Plant Archives Volume 20 No. 2, 2020 pp. 7979-7984 **e-ISSN:2581-6063 (online), ISSN:0972-5210**

HISTOLOGICAL FEATURES OF THE NASAL CAVITY IN INDIGENOUS DOGS

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Abstract

The present study was carried out to investigate the histological structure of the nasal cavity in indigenous dogs. Eight adult dogs were used. The tissue were fixed with 10% buffered formalin solution, prepared for paraffin embedding technique and stained with Hematoxyline & Eosin, Masson trichrom stains. The results revealed that, the nasal cavity has three regions: vestibular, respiratory and olfactory regions. The mucosa of the vestibule was lined with thick keratinized stratified squamous epithelium and supported by vascular connective tissue contained sebaceous and mucous vestibular glands. The mucosa of respiratory region has two types of epithelia, the septal respiratory epithelium & conchal respiratory epithelium. The septal epithelium was it was ciliated and non-ciliated pseudo stratified columnar epithelium showed furthermore of goblet cells. It wascovered the nasal septum, dorsal, and ventral meatus. The subepithelial connective tissue composed of loose connective tissue contained glandular acini of septal glands. The conchal epithelium was line nasal conchae and ventral meatus and the ciliated cells were the predominant in this type of epithelium. The mucosa of olfactory region was predominantly lined by olfactory and few of respiratory epithelia. The olfactory epithelium was thick pseudo stratified columnar epithelium has organized into apical three zones. The apical zone was occupied by the supporting cells. the middle zone was occupied by olfactory neuron and devoid of goblet cells and the basal zone was occupied by basal cells. The olfactory epithelium was measured $132.9 \pm 3.04\mu m$, while the respiratory epithelium measured $65.5 \pm 6.10\mu m$ in heights. The olfactory epithelium was supported by vascular connective tissue characterized by many of veins, nerves and bowman's glands. The result concluded that the nasal cavity of indigenous dog is similar that in other animals but the olfactory region showed thicker olfactory epithelium in compared other animals.

Key Word: nasal cavity, nasal concha, nasal septum, respiratory epithelium**.**

Introduction

The nasal cavity is divided into three regions: vestibular, respiratory and olfactory and each region is specialized with its main function which differed from others, in addition a lymphoid tissue presents in the regions of the nasal cavity those play important role in local immunity (Yang *et al.,* 2017). The nasal cavity is the initial rout for many of respiratory diseases such as influenza and pneumonia, so the nasal cavity has an important role in defense mechanism (Qin *et al.,* 2015; Tomosada *et al.,* 2013). Dogs have been proposed as an ideal animal model because the physiology of the canine respiratory tract is more active in smelling than are those of rodent models (Galibert *et al.,* 2016).). On the other hand the inhalation study in doge is once of scientific research to assess respiratory disease therapy and pharmaceuticals that will give via the inhalation route, which is very important in some species due to their close phylogenic relationship to man (Chamanza *et al.,* 2016), for this reason, the investigation of the doges nose is often essential to published an adequate data on the normal histology, as background for other basic science. The present work comprises a histological study of the nasal cavity of indigenous dogs.

Materials and Methods

Animals

Eightadult (2-3 years)indigenous doges were obtained from the department of surgery and obstetrics in the Faculty of Veterinary Medicine, University of Baghdad after which death due to unsatisfactory accidents. The study was carried out at department of Anatomy & Histology, College of Veterinary Medicine, during a period

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extended from April-December 2019.

