

GROSS ANATOMY, HISTOLOGY AND SCANNING ELECTRON MICROSCOPY OF THE NASAL COMMISSURES AND ALAE IN YOUNG PIGS

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ABSTRACT

The present study was conducted on 18 young pigs of 8-10 months age of local mixed breed to determine the anatomical features of nasal commissures and alae. The gross anatomical studies revealed that external nares were round to oval in shape and bounded by dorsal and ventral commissures and medial and lateral alae. The histological studies revealed that the commissures and alae were lined by stratified squamous keratinized epithelium. The dermis had dense irregular connective tissue mainly comprising of collagen, reticular and few elastic fibers, small sized blood vessels, fine blood capillaries and connective tissue cells. Some of the hair follicles were surrounded by blood vessels which were considered as sinus hair. The scanning electron microscopy presented that surface of snout was having flat epithelial cells with different patterns of microplacae observed at higher magnification. Large sized hair erupted from a trough like structure and their surface was smooth.

Key words: Alae, histology, nasal commissures, pig, scanning electron microscopy

The nasal cavity, being initial segment of the respiratory tract, is more prone to various pathological conditions. The nasal cavity plays an important role in preparation of the inspired air, olfaction, phonation, thermoregulation of body and removal of foreign dust particles (Mygind, 1978). There is paucity of literature on light and scanning electron microscopy of the nasal commissures and alae in pig except some work has been carried out in goat (Kumar *et al.*, 1993), camel (Suman *et al.*, 1998) and sheep (Ganganaik *et al.*, 2004 a,b). The present study was contemplated keeping in view the importance of nasal commissures and alae in pigs.

MATERIALS AND METHODS

The present study was conducted on 18 young male pigs of 8-10 months age, of local mixed breed. The heads were procured from local slaughter house immediately after decapitation and fixed in 10% neutral buffered formalin solution for 48 h. Six heads were used to study the gross anatomy of nasal cavity. The tissues for histomorphological and histochemical studies were collected from six heads. The tissues were collected from dorsal and ventral commissures; medial and lateral alae of snout. The fixed tissues were processed for routine paraffin technique of light microscopy. The paraffin sections of 5-6 μ were cut and stained by routine Harris² hematoxylin and eosin method, Gomori's method, Weigert's method, Ayoub Skhlar method McManus² PAS method (Luna, 1968) and Crossman's trichrome stain (Crossman, 1937).

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Fresh tissues from six pigs heads were processed for scanning electron microscopy. The tissues were fixed in 2% glutaraldehyde solution for 6-8 h after thorough washing in chilled 0.1 M phosphate buffer (pH 7.4). The tissues were again washed twice with 0.1M phosphate buffer and rest of the procedure was carried out at EM Lab., AIRF, Jawahar Lal Nehru University, New Delhi. The tissues after sputter coating with gold were viewed under scanning electron microscope (Zeiss EVO-40).

RESULTS AND DISCUSSION

Gross Anatomy: The external nares (snout) in pig were round to oval in shape (Fig. 1) whereas these were slit like in sheep (Ganganaik *et al.*, 2004a), and comma shaped in ox (Hare, 1975), buffalo (Dhingra and Kumar, 1978) and dog (Hare, 1975). The nostrils were bound by two alae (medial and lateral) and two commissures (dorsal and ventral). The medial ala was formed by the fold of skin on the medial side of the nostril, whereas the lateral ala was formed by the fold of skin on the lateral side of the nostril. The medial and lateral alae met at the upper side of the nostril to form the dorsal commissure, whereas the ventral commissure was formed by the union of medial and lateral alae on lower side of the nostril. The rostral part of the nasal septum was modified to form the os rostrale (Fig. 1). Os-rostrale in pig was a three-sided prism shaped structure whose apex was in apposition with the nasal septum and base facing rostrally under skin between the two nostrils as reported earlier (Hare, 1975).

