

HISTOMORPHOCHEMICAL CHARACTERIZATION OF MANDIBULAR SALIVARY GLAND IN YOUNG PIG (*Sus scrofa*)

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ABSTRACT

The present study was conducted on six healthy young pigs of local mixed breed of either sex to study the histology and histochemistry of the mandibular salivary gland. The gland was pear-shaped and located inferioposterior to the angle of mandible which lied superficial to the suprahyoid, and infrahyoid muscle groups. Histologically, the gland was compound tubuloacinar type and was characterized by a mixed parenchyma of mucous and serous secretory acini along with several orders of ducts distributed in the stroma. The myoepithelial cells appeared as flattened basal cells around the secretory acinar cells and formed the incomplete layer, enclosed by a basement membrane. The interstitial tissue contained scattered plasma cells and sparse lymphocytes in addition to the connective tissue fibres, numerous small blood vessels and nerves. The duct system was comprised of intralobular, interlobular and large excretory ducts. The intercalated as well as striated ducts were of intralobular type, which first opened into interlobular ducts and finally empty into the large excretory duct lined by stratified columnar epithelium. The interlobular ducts converged to form the long, horizontal main excretory duct which opened into the oral cavity. Histochemical studies revealed that the presence of neutral mucopolysaccharides, weakly acidic sulfated mucosubstances, sialomucins and hyaluronic acid in the secretory units. The goblet cells also showed the presence of all these muco-substances in the interlobular ducts of the mandibular gland.

Keywords: Histochemistry, Histomorphology, Mandibular salivary gland, Young pig

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The salivary glands are invaginations from buccal epithelium into the lamina propria-submucosa (Singh and Singh, 2017). These develop at different sites and they have very different architectures and produce different types of saliva (Jaskoll *et al.*, 2002). The salivary glands are known as multifunctional organs as they perform many important digestive, protective, excretory and endocrine functions (Miletich, 2010). The secretion (saliva) of the mandibular salivary gland plays an important role in the moistening and swallowing of newly ingested food and maintenance of oral hygiene. The saliva is mostly serous containing various enzymes, water, mucopolysaccharides and lubricating glycoproteins (Singh and Singh, 2021). The broader distributions of the salivary glands are advantageous for the protection of the oral cavity against pathogens (Sumi *et al.*, 2007). Dysfunction of salivary secretion (hyposalivation) causes xerostomia (dry mouth) and sequentially leads to severe dental caries as well as oral mucosal disorders (Featherstone, 2000).

The structure of mandibular salivary gland of prenatal pig (Zhou *et al.*, 2010), rabbit (Al-Saffar, 2014), sheep (Singh *et al.*, 2015) and buffalo (Singh and Singh, 2017) has been studied at both macroscopic as well as microscopic levels, but the mandibular salivary gland of postnatal pig have received little attention, especially from the histomorphochemical point of view. Keeping in view the importance of mandibular gland, the present study describes the light microscopic details of the gland in young pig and its comparison with other domestic animals.

