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Satish Kumar

Department of Veterinary
Anatomy, College of Veterinary
Sciences, Lala Lajpat Rai
University of Veterinary and
Animal Sciences, Hisar,
Haryana, India

Shailja Bansal

Department of Veterinary
Anatomy, College of Veterinary
Sciences, Lala Lajpat Rai
University of Veterinary and
Animal Sciences, Hisar,
Haryana, India

Pawan Kumar

Department of Veterinary
Anatomy, College of Veterinary
Sciences, Lala Lajpat Rai
University of Veterinary and
Animal Sciences, Hisar,
Haryana, India

Ravi Kumar

Department of Livestock
Production Management, College
of Veterinary Sciences, Lala
Lajpat Rai University of
Veterinary and Animal Sciences,
Hisar, Haryana, India

Corresponding Author

Satish Kumar

Department of Veterinary
Anatomy, College of Veterinary
Sciences, Lala Lajpat Rai
University of Veterinary and
Animal Sciences, Hisar,
Haryana, India

Age related scanning electron- microscopic studies on hard palate and oropharynx of the broiler chicks

Satish Kumar, Shailja Bansal, Pawan Kumar and Ravi Kumar

Abstract

The purpose of the present study to investigate the microscopic structure of the hard palate and oropharynx with the help of scanning electron microscope. The roof of oropharynx was formed by the hard palate. The oropharynx and palate was the first area for food selection and intake which was vital to the nutrition and growth of the bird and therefore its commercial viability. A common oropharyngeal cavity was observed that had no clear demarcation between the oral and pharyngeal cavities. The hard palate which was bounded on either side by maxillary ramphotheca. The anterior most portion of the hard palate constituted the upper beak. No obvious morphological distinction could be made between the oral cavity and the pharynx and both cavities formed a common chamber. The hard palate presented a median swelling, from the caudal end of which the lateral palatine ridges were diverging caudally and medially. On either side of the median palatine ridge, the lateral palatine ridge extended to the whole length of the palate. The anterior most portions of these orbital folds were united with each other. At the point of union the different rows of papillae were observed. These orbital folds presented transversely arranged rows of papillae which were of varying shapes and size. These rows of papillae were separated from each other by the smooth surface of the palate where these papillae were absent. The transversely oriented papillae extended up to the lateral margin of the palatine cleft and thus presented the corrugated appearance of the cleft. A large sized longitudinal groove extended up to the level of large sized papillae on either side of the orbital fold of the hard palate where the openings of the lateral palatine glands were present.

Keywords: maxillary ramphotheca, oropharyngeal, orbital folds

Introduction

Fowl production is one of the fastest growing segments of the agricultural sector in the world. It is gaining high attention among small holder farmers as an alternative source of income (Abubakar *et al.*, 2008; Obike *et al.*, 2011) ^[1, 22]. The fowl is an omnivorous bird and therefore has a diet that primarily includes worm and insect on the ground, along with seeds. They are highly adapted to scavenging conditions, and can feed by foraging for a diet of grass, invertebrates and water fleas etc. Variations in food resources have resulted in adaption of fowl to different environments. This has led to difference in shape and structure of the palate and oropharynx (Nickel *et al.*, 1997; King and McLelland, 1984) ^[21, 19]. The palate and oropharyngeal cavity plays a very important role in maintenance of food in oral cavity, movement and in swallowing of bolus as a reflection of the different lifestyle of avian (Dehkordi *et al.*, 2010) ^[6]. The macroscopic structure of palate differs greatly with the feeding habits (Hodges, 1974) ^[14]. The gross anatomical features of the palate and oropharynx of birds have been described in a number of wild and domestic species. One of the most comprehensive works is that of Goppert (1903) ^[10] which compared the oropharynx of numerous species and serve as the basis for later description of this region (McLelland, 1979) ^[20]. Some attention has been given to the study of the morphology of the avian oropharynx and tongue in recent times (Crole and Soley, 2009; Igwebuikwe and Eze, 2010; Tivane *et al.*, 2011; Erdogan and Alan, 2012) ^[4, 15, 29].

However, specific information on the anatomy and scanning electron microscopy of the hard palate and oropharynx of the fowl is still very minimum and also there is paucity of literature on the macroscopic anatomy of hard palate and oropharyngeal cavity in the domestic birds (Nickel *et al.*, 1977) ^[21]. This is an important region considering that these findings may be important in nutritional and medical management of fowl especially under the intensive system of production. Furthermore, this study may provide a foundation for identifying the structural features in the palate and oropharynx of the fowl.

