Histomorphological comparison of tonsil of Large White Yorkshire and indigenous pigs of Kerala, India


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ABSTRACT

The present study was undertaken to compare the distribution and histomorphology of the tonsil of soft palate in Large White Yorkshire and indigenous pigs of Kerala. Tissue samples were collected from 12 apparently healthy adult male animals. In both the groups of pigs, the tonsil of soft palate was located in the oropharynx and was found to be larger in Large White Yorkshire. Histological structure of tonsil of soft palate revealed that the average number of lymphatic nodules per microscopic field and lymphocyte count per nodule was maximum in indigenous pigs. Statistically significant differences were recorded in the average size of tonsils, crypt depth, number and size of lymphatic nodules and lymphocyte count per nodule in Large White Yorkshire and indigenous pigs.

Key words: Crypts, Lymphatic nodules, Pig, Tonsil of soft palate.

INTRODUCTION

Since tonsils are regarded as secondary lymphoid tissue and part of the acquired immune system which is subjected to induction through contact with antigens, an evaluation of the different lymphocyte populations in tonsils is useful to determine a tendency of the specific tonsils to more inductive or more effective immunity (Breugelmans et. al, 2011). The tonsil of the soft palate in pigs is a part of Waldeyer’s ring (Tenorio and Pabst, 2006; Casteleyn et al., 2011). It has been known for a long time to be the site of colonization of important swine and zoonotic bacterial pathogens (Kernaghan et al., 2012).

Since the pig is a highly prolific animal and is used as a model for most of the biological experiments, it is of utmost importance in the studies regarding immune system. Although extensive works have been done on the lymphoid organs of pigs, a detailed study on the porcine tonsil of soft palate is scanty. Since the indigenous pigs are comparatively more resistant to many of the respiratory and digestive disorders than Large White Yorkshire, the present study has been undertaken for a comparative study on the tonsil of soft palate of these two breeds.

MATERIALS AND METHODS

Histomorphological comparison of the tonsil of soft palate of Large White Yorkshire and indigenous pigs of Kerala was carried out using the tissue samples collected from 12 apparently healthy adult male animals of ages ranging from eight to ten months (six from each group) sold for slaughter from Centre for Pig Production and Research, Mannuthy. The age and body weight of the animals were recorded. The median sections of the heads were taken using Bone and Meat cutting machine (Yorco, Y172). Both the halves were washed thoroughly in fresh water and the shape, size, colour, location, extent, morphometry and topographic relation of tonsil of the soft palate in both the breeds of pigs were recorded. Then the tissue samples were dissected out of the head and fixed in 10 per cent neutral buffered formalin. Tissue samples were processed as such by routine histological methods, paraffin blocks were made and sectioned to a thickness of 5µm. The tissues were stained by Haematoxylin and Eosin (H&E), Gomori’s rapid one step trichrome method for connective tissue fibres and Ayoub Shklar method for keratin and prekeratin. Micrometric parameters were recorded using ocular micrometer and photographs were taken using Olympus microscope attached with a digital camera.

RESULTS AND DISCUSSION

The tonsil of the soft palate was very prominent and seen on the ventral surface of the soft palate as two elongated and oval plaques on either side of the median palatine raphe in both the groups (Fig. 1 and 2). The mucosal surface was flattened and pitted with openings called fossules, which were wider in Large White Yorkshire as compared to indigenous pigs (Fig. 3). Belz and Heath (1996) and Liu et al. (2012) made similar observations on the morphology of the tonsils in pigs. In contrast Cocquyt et al. (2005) and Yang et al. (2011) observed that the tonsil of the soft palate consisted of scattered nodules of lymphoid tissue located on the dorsal (nasal) side of the soft palate in sheep and camel respectively.

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These variations from the present study might be attributed to species difference.