Histological analysis

After death, animals were decapitated, and the skin, muscles around the nasal cavity was flashed. The nasal region was fixed in 10 % Buffered formalin solution for 48 hours, then cutting up into three transverse sections. The sectioning has made up through the hard palate from the level of the upper incisors to the level of the cribriform plate. The 1st section represented the vestibule of the nasal cavity, the 2nd section was involved the respiratory region of nasal cavity, while the 3rd section was represented the posterior part of nasal cavity involved olfactory region. Each all sections were transformed into solution of formic acid, which used as more save decalcifying solution that composed of equal parts of formic acid solution [250ml formic acid (98%) + 250ml distal water] and Sodium citrate solution [50 gr. Sodium citrate + 250 ml Distal water] (Luna, 1968). The decalcified solution have used for 20 days and the solution changed every 4 days to achieve well decalcification (Altaii, 2010, Abood and Hussein, 2017). Then the tissue sections were prepared for paraffin embedding technique and sectioned serially at 6-7µm. The prepared tissue sections were stained with the Hematoxylin and Eosin stain, Masson's trichrom stain (Bancroft 2008).

Results and Discussion

In indigenous dogs the nasal cavity was dividing into three regions (vestibular, respiratory and olfactory). This result is in dissimilar result of (Kumar, 2008) who divided the nasal cavity of the buffalo into four regions refereeing for transitional zone which located between the vestibular and respiratory region, in fact in our opinion this zone did not represents functional region as those of vestibule, respiratory and olfactory region, and this zone in belonged the respiratory region because that it line with respiratory epithelium. On the other hand this result agrees with that of (Ganganaiki *et al.,* 2004) in sheep, (Moussa and Mokhtar, 2005; Suman *et al.,* 1998; Zguigal *et al.,* 18) in camels, (Yang *et al.,* 2017; Kalita 2014) in pigs, (Kumar *et al*., 2000) in horse, (Kahwa and Balemba, 1998) in goat, (Yee *et al.,* 2016) in white-tailed deer.

Vestibular region: The mucosa of the vestibule was lined with thick keratinized stratified squamous epithelium. The epithelium has composed up to 26 layers of cells. The first layer was the germentive or basal layer which had large rounded darkly stained nuclei followed by 5-8 layers of granular cells, the third layer was flattened and the superficial layer was slightly keratinize flattened cells Fig. 1 and 2. This result disagrees with result of (Kumar *et al*., 2000) in horse who mentioned stratified cuboidal epithelium. On the other hands this result is compatible with the results of most authors (Kumar, 2008) in buffalo, (Ganganaiki *et al.,* 2004) in sheep, (Suman *et al.,* 1998; Moussa and Mokhtar, 2005) in camel, (Yang *et al.,* 2017; Kalita 2014) in pigs, (Sinha, *et al.,* 2015; Taher *et al*., 1989) in sheep, (Kahwa and Balemba, 1998; Kahwa and Purton 1996) in goat, (Zguigal *et al.,* 18) in one -humped camel, (Badawi and Fath El-Bab 20). The epithelium of vestibule was supported by a lamina propria which composed of thick dense irregular collagen bundles that intermingled with blood vessels, nerve and group of sebaceous glands at the entrance of vestibule while at the mid-part was mucous vestibular glands Fig. 3, 4 and 5. These results similar with results of (Suman et al., 1998; Moussa and Mokhtar, 2005) in camel, (Yang *et al.,* 2017; Kalita 2014) in pig. (Eidaroos *et al.,* 1997; Fath El-Bab (21). On the other hands the current result is dissimilar to result of (Ganganaiki *et al.,* 2004) in sheep which include many of hair follicles, sweat glands and serous glands, also dissimilar (Kahwa and Purton 1996) in goat.