Material and Methods

The present study was conducted on 30 broiler chicks of one week to one month of age (7-32 days) which were divided into 5 groups each group having 6 birds. The heads of dead birds were collected after post-mortem examination at 7, 11, 18, 25 and 32 days of age. The tissues were collected from rostral, middle and caudal portions of hard palate, oropharynx and associated glands. Fresh tissues from these selected sites were also collected for scanning electron microscopy and fixed in 2% glutaraldehyde solution for 6-8 hours after thorough washing in chilled 0.1M phosphate buffer (pH 7.4). The tissues were again washed twice with 0.1M phosphate buffer. The tissues were dehydrated in grades of ethanol, critical point dried and sputter coated. The processed tissues were viewed under scanning electron microscope (SEM) to record observation and photograph.

Results and Discussion

Only a limited number of studies were available on the anatomical and histological structures of the avian palate (Samar *et al.*, 1995, 1999, 2002; Tivane, 2008; Igwebuike and Eze, 2010; Crole and Soley, 2011; Erdogan and Alan, 2012) [23, 24, 25, 28, 5, 15] but almost no literature was available on scanning electron microscopy of hard palate.

The scanning electron micrograph of the hard palate of 7 days old birds revealed that the median swelling which was strongly convex in shape and sloped laterally (Fig. a). At its caudal extremity, it diverged into the lateral palatine ridges as reported by Gupta *et al.*, (2015) [11]; Nickel *et al.* (1977) [21] and Sisson and Grossman (1955) [26] in fowl. At the junction of the two on either side a small depression indicating the opening of maxillary glands which was indistinct in gross anatomical study. This median ridge was smooth and did not present any papillae. Similarly, the lateral palatine ridge was convex, sloped medially and laterally and its surface presented small nodular like papillae. The median ridge after the point of divergence led into a median structure which presented the small granular like arrangement and isolated blunt type of papillae on its either side. The boundaries and components of the oral and pharyngeal cavities agreed with the general avian pattern as previously described (Goppert 1903; Farmer and King 1972, McLelland 1979, 1993) [10, 8, 20] and with the brief description of Bezuidenhout (1999). As noted above, no obvious morphological distinction could be made between the oral cavity and the pharynx and both cavities formed a common chamber. This situation is apparent in most avian species due to the absence of a soft palate and oropharyngeal isthmus (McLelland, 1979, 1993) [20].

At 32 days of age, scanning electrograph revealed that the lateral palatine ridge appeared smooth, convex and sloped medially as well as laterally and its surface presented very faint small ridges from middle to the caudal aspect as revealed with the findings in the rhea (Gusseklou, 2006) [13], emu (Crole and Soley, 2009) [4] and ostrich (Tivane *et al.*, 2011) [29]. The first row of papillae was observed at its junction with the choanal cleft. The transversely oriented row of blunt type of papillae was observed whereas in the African pied crow (Igwebuike and Eze 2010) [15], common raven and European magpie (Erdogan and Alan, 2012), conical papillae were located particularly in the palatine mucosa surrounding the choanal cleft. Our study was in agreement with Erdogan and Alan (2012) in magpie and raven that, there were numerous conical backward directed papillae scattered singly or arranged in rows, in which the palatine surface have

numerous scattered singly caudo-medially directed conical papillae.

The middle one was the largest and oriented straight or parallel to the palate. On either side of this central papilla, two papillae were also observed. These papillae were smaller in size, obliquely placed and appeared to be emerged from the mucosa of the hard palate. These papillae presented an overlapping pattern. Sometimes a third very small sized papilla was also observed on either side. A narrow space was presented just caudal to these papillae and in the center choanal cleft was present. The first row of papillae 3-4 on each side was more pronounced at the 11 days of age. There were numerous orderly arranged rows of notches called lamellae situated on the lateral border of the hard palate in Muscovy duck (Igwebuike and Anagor, 2013) [17] and these were not observed during the present study.