Histology: The dorsal commissure was lined by stratified squamous keratinized epithelium (Fig. 2) as observed in domestic animals (Dellmann, 1993), goat (Kumar *et al.*, 1993; Sinha *et al.*, 2012), camel (Suman *et al.*, 1998) and sheep (Ganganaik *et al.*, 2004b). The stratum basale was comprised of the cell layer resting on the basement membrane. Their nuclei were oval shaped with their long axis parallel to the height of the epithelium and presented smaller clumps of chromatin towards periphery thus leading to vacuolated appearance to the rest of nucleoplasm. These cells contained one or two nucleoli which were centric/eccentric in position. The stratum spinosum was having four to five rows of nuclei which were narrow, oval or elongated shape and were vertically oriented. The chromatin material of these cells was comparatively less basophilic than those of stratum basale cells. The cytoplasm of the stratum basale and stratum spinosum cell layers was finely granular and eosinophilic in nature. The nuclei of stratum granulosum contained fine chromatin material which condensed in small clumps being distributed throughout the nucleoplasm. Some of the nuclei presented degenerative changes. The stratum corneum had two to three rows of the cell layers where nuclei showed almost total degenerative changes. The cytoplasm was strongly eosinophilic and finely granular in nature. Towards free surface, keratinized layer was observed desquamated irregularly throughout the extent as reported in goat (Kumar *et al.*, 1993).

Both the surfaces of the epithelium were irregular. The deeper surface presented papillated and large sized inter-papillary pegs. At some places, the epithelium dipped into deeper part of dermis especially where hair follicles were present. However, in goats (Ganganaik *et al.*, 2004b) arrector pili muscle were present along with hair follicles. The superficial part of dermis had dense irregular connective tissue mainly comprising of collagen, reticular (Fig. 3) and few elastic fibers oriented in different directions, small size blood vessels, fine blood capillaries, connective tissue cells as stated earlier in domestic animals (Dellmann, 1993), goat (Kumar *et al.*, 1993; and Sinha *et al.*, 2012), camel (Suman *et al.*, 1998) and sheep (Ganganaik *et al.*, 2004b). Elastic fibers were well developed in tunica intima of the blood vessels and around hair follicles. The bundles of striated muscles of varying lengths cut into different profiles extended from superficial part of the connective tissue to the deepest part. In the deepest part of the dermis, large number of nerve bundles, few fasciculi of muscles, fatty tissue, elastic fibers and collagen bundles were present. Large clusters of serous type of glands were observed in the deeper part of the

dermis. Their intra and inter glandular ducts were lined by simple cuboidal epithelium.

The ventral commissure was also lined by stratified squamous keratinized epithelium with its free surface showing keratinization whereas, the deeper surface had papillated appearance because of interpapillary pegs as reported in domestic animals (Dellmann, 1993), goat (Kumar *et al.*, 1993; Sinha *et al.*, 2012), camel (Suman *et al.*, 1998) and sheep (Ganganaik *et al.*, 2004b). The epithelium height of ventral commissure was more as compared to the dorsal commissure. In contrast, maximum epithelial height was recorded at dorsal commissure and the least at ventral commissure in goat (Kumar *et al.*, 1993). There were no significant differences in the histology of the different layers of epithelium as compared to the dorsal commissure except varying number of rows in different strata. The cytoplasm of the stratum granulosum and stratum corneum cells was darkly stained with basophilic tinge. The superficial part of the dermis was having loose irregular connective tissue. The other histological features were similar as described in the histology of the dorsal commissure.

The medial and lateral alae were lined by stratified squamous keratinized epithelium as reported earlier in domestic animals (Dellmann, 1993; Kumar *et al.*, 1993; Sinha *et al.*, 2012), camel (Suman *et al.*, 1998) and sheep (Ganganaik *et al.*, 2004b). The ventral surface presented inter-papillary pegs. The epithelium was comprised of strata basale, spinosum, granulosum and corneum with varying number of rows. The connective tissue of dermis was dense and regularly arranged; however, it was irregular towards the papillary pegs. The bundles of striated muscle cut in different profiles, bundles of connective tissue fibers, nerve bundles were extended throughout the dermis. In the deeper part, clusters of serous type of glands were present which were sandwiched in between collagen bundles and fasciculi of muscles. The deeper dermis had dense arrangement of collagen and elastic fibers and nerve bundles. The elastic fibers along with few collagen fibers were present in and around blood vessels and surrounding the muscles bundles. At few places, the fatty tissue was also observed.