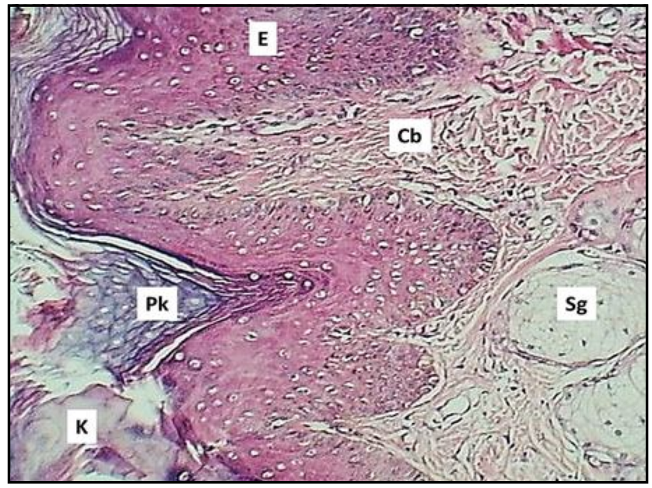

Fig. 1: Sections of at the entrance of the vestibular region show: epithelium (E), lamina propria (Lp), sebaceous gland (Sg), keratin layer (K) & Para keratosis (Pk). H&E stain. 100x.

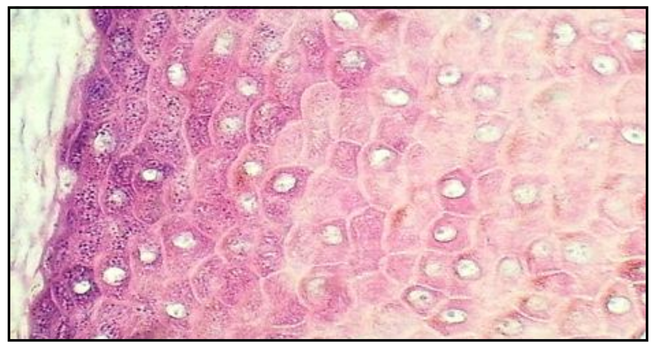

Fig. 2: Sections of stratified squamous epithelium in vestibular region of nasal cavity. H&E stain.400x.

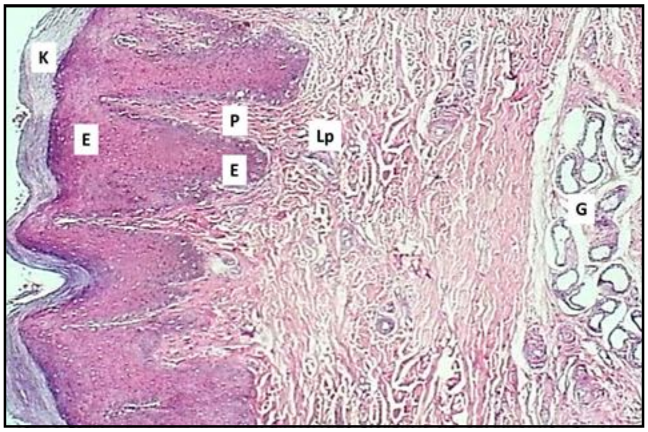

Fig. 3: Sections of at the mid-part of vestibular region show: epithelium (E), lamina propria (Lp), vestibular gland (G) keratin layer (K). H&E stain.40x.

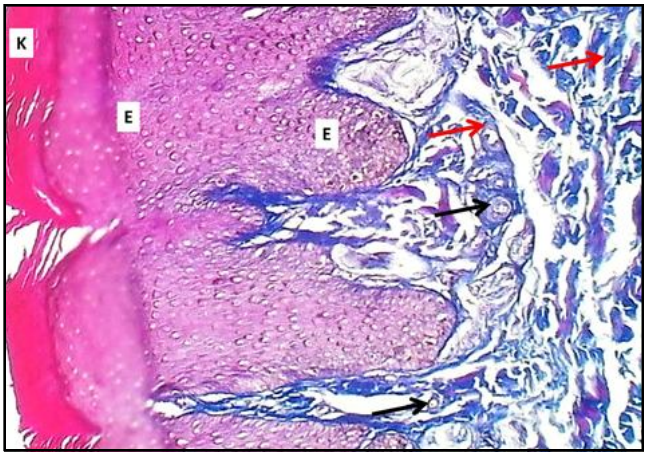

Fig. 4: Sections at of vestibular region show: epithelium (E), arteriole (black arrows), collagen bundles (Red arrows)&keratin layer (K). Masson' trichrom stain.100x.