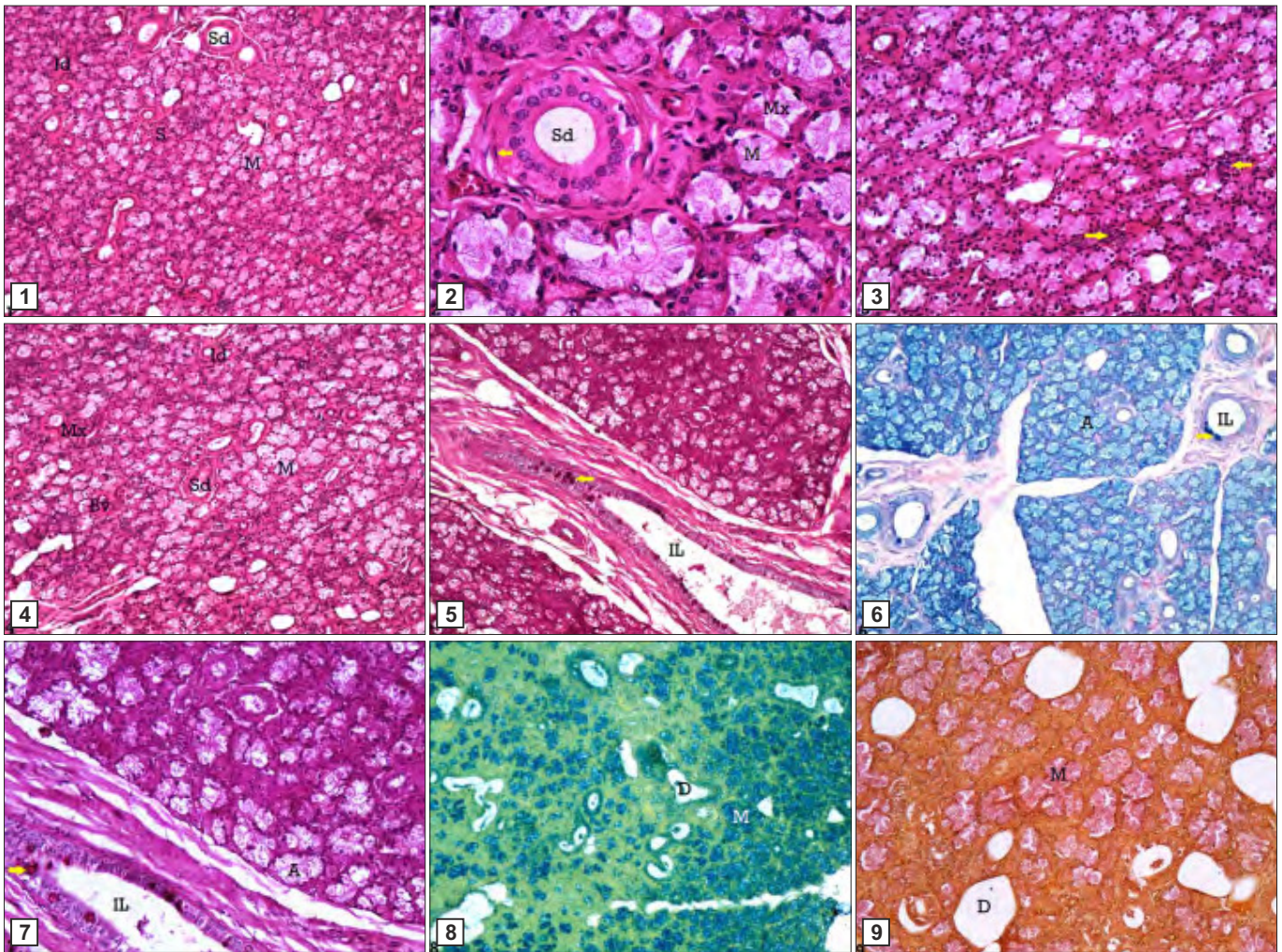
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MATERIALS AND METHODS

The present study was conducted on mandibular salivary gland of six healthy young pigs of local mixed breed of either sex of 6-8 months of age. The head of young pigs were procured from local slaughter house immediately after decapitation and the tissues were fixed in a 10% neutral buffered formalin solution for 48 hours, subjected to routine tissue processing for light microscopic examination and embedded in paraffin blocks. The paraffin sections (5-6 μ) were made through the entire gland and stained with routine Harris' hematoxylin and eosin stain for general histomorphological examination, Gomori's stain for reticular fibres, Weigert's method for elastic fibres (Luna, 1968) and Crossman's trichrome stain for collagen fibres (Crossman, 1937). In addition, selected sections were performed for histochemical demonstration of mucopolysaccharides using Periodic-Acid-Schiff-Alcian blue (PAS-AB), Alcian blue (AB) at pH 2.5, McManus' method, colloidal iron method and Mayer's mucicarmine method (Luna, 1968).

RESULTS AND DISCUSSION

The mandibular gland of pig was of compound tubulo-acinar type as observed earlier in camel (Mansouri and Atri, 1994), buffalo (Singh and Singh, 2017) and goat (Rauf *et al.*, 2004). The gland was enclosed in a thick layer of fibrous connective tissue known as capsule. The dense septa primarily composed of collagen fibres along with few reticular fibres, arise from the capsule and passed into the gland to divide it into lobes and lobules. The size of



Figs. 1-9. (1) Photomicrograph of mandibular salivary gland of pig showing the lobule consisted of mixed type of secretory cells comprised of mucous (M) and serous (S) secretory acini along with several orders of intercalated (Id) and striated (Sd) ducts. H & E \times 40. (2) Photomicrograph showing the glandular parenchyma of mandibular salivary gland consisted of pyramidal shaped mucous (M) and mixed (Mx) acinar cells along with striated duct (Sd). The incomplete layer of myoepithelial cells (arrow) were also seen around the striated ducts. H & E \times 400. (3) Photomicrograph of mandibular salivary gland of pig showing the interstitial tissue comprised of sparse amount of lymphocytes and scattered plasma cells (arrow) in addition to the connective tissue fibres, numerous small blood vessels and nerves. H & E \times 100. (4) Photomicrograph of mandibular salivary gland of pig showing the various orders of intralobular duct system (intercalated (Id) and striated (Sd) along with the secretory acini (mucous = M, mixed = Mx) and blood vessels (Bv). H & E \times 40. (5) Photomicrograph showing the presence of goblet cells (arrow) among the epithelium of interlobular ducts (IL) of mandibular salivary gland of pig. H & E \times 40. (6) Photomicrograph showing the presence of weakly acidic sulfated mucosubstances in the secretory end-pieces (A), whereas a strong positive reaction was seen in the goblet cells (arrow) of interlobular ducts (IL) of the mandibular salivary gland of pig. Alcian blue 2.5 \times 40. (7) Photomicrograph of mandibular salivary gland of pig showing strong positive reaction of neutral mucopolysaccharides in the goblet cells (arrow) of interlobular ducts (IL), however weak to moderate reaction was seen in secretory acinar cells (A). PAS \times 100. (8) Photomicrograph showing the moderate to strong reaction of acidic mucosubstances in the mucous secretory cells (M) and ducts (D) showed weak reaction in the mandibular salivary gland of pig. Colloidal iron method \times 40. (9) Photomicrograph of mandibular salivary gland of pig showing the moderate to strong reaction of mucosubstances in the mucous secretory cells (M) and ducts (D) showed weak reaction. Mayer's mucicarmine method \times 100.

