

Regional Histological Variations of the Nasal Mucosa of Cattle

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SUMMARY

Aspects of the histology of the nasal mucosa of calves from four to six months of age have been described.

The functional significance of the type of epithelium, its thickness, the degree of neutrophil invasion, and its relationship to the numbers of *Pasteurella haemolytica* in the nasal cavity have been discussed. In addition, the arrangement and depth of glands in the lamina propria and the presence of lymph follicles have been described.

RÉSUMÉ

Description des particularités histologiques de la muqueuse nasale de veaux âgés de quatre à six mois.

Signification fonctionnelle du type d'épithélium, de son épaisseur ainsi que du degré d'infiltration neutrophilique et de sa relation avec le nombre de *Pasteurella haemolytica* observées dans la cavité nasale. Description de l'agencement en profondeur des glandes dans le chorion ainsi que la présence de follicules lymphoïdes.

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INTRODUCTION

During the course of a study on the distribution of *Pasteurella haemolytica* in the nasal cavity of cattle (8), nasal tissue was obtained for routine histological study. Although the general histology of the bovine nasal cavity has been described together with that of other domestic animals (12), a detailed account pertaining specifically to cattle was not found. Bovine respiratory diseases are of great economic importance to the cattle industry in North America (5) and since the upper respiratory tract is usually involved in these diseases (2), it was considered worthwhile to present the histological observations made on the bovine nasal mucosa.

MATERIALS AND METHODS

Fifteen beef calves, four to six months of age, were purchased on arrival by train from Western Canada and were handled similarly to that described by Thomson *et al* (11).

Sixteen sides of the nasal cavities of these animals were used for the histological study. Small pieces of tissue, approximately 5 x 5 mm, from 15 areas (Fig. 1) of each nasal cavity were fixed in 10% buffered formalin, processed and embedded in paraffin and sectioned at six microns. Tissues were stained with haematoxylin and eosin (H & E).

Various aspects of the epithelium and lamina propria were examined. The study was conducted according to a randomized complete block design. The 16 nasal cavities served as the replications and the 15 areas constituted the treatments.

The epithelium was designated as either respiratory or stratified squamous type. A respiratory type epithelium is one in which there is pseudostratification of cells in addition to goblet cells and/or ciliated cells. Stratified squamous epithelium is composed of successive layers of cells which are cuboidal in the lower layers and assume a flattened shape towards the surface. Goblet cells and ciliated cells are not present in stratified squamous epithelium.

The epithelium was recorded as folded if

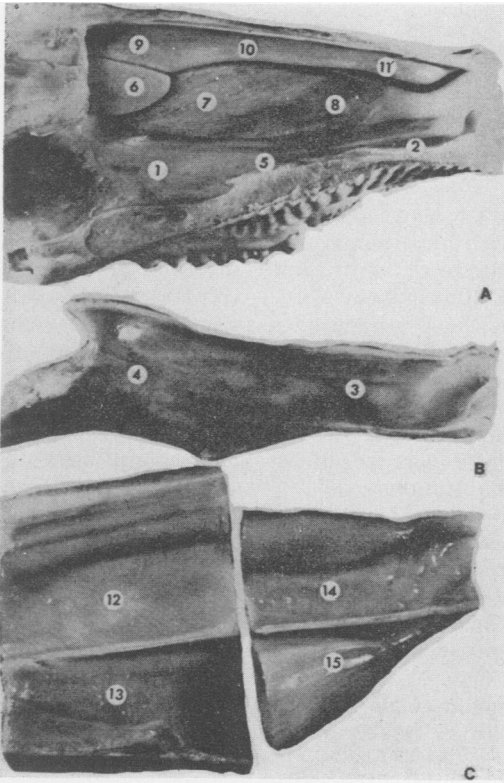


Fig. 1. Fifteen areas of the nasal cavity selected for detailed examination. A square piece of tissue from the areas indicated was sampled.

- A. One half of the bovine nasal cavity with the septum removed.
- B. Nasal septum.
- C. Exposure of areas 12 to 15 by removal of the ventral turbinate. The anterior edge of the lateral wall is to the left of the picture. The ventral turbinate was removed by cutting along the bony ridge (midway between 12 and 13). It was then laid out so that the anterior edge of the ventral turbinate is to the right of the picture (i.e. — analogous to an open book). The dorsal scroll of the ventral turbinate has been partly unrolled.