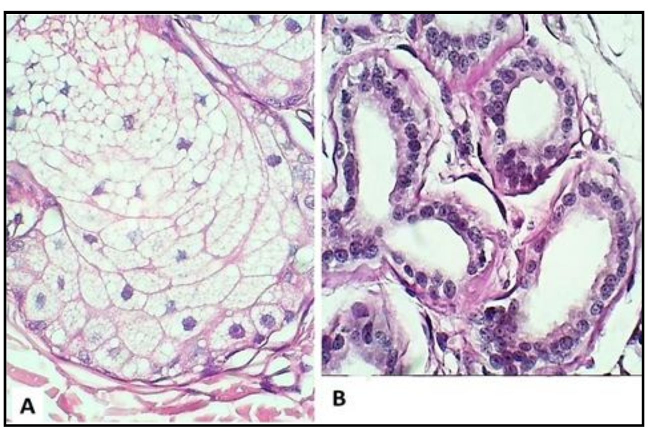

Fig. 5: Section of glandular component of vestibule show: (A) sebaceous gland & (B) alveolar mucus gland. H&E stain.400x.

Respiratory region: The respiratory region was the largest region and its mucosa showed two types of epithelia according for their position: septal respiratory epithelium and conchal respiratory epithelium. Septal respiratory epithelium was coated the nasal septum, dorsal, and ventral meatus and measured 65.5±6.10µm in heights. It was ciliated and non-ciliated pseudo stratified columnar epithelium; involved three cells types, basal cells, ciliated columnar cells and more of the goblet cells, the goblet cells were the predominant type Fig. 6 and 7. This result is agrees with (Kumar, 2008) in buffalo, (Ganganaiki *et al.,* 2004; Sinha, *et al.,* 2015) in sheep, (Moussa and Mokhtar, 2005) in camel, (Yang *et al.,* 2017; Kalita 2014) in pigs, (Kahwa and Balemba, 1998; Kahwa and Purton 1996) in goat and (Zguigal *et al.,* 18) in one-humped camel. The current result suggest that the main function of the upper respiratory tract is to adapt the air entering through the respiratory ducts to suit the nature of the body, and this requires the largest area covered by a specialized epithelium with this their primary functions for producing and secreting mucus by goblet cells which

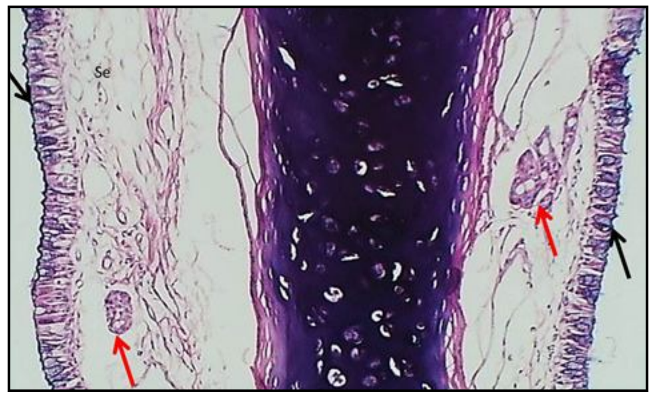

Fig. 6: Section of the nasal septum show: Septal epithelium (Black arrows) & septal nasal gland & (Red arrows). H&E stain. 40x.

