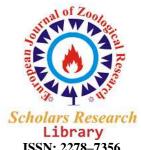


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European Journal of Zoological Research, 2013, 2 (5):26-31 (http://scholarsresearchlibrary.com/archive.html)



ISSN: 2278-7356

# Histogenesis study of lingual papillae in sheep fetus

# Sajjad Hejazi<sup>1\*</sup>, Rahman Baroughi<sup>2</sup>

<sup>1</sup>Department of Anatomy, Faculty of Veterinary Medicine, Tabriz Branch, Islamic Azad University, Tabriz, Iran

<sup>2</sup>Veterinary Student, Faculty of Veterinary Medicine, Tabriz Branch, Islamic Azad University, Tabriz, Iran

#### **Abstract**

The aim of this study is determining process of lingual papillae tissue development in sheep fetus. The study was observation and descriptive type and the survey was done on 100 sheep fetuses of Ghezel breed from different age groups. The desired samples were collected by random sampling. The crown- rump length of collected fetus (CRL) were measured, and according to the formula: X=2.1 (Y+17) their ages were calculated. The tongue of the fetus was separated from its head after fixation in formalin 10%, They were surveyed for sampling under microscope for observation of growth area and development of lingual papillae. The separated samples were examined under light microscope after histological processing and staining (H&E)the tissue sections. By microscopic observation, which has been surveyed from 40 days of pregnancy, at first epithelium of tissue was cuboidal and stratified cuboidal cells and by progress of the developmental period, they obtiand the final form of stratified squamous cells. The development of the lingual papillae appeared after complete formation of the tongue epithelium tissues. At first, we observed the mechanical papillae appearance and in the later stages, The taste buds were formed from collection of tongue epithelium cells which appeared on the tasting papillae. During this process we observed keratinized epithelium filiform papillae. It may be concluded that the time of developing lingual papillae are dependent to the final formation of lingual epithelium tissue. It can be Concluded that there is no relationship between the beginning time of filiform papillae keratinization and the diet of the animal after the birth.

**Key words:** fetus, sheep, lingual papillae,

## INTRODUCTION

The tongue development begins from fourth week of embryo. The tongue is formed as five ridges originating from the pharyngeal arches[14].Dorsal surface of tongue mucosa has different kinds of lingual papillae so in general they are divided into two groups of lingual papillae: mechanical and taste. This classification is based on the presence or absence of taste cells in the taste bud [13]. The tongue epithelial tissue is of non-keratinized stratified squamous [2]. However, the tongue mucosal papillae have the keratinized mechanical form and seen as stratified squamous kind [2]. Taste buds are found in large numbers in the taste papillae. In histological sections three types of cells are identifiable in the buds. Sensory cells of taste bud are fed by even nerves of ninth and tenth cranial nerves [3]. In comparative morphological studies, scientists have found a variety of vertebrate tongue morphology and development [11]. As in the study of vertebrate tongue morphology, they achieved the relationship between the appearance and evolution of the tongue and the changes in animal habitat [11]. Also they proposed a direct relationship between tongue mucus and ecology. As in wetlands habitat to dry lands habitat, the tongue epithelium changes from keratinized to squamous [10].

## MATERIALS AND METHODS

The population in this study was ewes. The sample consisted of 100 fetuses removed from the uterus of pregnant ewes which were slaughtered. Random Sampling was performed. Then, gestational age was calculated according to the following formula X = 2.1 (Y +17), where Y= the fetus 's caput to rump length (CRL). After determining the age, fetuses were labelled and recorded in a data note and finally were stabilized at 10% buffered formalin. The age of collected embryos was from 40 to 130 days after the pregnancy. In this study, the subjects were divided into 4 groups, The first, second, third and fourth groups included the ages of 40-60, 60-90, 90-120, and 120-150 day-olds, respectively. Before the final sampling for histological study, the samples were observed under a dissecting microscope to determine the location of the emergence and growth of the tongue papillae in order to determine the location of tissue sampling. The isolated samples were examined under light microscope after taking histological sections and stained with hematoxylin - eosin and trichrom's mason.