The average size of the tonsil and number of fossules were found to be significantly different (p ≤ 0.01) between both the groups. Longer (6.16 ± 0.05 cm) and wider tonsils (2.85 ± 0.05 cm) with more number of fossules (127.00 ± 6.49) were seen in the Large White Yorkshire than in the indigenous pigs.

The tonsil of soft palate was lined by a non-keratinized stratified squamous surface epithelium which could be differentiated into stratum basale, stratum spinosum and stratum superficiale (Fig. 4). The stratum basale was composed of a single layer of cells with oval nuclei and a slightly basophilic cytoplasm. Cells of the stratum spinosum were arranged in 10 to 12 layers. The nucleus of the deepest cell layer was similar to those of the stratum basale. However towards the superficial layers cells with round to oval nuclei and less basophilic cytoplasm were seen. These cells were placed parallel to the longitudinal axis of the epithelium. The cells of the outermost stratum superficiale were arranged in varying number of cell layers with elongated basophilic pyknotic nuclei and eosinophilic uniform cytoplasm. The surface epithelium had an irregular outer surface and an unevenly placed deeper surface which extended into the lamina propria mucosa as interpapillary pegs. These observations confirmed the earlier reports of Belz and Heath (1996) in pigs and Kumar and Timoney (2006) in horse. However Casteleyn et al. (2010) reported that in sheep the rostral half of the tonsil of the soft palate was lined by a pseudostratified columnar ciliated epithelium and the epithelium of the caudal part was stratified squamous non-keratinized. These variations in the epithelium might be due to the position of the tonsils in nasal surface in sheep.

The surface epithelium was perforated by crypts with very small openings which penetrated into the substance of the tonsil and presented a neck and a body. The neck was formed by an invagination of soft palate surface epithelium and was lined by the same stratified squamous epithelium

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**Fig 1:** Ventral view of the soft palate of Large White Yorkshire pig showing the position of tonsil of soft palate
1. Tonsil of soft palate  2. Median palatine raphae  
3. Root of tongue  4. Epiglottis

**Fig 2:** Ventral view of the soft palate of indigenous pig showing the position of tonsil of soft palate
1. Tonsil of soft palate  2. Median palatine raphae  
3. Root of tongue  4. Epiglottis  
5. Hard palate

**Fig 3:** Surface of the tonsil of soft palate of 1. Large White Yorkshire, 2. Indigenous pig showing tonsillar fossules
while a lymphoepithelium or reticular epithelium lined the body. Similar observations were also documented by Anderson (1974) in pigs. The deep crypts and body provided an intimate relationship between the lymphocytes of the parenchyma and the epithelium. Belz and Heath (1996) suggested that although the crypts in pig tonsils provided a large surface area for contact between the crypt epithelium and luminal contents, their openings into the oropharynx were very narrow. Since antigen sampling within the crypt epithelium could occur only after the organisms penetrated through the narrow crypt lumen, the upward and backward forces generated during deglutition were involved in forcing the oropharyngeal contents into the crypts and in the expulsion of mucus and entrapped particles and micro-organisms.

In both the groups of pigs, the lymphoepithelium consisted of two to five cell layers of cuboidal or columnar cells, intraepithelial lymphocytes, basal cells and goblet cells overlying an incomplete basement membrane and thin layer of connective tissue, above the apex of the follicles (Fig. 5 and 6). All these observations made in the present study supported the earlier findings of Belz and Heath (1996) in pigs. However goblet cells were not recorded in oropharyngeal tonsils of other domestic animals by Banks (1993). These cells might contribute mucus to the crypt and facilitate the removal of detritus as opined by Ramos et al. (1992) in pigs.