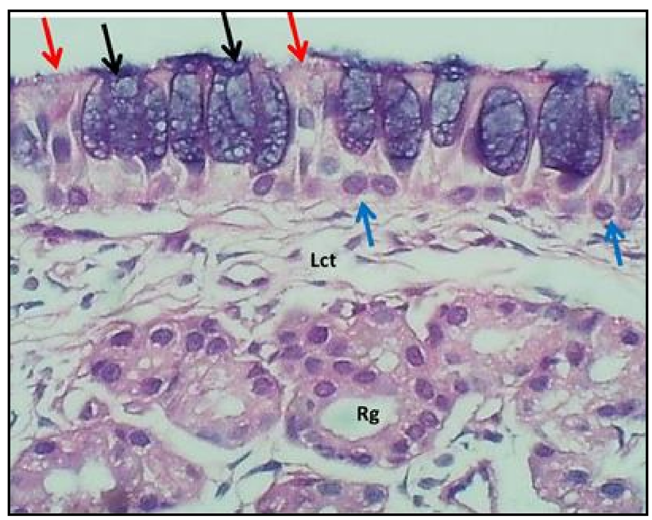

Fig. 7: Section of mucosa of respiratory region (Nasal septum) show: goblet cells (Black arrows) & ciliated cells (Red arrow), nasal septal gland (Rg), basal cells (Blue arrows) & loose connective tissue (Lct). H&E stain.400x.

can be central to mucosal immunity, the goblet cell-derived resistin-like molecule-[2] plays a critical role in recruiting CD4+ T cells (Bergstrom, *et al.,* 2015 & Yang *et al.,* (2017). The subepithelial connective tissue composed of loose connective tissue contained glandular acini of septal glands Fig. 8. This result is compatible with results of (Ganganaiki *et al.,* 2004; Taher *et al.,* 1989) in sheep, (Moussa and Mokhtar, 2005; Abdel-Salam *et al.,* 2014; Zayed, 2004) in camel, (Yang *et al.,* 2017; (Kalita 2014), in pigs, (Eidaroos *et al.,* 1997), Pirie *et al.,* (23) (Suman *et al.,* 1998; Moussa and Mokhtar, 2005). This result suggest that the once function of the respiratory region of nasal cavity is to humidifying the air inside nasal cavity as well as the presence of mucous secretion, helps to catch dust particles and foreign bodies them entering. The conchal epithelium was line nasal conchae, and ventral meatus. It was ciliated and non-ciliated pseudo stratified columnar epithelium; involved three cells types; basal cells, ciliated columnar cells and few goblet cells.

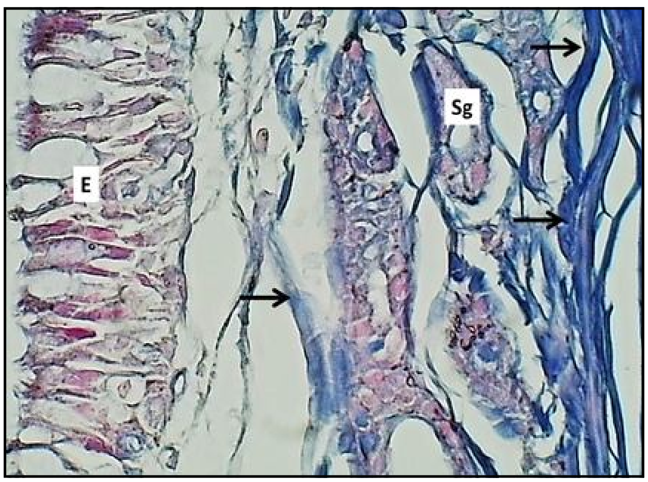

Fig. 8: Section of mucosa of nasal septum show: Epithelium (E), nasal septal gland (Sg) & collagen fibers basal. Masson's trichrom stain.400x.