Scanning Electron Microscopy: The surface of snout presented irregular surface because of flat epithelial surface which were continuous with each other except a few places where sloughing of cells was also observed (Fig. 4). These cells at higher magnification presented microplicae of different arrangements (Fig. 5). Large sized hair of uniform shape and dimensions emerged from

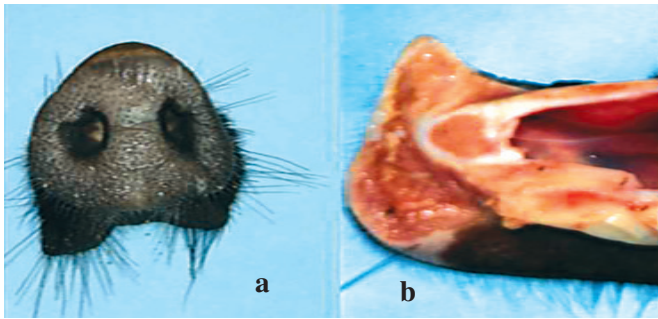
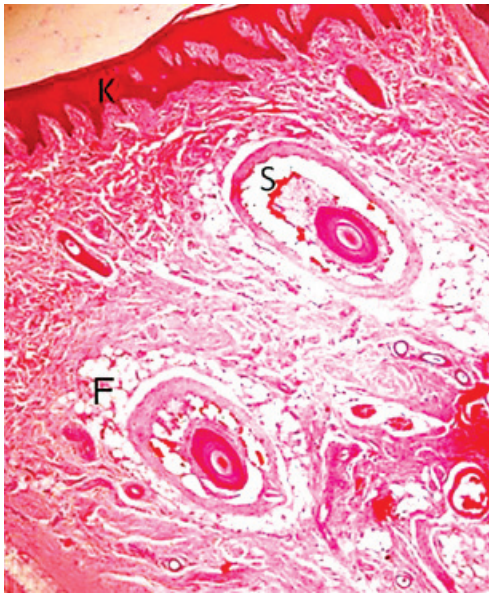
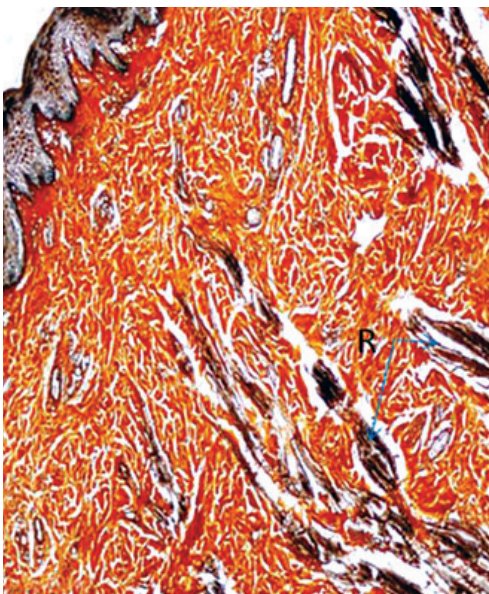


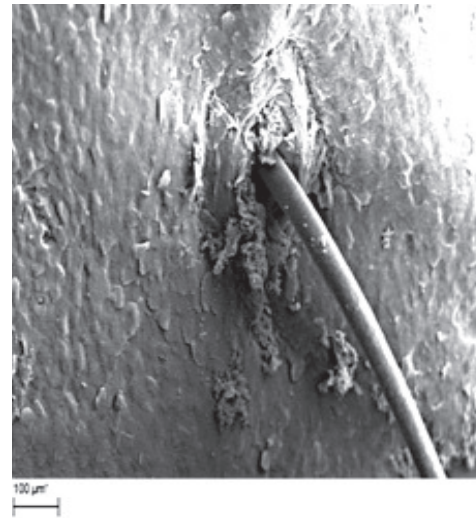
Fig 1. Photograph of snout region of pig showing round to oval shaped external nares (a) and mid-sagittal section showing os-rostrale (S) towards rostral end (b).