The median surface of choanal cleft was smooth up to the portion where the 2nd row of transversely oriented papillae was observed (Fig. b). An elongated median choanal cleft with the long rostral narrow and the caudal wide part observed during present study was similar to that of Eurasian hobby (Abumandour, 2014) [2], oval depression with two compartments in Muscovy duck (Igwebuike and Anagor, 2013) [16], very long in pigeon, short in duck and goose (Nickel *et al.*, 1977) [21] and inverted V shaped depression in herons and ducks (McLelland, 1979) [20], while the choanal cleft may take the bell-shape in ostrich (Catarina *et al.*, 2011) [3]. The mucosa was devoid of the papillae between the rows of papillae. These papillae were arranged in a comma shaped arrangement and generally six papillae were observed on either side of the mid plane. The size of the papillae decreased as moved from medial to the lateral position. However, all the papillae were conical shaped with pointed free ends. Just caudal to these papillae, the choanal cleft was comparatively narrow and its medial border presented the irregular surface because of presence of small sized papillae. Sometimes, it presented a corrugated appearance. Lateral to this choanal cleft was the location of the orbital fold which presented the papillae. These papillae were distributed randomly except in the caudal part where again a transverse row of papillae was observed. The rest of the surface of orbital fold presented small scales like arrangement which were separated by small spaces. The papillae present in the orbital fold appeared as if embedded in the surface of the palate. These were generally covered by the scales of the surface. The papillae presented in the caudal part having a regular arrangement. These were generally 5-6 papillae on either side of the narrow part of the choanal cleft. These papillae were conical shaped and directed caudally. These papillae were separated by each other by narrow spaces. These papillae were comparatively smaller in size than those which were present at the caudal border of the choanal cleft. These papillae placed on both sides were separated by choanal cleft. These papillae increased in the size as moved from medial to the lateral portion. These papillae were generally 16 on each side of the median plane (Fig. c). These papillae were larger in size than rest of papillae presented on the mucosa of the hard palate. Papillae present on the rostral portion of the orbital folds were conical in shape and some were fused at their bases. These papillae were showing different pattern of micropliae. Just caudal to these papillae there was another large row of conical papillae which were projecting caudally and the caudal free extremity was above the level of the surface of orbital folds. The first five papillae were almost of equal size with very blunt or

rounded free ends. These papillae were slightly broader towards the base and separated from each other by narrow groove. The free tips presented the shape as if have been cut transversely. The rest of the papillae increased in size as moved laterally. The free ends of papillae were blunt and showed transition towards the free ends. These papillae were separated from each other by spaces, which extended up to the middle portion whether towards the base of the papillae the mucosa was continuous. At some places either two papillae were fused with each other or some of the papillae were bifurcated into two portions towards the free ends. This arrangement was seen up to the 12th papilla but these papillae were generally 15-16 on each side of the median plane at 25 days of age. The next four papillae were larger in size and they showed their origin from the more cranial portion of the mucosa. It was common finding that the caudally pointed papillae that had been reported as a typical feature of the hard palate in most avian species (McLelland, 1979; Igwebuike and Eze, 2010) ^[20, 15]. A similar lack of papillae on the hard palate and roof of the oropharynx had been demonstrated in the rhea (Gussekloo and Bout, 2005) ^[12] and ostrich (Tivane *et al.*, 2011) ^[29].

At 18 days, of age the papillae showed growth in the first row of papillae. The central one was more pronounced and clearly visible. The anterior part of the choanal cleft became wider. In the 2nd row of papillae, 6 papillae were observed. There were six transverse rows of caudally directed papillae on either side of the choanal cleft as reported in pigeon (Getty, 1975; Nickel *et al.*, 1977) ^[9, 21] whereas there was lack of papillae on the hard palate and roof of the oropharynx of Muscovy duck (Igwebuike and Anagor, 2013) ^[16], rhea (Gussekloo and Bout, 2005) ^[12] and ostrich (Tivane *et al.*, 2011) ^[29] which showed faint papillae only in its caudal part. Tadjalli *et al.*, (2008) ^[27] also reported that the anterior two third of hard palate contained no papillae, while the caudal part of the palate had short and slender papillae surrounding choanal cleft in ostrich. At 25 days of age, a second row of papillae started to appear between the choanal cleft and the point of bifurcation of the median swelling. The transversely oriented row of blunt type of papillae was observed at the cranial end of choanal cleft. The middle one was the largest and oriented straight or parallel to the palate. On either side of central papilla, 3-4 papillae were also observed. At 32 days of age of birds the border of the choanal cleft was margined by a longitudinally oriented row of papillae which comprised of only single row of papillae. The choanal cleft became wider just caudal to the level of palatine papillae. These papillae were more conical in shape than previous age groups. The orbital folds lodged the caudal extremity of the broader part of the choanal cleft up to the level of palatine papillae. The palate of fowls and pigeons has caudally pointing papillae arranged in several transverse rows, but the palate of goose had a median and 2-3 paramedian longitudinal rows of blunt papillae and in the duck, these papillae were confined only to the apical region (Nickel *et al.*, 1977) ^[21].