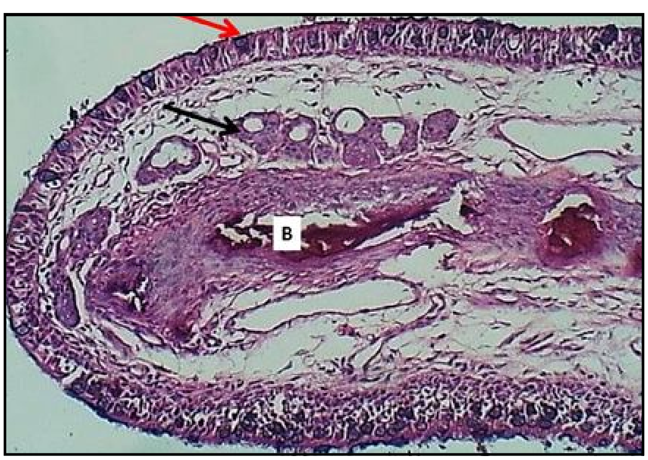

Fig. 9: Section of ventral nasal concha of respiratory region show: epithelium (E) respiratory glands (Black arrow) & conchal bone. H&E stain.100x.

The ciliated cells were the predominant type Fig. 9, the subepithelial connective tissue composed of loose connective tissue contained glandular acini of respiratory gland and supported with conchal bony plate Fig. 9 and 10.

Olfactory region: The mucosa of olfactory region has composed of two types of epithelia; olfactory and respiratory epithelia. The olfactory epithelium was predominated lined the ethmoid concha, most of the nasal and caudal part of nasal septum Fig. 11. This result disagrees with results of (Yang *et al.,* 2017; Yee, *et al*., 2016; Park *et al.,* 2015; Abdel-Salam *et al.,* 2014; Kalita, 2014; kumar 2008; Ganganaiki *et al.,* 2004) who referred for only one type "olfactory epithelium". The current result revealed that the olfactory epithelium was thick pseudo stratified columnar epithelium organized into three cellular zones: Basal zone consist of basal cells which lied on the

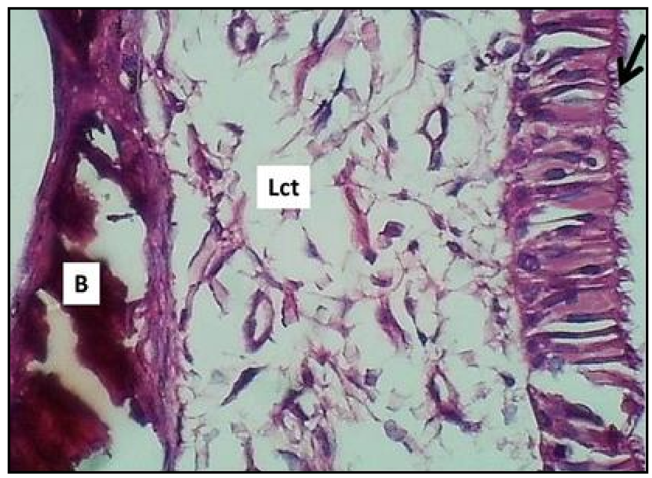

Fig. 10: Section of mucosa of respiratory region (ventral nasal concha) show: predominant ciliated columnar cells (Black arrows), loose connective tissue (Lct)& bone plate (B) H&E stain.400x.

Fig. 11: Section of mucosa of olfactory region show: olfactory epithelium (Oe), respiratory epithelium (Arrows), wide vein (Wv), Bowman glands (Red arrows), axons (Ax). H&E stain.100x.

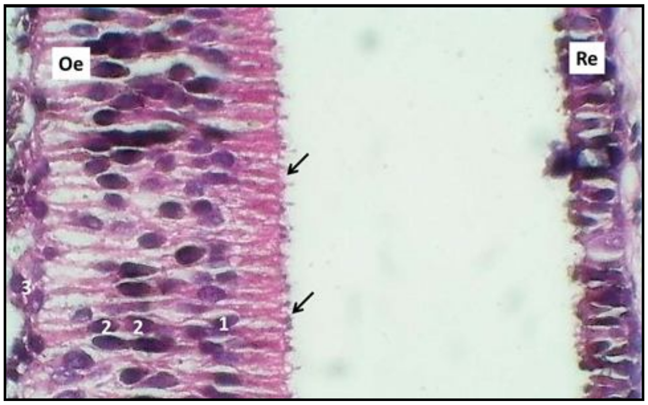

Fig. 12: Section of olfactory region show: respiratory epithelium (Re) & olfactory epithelium (Oe), basal cells (1), olfactory cells (2), sustanticular cells (3) & line demarcation of microvilli (arrows) .H&E stain.400x.

