure of the adult epithelium provided general results consistent with previous studies. However, gap junctions were found in the stratum basale, stratum spinosum and were extensive in the stratum granulosum. Tight junctions (zonulae occludentes) were seen between the cells of the stratum corneum.

Langerhans cells and mast cells/globule leukocytes were classified as non-keratinizing cells. Other non-keratinocytes recognised were the 'indeterminate cells', lymphocyte-like cells and cells similar to Merkel cells.

Completely keratinized cells appeared in the epithelium at about 12 days of age, in association with the increased production of tonofilaments, keratohyalin granules, endoplasmic reticulum protein and membrane-coating granules. Proliferation of mitochondria in the basal layers started at about 12 days of age, and glycogen deposits in the intermediate layers were not found after this age.

Increased folding of the basolateral membrane surfaces of basal cells and progressive thinning of the endothelium of sub-epithelial blood vessels were also observed. Gap junctions in the stratum granulosum became progressively more obvious between birth and 56 days of age. Annular gap junctions were also found.

Na+-K+-ATPase enzymatic sites were identified from 12 days of age, on the cytoplasmic membranes of lower granular, spinous and basal cells: the reaction products being localised on both the cytoplasmic and inter-cellular-space surfaces of the plasma membranes. Ouabain inhibited the formation of deposits only on the cytoplasmic side. Alkaline phosphatase activity was localised in the stratum corneum. Mg++-ATPase was demonstrated in the stratum corneum, the intercellular spaces in the stratum granulosum and stratum transitionale, and in mitochondria.

It is concluded that the ultrastructural features and Na+-K+-ATPase cytochemistry of the epithelium at 12 days of age appeared to be similar to those found in older animals. However, structural (and presumably functional) maturity did not appear to be complete until after 45 days of age at which stage the stratum transitionale had become complete and the mitotic index and the thickness of the epithelium had become stable. The increase of non-keratinocytes suggests the increasing immunocompetence of the epithelium.

Tight junctions and extruded contents of membrane-coating granules in the intercellular spaces of the stratum corneum could provide a barrier to the diffusion of solutes across the epithelium. The development of gap junctions and the presence of Na+-K+-ATPase enzymatic sites in the membranes are consistent with the absorptive and transport functions of the epithelium, particularly the active transepithelial movement of sodium ions. Future studies could well show hormones, hormone-like substances and antibiotics to be important in the development of the forestomach epithelium in ruminants.

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