The orbital folds were separated by narrow portion of choanal cleft. It was similar to what has been described in guinea fowl (Jayachitra *et al.*, 2015) ^[18]. Each half of these folds presented the irregular row of conical shaped papillae which were arranged in 2-3 in number. Except these conical shaped papillae the surface of the orbital fold presented the small scales which were of varying shape and size and separated by small spaces. The border of the choanal cleft was margined by a longitudinally oriented row of papillae which comprised of only single row of papillae. The caudal most portion of the orbital fold was separated by a narrow groove from the transversely oriented fold of the mucosa presenting the palatine papillae. The choanal cleft again became wider just caudal to the level of palatine papillae and its middle wall presented small conical shaped papillae which showed the corrugated appearance. The caudal portion of choanal cleft was blunt and followed by a smooth surface being followed by an infundibular cleft.

Scanning electron microscopy of oropharynx:

The pharyngeal fold was present on both sides of choanal and infundibular cleft. The pharyngeal folds on either side of the choanal cleft were smooth however towards the infundibular cleft irregular rows of papillae were present. On either side of infundibular cleft flat cells were observed. These cells were showing the microplicae at higher magnification. The pattern of microplicae was different in different cells and in some cells microplicae appeared depressed. The surface of conical papillae showed a regular surface presenting the flat squamous cells and these cells were having the arrangement of depressed type of microplicae. The anterior most part of pharyngeal fold was smooth whereas, at the caudal portion papillae were present. At most caudal part of pharyngeal fold a transversely placed row of papillae was observed. These papillae were increased in number (12) on each side at 11 days of age. These papillae presented the arrow head appearance. These papillae having corniculate shape were directed caudally. The surface of these papillae was flat and having embedded type of microplicae. The most medially placed papillae were most caudally placed whereas most caudally placed papillae were anteriorly placed. These folds were separated from the esophagus by these transversely placed papillae. At 25 days of age the papillae in pharyngeal mucosa were in 6-8 rows and these papillae were conical in shape with broad base (Fig. d). At 32 Days of age on either side of choanal cleft there were appearance of single row of longitudinally arranged papillae which were over hanged the choanal cleft and these were slender in shape with pointed apices and were caudally directed. The surface of conical papillae showed an irregular surface presenting the flat squamous cells. The infundibular cleft became wider and presented papillae on its caudal end.