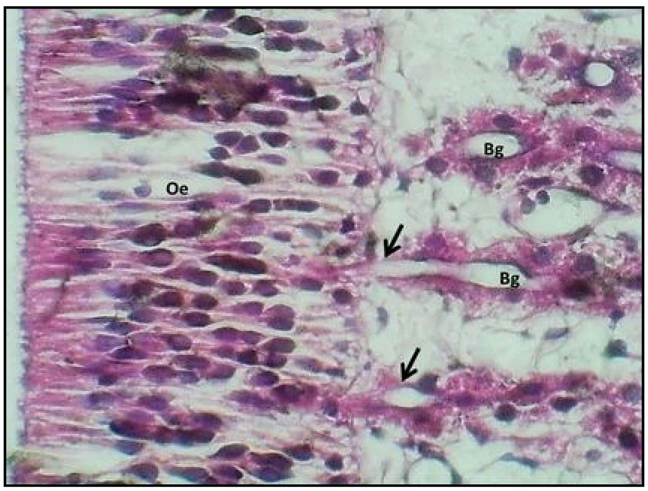

Fig. 13: Mucosa and lamina propria of olfactory region show: olfactory epithelium (Oe) acini of bowman gland (Bg) and their opening into epithelium (arrows). H&E stain.400x.

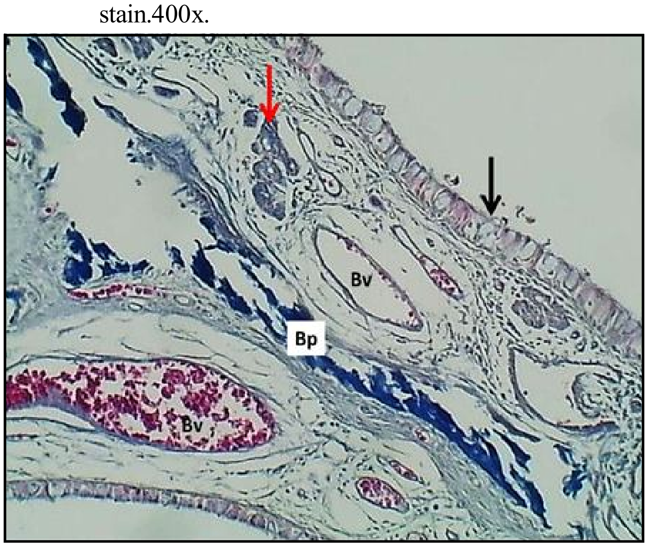

Fig. 14: Section of ventral concha (respiratory region) show: bony plate (Bp), epithelium (Black arrow), serous conchal glands (Red arrows) & blood vessels (Bv). Masson's trichrom stain.100x.