## **RESULTS**

The tongue tissue is mesenchymal until the age of 40± 2 days of pregnancy and contains abundant mesenchymal cells that some of which are differentiated into skeletal muscle fibres in the centre of the tongue.

The epithelial lining of the tongue is cubic in this age of. At this stage there was no indication of differentiation of the tongue mucosa epithelium (figure1).

It seemed in the samples aged  $43 \pm 1$  days of gestation that the dorsal lining of the tongue epithelium was thicker than the ventral side and there were signs of forming domed papillae at several points of the tongue mucosa (figure 2). So we can say that since day  $43 \pm 1$  of gestation, the tongue papillae of the sheep have appeared. Since  $50 \pm 2$  days of age, the tongue epithelium changes from stratified cuboidal to stratified squamous. At this age, the tongue papillae on the anterior part of the tongue were clearly emerging (figure 3). At this stage we observed villus morphological differentiation which are sharp at the anterior segment and domed gradually towards the posterior part of the tongue.

In the observations made on samples of  $72 \pm 3$  days of age some buds were formed from the generative cells of dome form epithelial papillae so it seemed that taste buds appear only in domed papillae (figure 4). At the age of 78  $\pm 2$  days, the taste people were formed in the vicinity of the filiform papillae on the posterior part of the tongue (figure 5).

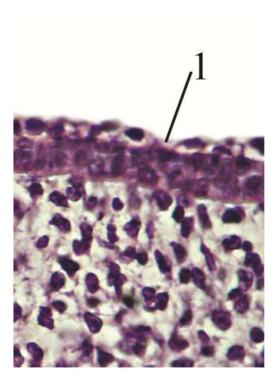


Figure 1: 1 - Microscopic view of Cubic stratified epithelium, on 40 days (H&E Stained with magnifying ×400)

Until the age of 90± 5 days of gestation the filiform papillae had non keratinized stratified squamous epithelium but later on the filiform papillae of epithelium became keratinized.

In the observations made on samples of aged  $110 \pm 5$  days of gestation, cup, fungiform and filiform papillae with a variety of taste bud cells, were clearly distinguishable from each other (figure 6). It seems that the lingual tissue at  $115\pm 5$  days of gestation finds its final form. So Mucosal epithelium of the tongue takes its fix form from birth to 120 days of age(figure7). The observations of the samples after 120 days of age demonstrated that filiform papillae of epithelial keratinised cells had scaling and degenerative forms (figure 8).

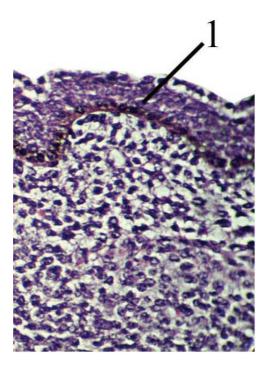


Figure 2: 1 - Microscopic view of differentiating the epithelium to primary papillae on 43 days. (H&E Stained with magnifying  $\times 200$ )

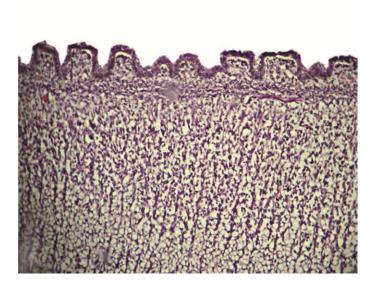


Figure 3: Microscopic view of e initial papillae growth on 52 days. (H&E Stained with magnifying  $\times 100$ )

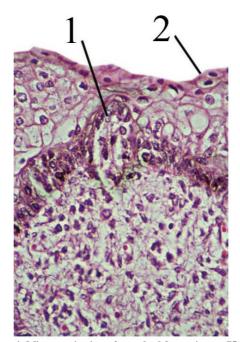


Figure 4: Microscopic view of taste bud formation on 75 days. 1 - The initial taste bud 2 – non-keratinized stratified squamous epithelium (H&E Stained with magnifying  $\times 200$ )