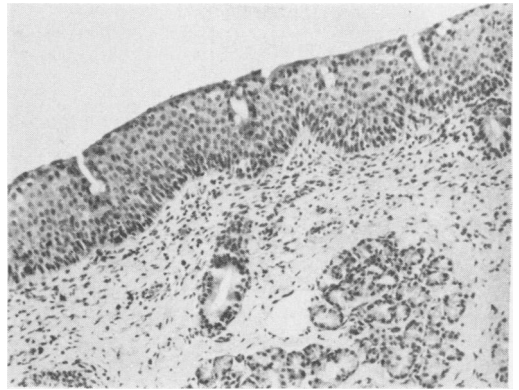


Fig. 2. Nasal mucosa from area 3 (Figure 1) to demonstrate folding of the base of the epithelial layer.

the basal cell-lamina propria junction displayed an undulating appearance (Fig. 2). The mean value of the thickness (microns) of three random fields observed at 100 times magnification was used as the measurement of the thickness of the epithelium of each area of each nasal cavity examined. The measurement was made to the nearest ten microns. The goblet cells were counted and were recorded as Low, for 1 to 20, and High, for greater than 20 cells, per section.

The number of neutrophils present in three power (400X) fields of the epithelium and lamina propria was counted and recorded as +, ++, +++. The designations corresponded to 1 to 25, 26 to 100, and greater than 100 cells respectively. The number of neutrophils was compared with the number of *P. haemolytica* present in an area (8). The presence of other bacteria were noted but the number was not compared with the number of neutrophils.

The number and mean of the greater diameter (microns) of lymph follicles per section were recorded. The mucous and serous glands were designated as having a "regular" arrangement if the groups of glands were aligned in a regular array below the epithelium. If groups of glands did not have this arrangement, they were designated "scattered". The distance from the basal layer of the epithelium to the deepest extremity of glandular tissue was recorded as the Gland Depth. The distance was measured to the nearest ten microns.

The analysis of the data consisted of analysis of variance and Duncan's Multiple Range Test (6) employed for testing the significance of the mean differences of the thickness of the epithelium and the depth of glands. The five per cent probability level was used for all tests of significance.

RESULTS

The majority of sections examined had a respiratory type epithelium (Fig. 3). Four sections of area 11, two sections of area 2, and one section of area 3 had a stratified squamous epithelium. Areas 2 and 3, particularly the latter, displayed folding most prominently.

Although there was little variation in the mean thickness of the epithelium between the 15 areas (Table I), there was a significant difference in some cases (Table II). Apart from area 4, the areas with the thickest epithelium were situated at the anterior end of the nasal cavity.

The variation between areas in frequency of High and Low numbers of goblet cells, represented as a fraction of the total number of sections examined per area, was marked (Fig. 4).

In most of the sections examined, neutrophils were present between epithelial cells. The numbers varied greatly. In the majority of sections, single neutrophils were present between cells of the epithelium but in several sections there was a tendency for the neutrophils to be clumped. A ++ and

TABLE I. The Mean (microns) and One Standard Deviation of the Thickness of Epithelium and Depth of Glands in 15 Areas of 16 Nasal Cavities

Area	Thickness of Epithelium	Depth of Glands	
		Log 10	(Antilog)
1	63	2.9583	(908)
2	76	3.2451	(1758)
3	81	3.0269	(1064)
4	72	2.6831	(482)
5	54	2.7752	(596)
6	64	— ^a	
7	55	2.5349	(343)
8	58	2.6466	(443)
9	58	2.6160	(413)
10	56	2.8064	(640)
11	68	2.9681	(929)
12	66	2.6756	(474)
13	65	2.6535	(450)
14	59	2.4812	(303)
15	65	2.5661	(368)

Standard Deviation	11	0.1558
Error DF ^b	210	195

^aOwing to the inadequate sample size of this area it was deleted from the calculations

^bDF — degrees of freedom

+++ neutrophil count was often associated with hyperaemia of the lamina propria. The degree of neutrophilic invasion of the epithelium appeared to bear little relationship to the presence of a surface exudate seen microscopically. In general, the number of neutrophils present in the lamina propria appeared to bear no relationship to the numbers present in the epithelium. Similarly, there was no direct relationship between the numbers of *P. haemolytica* and the number of neutrophils. *Pasteurella multocida* and/or beta haemolytic streptococci were present in many areas. The exact number was not determined and the relationship to the number of neutrophils present was not determined.