lobules varied without a definite pattern. Similar type of arrangement was observed in the horse (Dellmann and Eurell, 1998), goat (Rauf *et al.*, 2004) and neonatal buffalo (Singh and Singh, 2017). Each lobule consisted of mixed type of secretory cells of mucous and serous secretory acini along with several orders of ducts (Fig. 1). Although, the gland was reported to be of mucous type in dogs and cats, serous in rodents, mixed in horses, humans and ruminants (Banks, 1992). Furthermore, Mansouri and Atri (1994) also mentioned that the secretory cells of camel were of two types; mucous cells grouped into secretory tubules and acini and seromucous cells grouped into acini

and demilunes. Adnyane *et al.* (2010) found that in barking deer (*Muntiacus muntjak*) the mandibular gland had serous and mucous cells with the mucous type predominating.

In present findings the pyramidal shaped acinar cells were arranged around a narrow lumen and characterized by the presence of flat nucleus located near the periphery of the cell (Fig. 2). These findings were in parallel agreement with those of Singh and Singh (2017) in buffalo. Myoepithelial cells were observed around the secretory end-pieces as well as intercalated and striated ducts of the mandibular salivary gland which result in the formation the incomplete layer, enclosed by a basement membrane (Fig. 2).

However, Singh and Singh (2017) stated that the presence of myoepithelial cells surrounding the intercalated duct was considered as a distinguishing feature to identify these ducts in mandibular salivary gland of buffalo. The interstitial tissue comprised of sparse lymphocytes and scattered plasma cells in addition to the connective tissue fibres, numerous small blood vessels and nerves (Fig. 3). The duct system was comprised of intercalated, striated, interlobular and large excretory ducts (Fig. 4). The secretory cells first opened into small intercalated ducts, lined by simple cuboidal epithelium as reported in mandibular gland of horse (Dellman and Eurell, 1998). These intercalated ducts were then empty into the large striated ducts, lined by simple columnar epithelium. These large striated ducts with a wide lumen were characterized by longitudinal striations in the basal third portion of cells, extending from the base of cells to the level of the nucleus. The cytoplasm of the cells lining the striated ducts was eosinophilic, while the nuclei were basophilic and darkly stained. Lemmon (2008) reported that the striated ducts occurred in groups and lacked distinct striations in buffalo calves. These ducts then opened into the interlobular ducts and finally empty into the large excretory duct lined by stratified columnar epithelium. These findings were in agreement with observations of Ikegbu *et al.* (2014) in African giant rat. The interlobular ducts converged to form the main duct which opened into the oral cavity. Goblet cells were present among the epithelium of interlobular and large excretory ducts with their varied in numbers (Fig. 5) which is in agreement with the finding of the Nawar and El-Khaligi (1977) in camel.

Histochemical studies revealed the presence of neutral mucopolysaccharides, weakly acidic sulfated mucosubstances, sialomucins and hyaluronic acid in the secretory end-pieces and the stroma of the gland (Fig. 6). A strong to intense Periodic-Acid-Schiff (PAS) positive reaction was seen in the goblet cells of epithelium of interlobular and large excretory ducts (Fig. 7); however, strong Alcian blue (AB) reaction was noticed in these goblet cells (Fig. 6). Similar type of activity was observed in mandibular gland of camel (Mansouri and Atri, 1994), rabbit (Al-Saffar, 2014) and buffalo (Singh and Singh, 2017). Localization of neutral mucopolysaccharides in acinar cells of bovine mandibular salivary gland was observed, whereas these cells lacked acidic mucopolysaccharide content (Lemmon, 2008). Colloidal iron (Fig. 8) and Mayer's mucicarmine (Fig. 9) methods also showed the presence of mucosubstances in the secretory acinar cells.

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