Statistically significant differences were recorded in the height of the crypt epithelium, depth of the crypts and diameter of crypt openings between both the groups (p≤ 0.01). Comparatively smaller crypt epithelium (88.33 ± 20.27 µm) with deeper crypts (1022.00 ± 18.43 µm) and smaller crypt openings (118.80±4.76 µm) were seen in the indigenous pigs. Belz and Heath (1996) observed that the crypt openings varied from 90 to 500 µm across their long axis in pigs. In indigenous pigs the deeper crypts with comparatively smaller crypt epithelium will increase the surface area and provide an intimate contact between the antigen and the underlying lymphatic tissue and assist in maintaining better mucosal immunity.

The lamina propria of the tonsil of the soft palate was mainly composed of loose irregular connective tissue, lymphatic tissue and glandular tissue in both the groups of pigs. The parenchyma of the tonsil was mainly composed of lymphatic nodules and diffuse lymphatic tissues. Some of the nodules presented germinal centres. Cryptolymphatic units and tonsillar nodules were present. The average internodular space was also significantly different statistically.
and it was more in Large White Yorkshire pigs due to which comparatively less number of lymphatic nodules were seen in an unit area. In the interfollicular and parafollicular areas meshwork of reticular and collagen fibres contained small, medium and large lymphocytes, plasma cells, macrophages, blood capillaries and HEVs (Fig. 7). All these observations made in the present study tally with the reports of Belz and Heath (1996) and Liu et al. (2012) in pigs and Kumar and Timoney (2006) in horse.

A connective tissue capsule made of collagen and reticular fibres separated the lymphatic tissue from the adjacent glandular tissue. Its average thickness was 310±34.48 µm and 308±0.05 µm in Large White Yorkshire and indigenous pigs respectively. Connective tissue septa arose from the capsule and penetrated into the lymphoid tissue (Fig. 8). Similarly Belz and Heath (1996) observed that the tonsil of soft palate in pigs were capsulated and penetrated by septa. However, Cocquyt et al. (2005) studied the tonsil of the soft palate in sheep and concluded that they were not macroscopically distinguishable from the surrounding tissue and were not encapsulated. The variation in the present study might be because of the difference in size and location of tonsils in the nasal cavity in the sheep.

The average number of lymphatic nodules per field did not differ significantly (p ≤ 0.01) between both groups of pigs. Statistically significant difference was recorded in the average diameter of the small lymphatic nodules and number of lymphocytes in the lymphatic nodules and it was more in the indigenous pigs. This may provide better immunological response to antigens which enter the body by the oral route. The average internodular space was also significantly different statistically and it was more in Large White Yorkshire pigs due to which comparatively less number of lymphatic nodules were seen in an unit area. In the present study the average lymphocyte count per lymphatic nodule was maximum in the tonsil of soft palate as compared to other tonsils in both the groups of pigs. Contrarily Kumar and Timoney (2006) described that in horse lymphoid tissue in the tonsil of soft palate was lesser than when compared to other tonsils. The difference in the present study may be due to species difference since in horse well developed paired palatine tonsils are also present to compensate the reduced amount of lymphoid tissue in soft palate tonsils, while in pigs due to the absence of the former, soft palate tonsils were well developed.

It was concluded that the tonsil of the soft palate was the largest among all the tonsils present and visible macroscopically at the ventral surface of the soft palate. On statistical analysis significant differences were noticed in the average size of tonsils, crypt depth, number and size of lymphatic nodules and lymphocyte count per nodule in the Large White Yorkshire and indigenous pigs studied. Though the average size of tonsils of soft palate was larger in Large White Yorkshire, the crypt depths were more in the indigenous pigs that provided a larger surface area and intimate contact between the lymphocytes of the parenchyma and the surface epithelium probably leading to an intense and instantaneous immune response. The average size of lymphatic nodules and number of lymphocytes per nodule was more in indigenous pigs when compared to Large White Yorkshire. Since the lymphocytes carry out the activities of immune system, it may be interpreted that in the indigenous pigs the tonsils offered better immunological protection against antigen that entered the body by oral or nasal routes and hence they are resistant to many of the respiratory and digestive disorders when compared to Large White Yorkshire pigs.
REFERENCES