Small focal areas of degeneration of epithelial cells, manifest as small vesicles containing neutrophils, were present in a few sections. They were associated with a ++ or +++ neutrophil count.

Lymph follicles were consistently found in area 1. On the average there were five per section, the mean of their greater diameter being 350 microns. Occasionally a few smaller follicles were found in other areas but they were not found on the dorsal turbinate (areas 9, 10, and 11), the anterior medial wall of the ventral turbinate (area 8) or in the lamina propria of the ventral scroll of the ventral turbinate (area

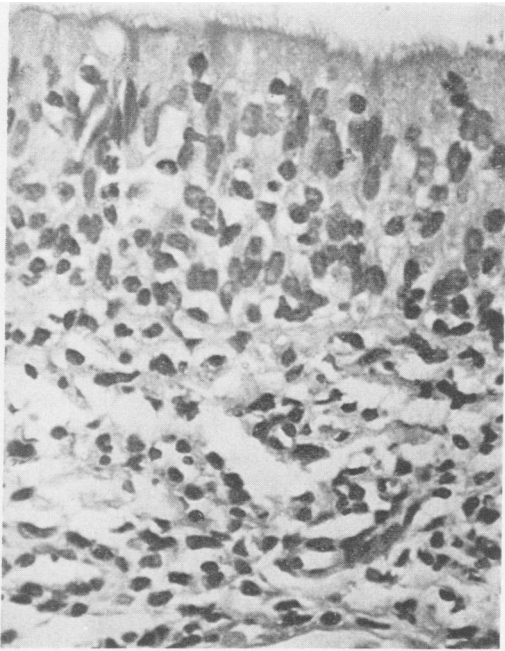


Fig. 3. Nasal mucosa from area 6 to demonstrate the typical appearance of the respiratory mucosa of the bovine nasal cavity.

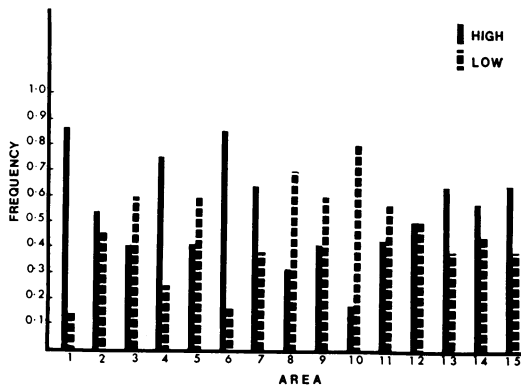


Fig. 4. Frequency of occurrence of High and Low groups of goblet cell numbers per area of nasal cavity expressed as a fraction of the total number of sections examined.

15). Lymphocytes, plasma cells, and other mononuclear cells were present in the lamina propria in all areas (Fig. 3). In most cases, apart from those areas in which lymph follicles were present, these cells were diffusely arranged; however, in some cases there was a focal arrangement. The number of cells was variable and appeared to bear no relationship to the degree of neutrophilic infiltration of the tissue.

A scattered arrangement of glands was present consistently in area 2. Areas 4 to 15 presented a regular arrangement; in areas 1 and 3 regular and scattered arrangements occurred with about equal frequency.

There was marked variation in the mean depth of mucous and serous glands in areas 1 to 15 of the 16 nasal cavities examined (Table I).

There was no significant difference in the depth of glands between nasal cavities

but there was a significant difference between areas within nasal cavities. The means of the depth of glands that were significantly different are represented in Table III.

DISCUSSION

In the majority of sections examined the epithelium was of a respiratory type apart from seven sections from the anterior regions (areas 2, 3, and 11) in which a non-cornified, stratified, squamous epithelium was present. These findings are consistent with those described for man and domestic animals in standard texts on the subject (3, 10, 12).