basement membrane and characterized by round-oval nucleus Fig. 12. The Middle zone was the thickest, it consisted of olfactory sensory neurons which were bipolar spindles shape and they had large, pale rounded nucleus. The apical zone was occupied by the supporting (sustentacular) cells which were had epileptical shape, darkly stained nucleus and arranged to form arrow of nuclei near the apical surface Fig. 12. The olfactory epithelium was devoid of goblet cells and measured 132.9 ± 3.04μm in height. The epithelial height of dogs olfactory epithelium was exceeded those recorded in ox (Altaii, 2010) and in Gazella (Abood and Hussein, 2017) and similar to result in a comparion study in dogs and sheep (Kavoi *et al.,* 2010). This suggest that the increased epithelial height is related with the possibility of dog for both day and night operations offers in security such feature will enable more efficient training, leading to improved performances of smelling (Gazit and Terkel, 2003). Also this feature in the dog olfactory region make the dogs can be trained to detect some of volitaile organic compound in patient by smelling urine with a significant success in detection prostate cancer (Cornu *et al.,* 2011). The lamina propria of olfactory epithelium was well vascular connective tissue contained many of wide veins, bundles of nerve fibers and groups of bowman's glands. Bowman glands were compound tubuloacinar glans lined simple cuboidal cells which opened into olfactory epithelium Fig. 11 and 13. This result agrees with result of (Park *et al.,* 2015; Barrios *et al*., 2014; Kalita 2014; Kavoi *et al*., 2010; Suman *et al.,* 1998; Moussa and Mokhtar, 2005; Kumar *et al.,* 1993). On the other hand this result disagrees with result of (Lee *et al.,* 2016) who mentioned that he Bowman's gland acini contains both neutral and acidic mucopolysaccharids. The current result suggests that the importance of neutral serous secretion of bowman's gland that plays an important role add in dissolving and absorption the odiferous molecules. On the other hand there was no acidic secretion seen in this region.

Nasal conchae: The ventral, middle and dorsalconchae were covered by ciliated pseudo stratified columnar epithelium contained much of goblet cells. The subepithelial tissue was composed of highly vascular connective tissue contained serous acini of conchal gland. The core of ventral concha was supported by a thin bony plate Fig.14, 15 and 16 and at the part of dorsal concha near the olfactory region was covered by olfactory epithelium Fig. 17. The caudal (Ethmoturbinate) conch was covered with olfactory epithelium and supported by very thick layer of sub epithelial connective tissue that occupied with serous acini of bowman glands, wide veins axons of olfactory nerve fibers and bone plate Fig. 17. In indigenous dogs the ventral and middle conchae were lined with respiratory epithelium. This result was similar

Fig. 15: (A) section of middle concha (respiratory region) show: bony plate (Bp), epithelium (Black arrow), blood vessel (Bv) and serous conchal glands (Red arrow). H&E stain.100x. (B) Magnified section 400x.

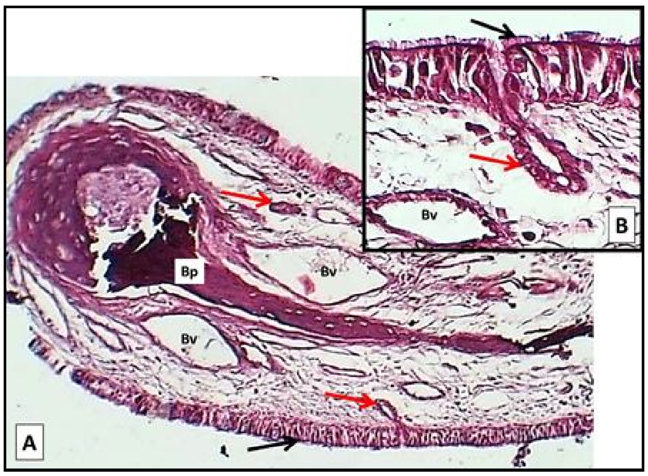

Fig. 16: (A) section of dorsal concha (respiratory region) show: bony plate (Bp), epithelium (Black arrow), blood vessel (Bv) and serous conchal glands (Red arrows). H&E stain.100x. (B) Magnified section 400x.

Fig. 17: (A) Section of dorsal concha (respiratory region) show: bony plate (Bp), olfactory epithelium (Oe), blood vessel (Bv) and serous bowman glands (Back arrows). H&E stain.100x. (B) Magnified section 400x.

that in (Abdel-Salam *et al.,* 2014; Zayed, 2004; Suman *et al.,* 1998; Moussa and Mokhtar, 2005), (Kalita 2014) in pigs, (Kumar *et al*., 2000) in horse, (Taher *et al.,* 1989) in sheep, (Yee, *et al*., 2016) in cat.

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