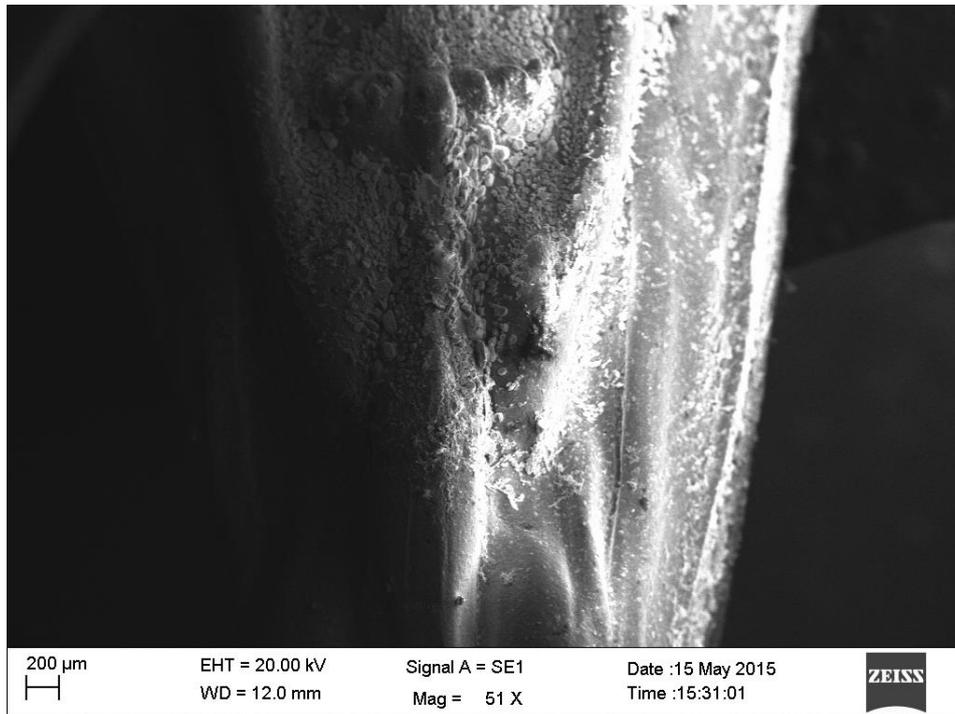


Fig (a): The median swelling which was strongly convex in shape and sloped laterally and at its caudal extremity, it diverged into the lateral palatine ridges

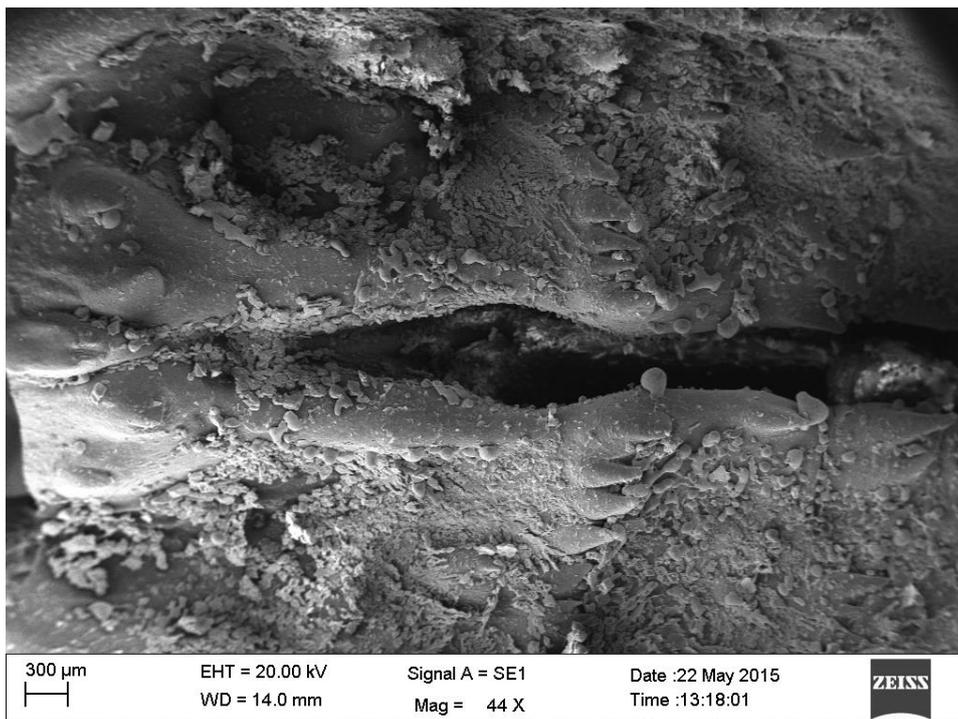


Fig (b): The median surface of choanal cleft was smooth up to the portion where the 2nd row of transversely oriented papillae was observed.