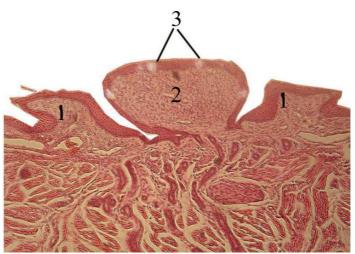
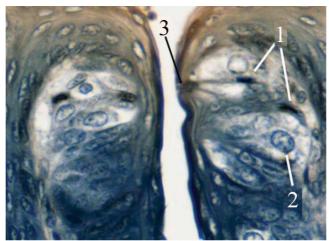
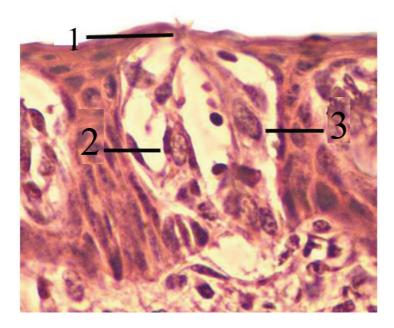


Figure 5: Microscopic view of the legal papillae on 80 days.

1. Filiform papillae, 2. Fungiform papillae 3. Taste bud. (H&E Stained with magnifying ×100)



 $\label{eq:Figure 6: Microscopic View of taste bud on 115 days. 1. Supporting cells, 2. Sensory cells, 3-taste bud. \\ (Trichrom mason Stained with magnifying <math display="inline">\times 800)$ 



 $Figure~7:~Microscopic~View~of~taste~bud~on~120~days.\\1.~Taste~buds,~2.~Supporting~cell,~3.~Taste~supporting~cell.~(H\&E~Stained~with~magnifying~×800)$ 



 $\label{eq:figure 125:microscopic view of filiform papillae on 125 days. 1. Keratinized stratified squamous epithelium. \\ (H\&E Stained with magnifying \times 100)$ 

Table 1: Tongue epithelial demonstrations based on age of fetuses

Age of fetus (days)	40±2	43±1	50±2	67±3	75±3	90±5	115±5	120
Variable								
Cubic stratified epithelial	*							
Non-keratinized Stratified squamous epithelium			*					
Keratinized Stratified squamous epithelium						*		
Emergence of initial papillae of the tongue		*						
Differentiation of mechanical papillae of the tongue				*				
Differentiation of taste papillae of the tongue					*			
Differentiation of taste cells							*	
The final form of the tongue mucosa							·	*

# DISCUSSION

Based on the obtained results of this study it can be concluded that the tongue epithelium development starts from a simple cube, then changes to stratified and finally changes to non-keratinized stratified squamous form. Histological studies identified that the tongue epithelium is as non-keratinized stratified squamous [2].

According to the observations made in the mucosa of the tongue there was no lingual papillae with cubic epithelium, while all lingual papillae appeared with stratified squamous epithelium. It can be concluded that the time of emergence and growth of the lingual papillae depends on the time of forming the stratified squamous epithelium. Histological studies have shown that all of the papillae epithelium is stratified squamous one [8]. The observations showed that the first appearance of the tongue was as sharp form and eventually was converted to filiform papillae and taste papillae appeared in the evolutionary process It has been reported in a study that the mechanical papillae are the first differentiated papillae of the tongue and Taste bud cells of the tongue papillae are the last one so they emerge near the delivery [4]. There is also a significant correlation between the morphology of the taste papillae with the nervous system of the region [4]. According to the observations about the appearance of taste buds, it can be concluded that all of the taste bud cells are formed by gathering the generative epithelial cells. So it has been reported in a study that the taste buds are originated from the taste papillae [12]. In another study the contrast of epithelial and mesenchymal tissues in order to form taste papillae during the organogenesis process [11]. It was seemed from the examination of the kind of tongue filiform papillae that filiform papillae epithelium is as non-keratinized at the initial stages of differentiation but on the last months of pregnancy it becomes a keratinized mood.

## **CONCLUSION**

It can be Concluded that there is no relationship between the beginning time of filiform papillae keratinization and the diet of the animal after the birth. So the keratinisation of filiform papillae epithelium occurs during embryonic life.

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