Figs 2. Photomicrograph of dorsal commissure of nostril showing stratified squamous keratinized epithelium (K), fatty tissue (F) and sinus hair (S). (H & E $\times 40$)



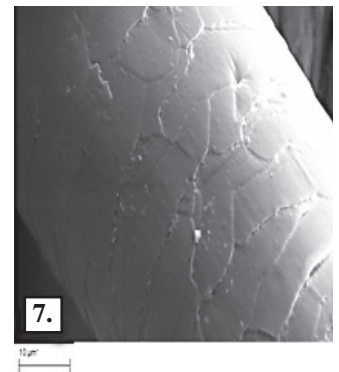
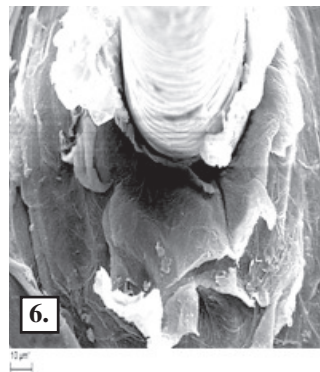
Figs 3. Photomicrograph of lateral ala of nostril showing reticular fibers (R) in dermis. (Gomori's method $\times 100$)



Figs 4. Scanning electron micrograph of snout region showing flat surface and a hair emerging from the deeper part. ($\times 278$)



Figs 5. Scanning electron micrograph of surface showing different arrangements of microplacae. ($\times 3610$)



Figs 6-7. 6. Scanning electron micrograph at higher magnification showing petal like arrangement at the base of hair. Note thread like arrangement at the base of hair and sloughing of epithelial cells. ($\times 1960$); 7. Scanning electron micrograph of hair showing flat cells continuous with each other forming the smooth surface. ($\times 4700$)

deeper surface of snout from a trough like structure. The surface epithelial cells were not continuous at these points. The base of hair was surrounded by cells having a pattern similar to that of petals of a flower and these cells also had microplicae of varying patterns (Fig. 6). The surface of hair was smooth and regular and presented flat cells of different shapes which were lacking microplicae (Fig. 7).

REFERENCES

- Crossman, G.A. (1937). A modification of Mallory's connective tissue stain with a discussion of principles involved. *Anat. Rec.* **69**: 33-38.
- Dellmann, H.D. (1993). Textbook of Veterinary Histology. (4th edn.). Lea and Febieger, Philadelphia, USA.
- Dhingra, L.D. and Kumar, S. (1978). Nasal cavity of buffalo (*Bubalus bubalis*). *H.A.U. J. Res.* **2**: 107-119.
- Ganganaik, S., Jain, R.K. and Kumar, P. (2004a). Gross anatomy of the nasal cavity in sheep (*Ovis aries*). *Indian J. Anim. Sci.* **74**: 605-608.
- Ganganaik, S., Jain, R.K. and Kumar, P. (2004b). Histological structure of the anterior and posterior nares and median nasal septum of sheep (*Ovis aries*). *Indian J. Anim. Sci.* **74**: 754-756.
- Hare, W.C.D. (1975). Porcine Respiratory System. In: Sisson and Grossman's The Anatomy of the Domestic Animals. Getty, R. (edt.). (5th edn.). Vol. 2, W.B. Saunders Co., Philadelphia, USA.
- Kumar, P., Kumar, S. and Singh, Y. (1993). Histology of the nasal cavity boundaries in goat. *Indian J. Anim. Sci.* **63**: 34-37.
- Luna, L.G. (1968). Manual of Histologic Staining Methods of Armed Forces Institute of Pathology (3rd edn.). McGraw Hill Book Co., New York.
- Mygind, N. (1978). Applied Physiology of the Nose. Nasal Allergy. (1st edn.). Blackwell, London, Edinburgh.
- Sinha, M.K., Ray, S., Das, P. and Choudhary, R.K. (2012). Histological studies on the organs of upper respiratory tract from nostril to larynx in black Bengal goat (*Capra hircus*). *J. Cell Tissue Res.* **12**: 3049-3053.
- Suman, Singh, G. and Nagpal, S.K. (1998). Histological studies on the nasal cavity of Indian camel (*Camelus dromedarius*). *J. Camel Prac. Res.* **1**: 99-104.