In this study the mean thickness of the epithelium between some areas within the nasal cavity differs significantly. The thickness of the epithelium is related to the paths of the major airstreams (1). The airstreams in the domestic animals examined curved downward after entering the nares and flowed along the ventral regions of the nasal cavity. The paths of the major airstreams in cattle have not been described. However, if the same principles apply to cattle as they do to other animals, the rate of mucous flow can be roughly correlated with the paths of principal air currents (1). Lucas and Douglas (7) have described the mucous flow rate in cattle, the slowest flow being over the anterior third of the medial aspect of the dorsal and ventral turbinates, along the floor and over the opposing surface of the nasal septum.

TABLE II. Means of Thickness of Epithelium (microns) of the 15 Areas of 16 Nasal Cavities Examined

Areas of the Nasal Cavity ^a	3	2	4	11	12	13	15	6	1	14	8	9	10	7	5
Means	81	76	72	68	66	65	65	64	63	59	58	58	56	55	54

^aRanked in descending order of thickness of epithelium. The means that are not significantly different from each other are under-scored, i.e. the thickness of the epithelium of area 3 is not significantly different from that of area 2, but it is from that of the remaining 13 areas.

TABLE III. Means of Depth of Glands (microns) of 14 Areas of 16 Nasal Cavities Examined

Areas of the Nasal Cavity ^a	2	3	11	1	10	5	4	12	13	8	9	15	7	14
Mean Log 10	3.2451	3.0269	2.9681	2.9583	2.8064	2.7752	2.6831	2.6756	2.6535	2.6466	2.6160	2.5661	2.5349	2.4812
Antilog (microns)	1758	1064	929	908	640	596	482	474	450	443	413	368	343	303

^aRanked in descending order of depth of glands. The means that are not significantly different from each other are under-scored, i.e. area 2 is significantly different from all other areas, areas 3, 11, and 1 are not significantly different from each other but they are from all other areas, etc.

Therefore, we would expect the epithelium to be thickest in areas 1, 2, 3, 5, 8, 11, and possibly 4 (Fig. 1). Apart from areas 1, 5, and 8 (Table 11), this relationship appears to hold. The obvious discrepancy for area 5 cannot be explained.

The functional significance of folding of the base of epithelium, as seen in areas 2 and 3, is not understood.

The relative number of goblet cells in the epithelium was variable throughout the nasal cavity in this study and agrees only in part with those of Hilding (4) and Lucas and Douglas (7), who found greater numbers of goblet cells in the epithelium of the roof of the nasal cavity. They related this to the lack of major air currents in these regions.

The mucous and serous glands in the lamina propria of areas 1, 2, 3, and 11 reached a greater depth than those in other areas and this corresponded with macroscopic observations on the thickness of the mucosa observed during processing; i.e., deep glands are found in thick mucosa. Apart from area 11, these areas (1, 2, and 3) were the areas in which a scattered arrangement of glands were found. The significance of this, if any, is not clear. The depth of the entire mucosal layer and its glands, as well as the depth of the epithelium, may be related to the presence of contaminating micro-organisms and/or particulate matter. A remarkable reduction in overall mucosal depth and a reduction in gland depth in all areas of the nasal cavity has been demonstrated in chickens reared in a germfree environment (1).

Lymphocytes, plasma cells, and other mononuclear cells were found in the lamina propria of all areas but lymph follicles were only found consistently in the nasopharynx (area 1) and occasionally in the anterior regions (areas 2, 3, and 11). Proetz (9) stated that the occurrence of lymph follicles in the nasal cavity coincides with the areas of maximal accumulation of particulate matter. As the mucus from most areas of the nasal cavity eventually passes the nasopharynx (7) one would expect to find lymph follicles in this area (area 1).

The presence of lymphocytes and plasma cells in the lamina propria is similar to that seen in other exposed mucosal surfaces which are subject to continual antigenic stimulation.

A direct relationship between the numbers of *P. haemolytica* present and the intensity of neutrophilic invasion of the epi-

thelium was not demonstrated. The sections in which a ++ or +++ neutrophil count was associated with hyperaemia could be interpreted histopathologically as being inflamed, but the cause of the inflammation was not determined. It appeared that if *P. haemolytica* was involved, it was so only in part. The presence of other bacteria such as *P. multocida* and beta-haemolytic streptococci may have been associated with the reaction but attempts at correlation were not performed.

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