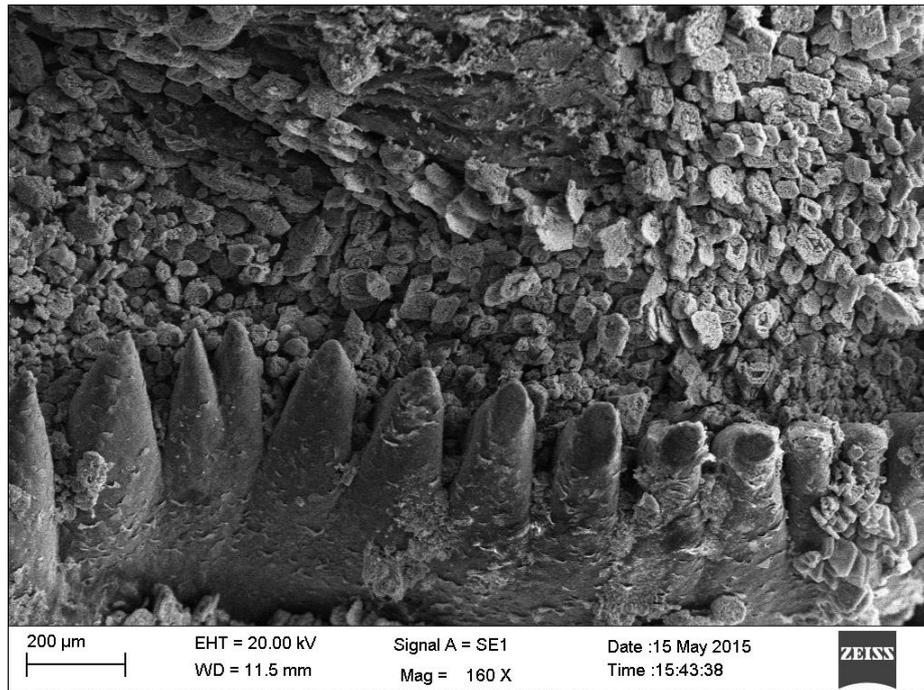


Fig (c): At the caudal border of the choanal cleft, the papillae were generally 16 on each side of the median plane

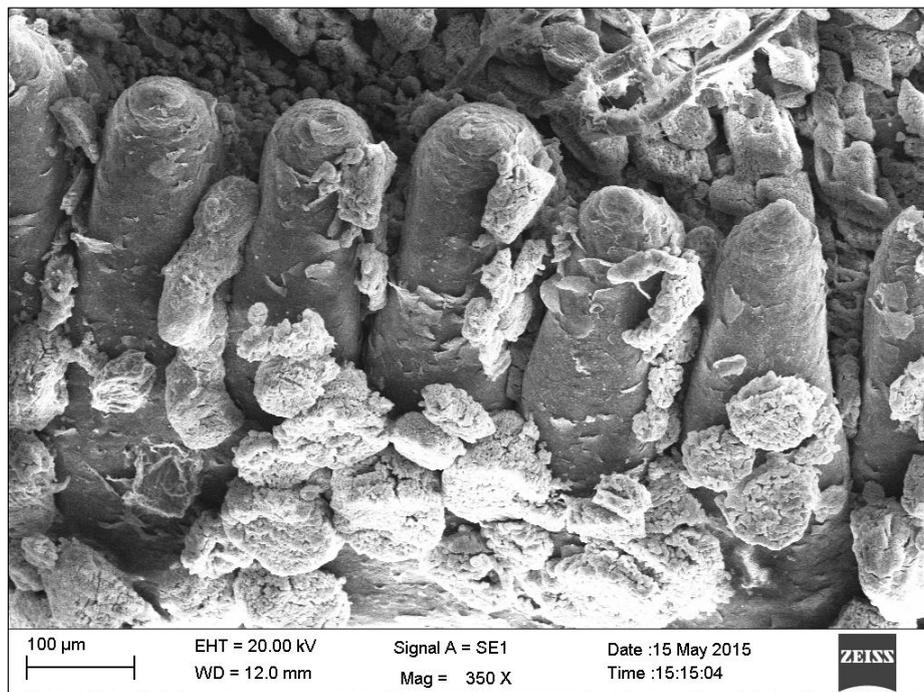


Fig (d): The papillae were conical in shape with broad base.

Conclusion

The present study was conducted on hard palate and oropharynx in a systematic manner in broiler chicks of the different age group. The scanning electron micrograph of the hard palate revealed that the median swelling was strongly convex in shape and sloped laterally. At its caudal extremity, it diverged into the lateral palatine ridges. This median ridge was smooth and did not present any papillae. Similarly, the lateral palatine ridge was convex, sloped medially and laterally and its surface presented small nodular like papillae. The first row of papillae was observed at its junction with the choanal cleft. The middle one was the largest and oriented straight or parallel to the palate. These papillae were smaller in size, obliquely placed and appeared to be emerged from the

mucosa of the hard palate. The median surface of choanal cleft was smooth up to the portion where the 2nd row of transversely oriented row of papillae was observed. These papillae were arranged in a comma shaped arrangement and generally six papillae were observed on either side of the mid plane. However, all the papillae were conical shaped with pointed free ends. Just caudal to these papillae, the choanal cleft was comparatively narrow and its medial border presented the irregular surface because of presence of small sized papillae. These papillae were distributed randomly except in the caudal part where again a transverse row of papillae was observed. The rest of the surface of orbital fold presented small scales like arrangement which were separated by small spaces. These were generally covered by the scales

of the surface. The papillae present in the caudal part were having a regular arrangement.

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