Investigations on postnatal development of the foliate papillae of the tongue by the light and scanning electron microscopy in the white laboratory mice

B. TOPRAK*, YILMAZ*

1 Etlik Central Veterinary Control and Research Institute, Ankara, TURKEY
2 Department of Anatomy, Veterinary Faculty, Fırat University, 23119 Elazig, TURKEY
* Corresponding author: E-mail: syilmaz@firat.edu.tr

SUMMARY

The postnatal development and structure of foliate papillae of the tongue were examined by the light and scanning electron microscope in the mice. In that aim, the animals were divided into eight groups according to their age (0, 15, 30, 60, 90, 120, 150 and 180 days, n=8 per group).

Foliate papillae were located between the body and the radix of the tongue at the caudo-lateral sides of it. Foliate papillae grooves started to open at day 3 and were completely opened at postnatal day 8. Taste buds with developed pores were seen at this stage. Two different, light and dark cells of taste buds were distinguished in taste buds. The foliate papillae developed more rapidly during the first 30 days than between day 30 and day 90 and completed their development at day 90. Foliate papillae were shown to exhibit mucosal crypts rather than laminas by scanning electron microscope. The thickness of epithelial border cells was apparent with high magnification. There were lots of micro-ridges on the surface of these cells.

In conclusion, it was shown that foliate papillae of the tongue achieved their development at day 90 and that taste buds became functional at day 8. No differences were evidenced on the surface structure of the papillae from the birth to the adulthood period in mice.

Keywords: Development, foliate papillae, mice, scanning electron microscope.

RÉSUMÉ

Étude du développement post-natal des papilles foliées de la langue par microscopes photonique et électronique à balayage chez la souris blanche de laboratoire

Le développement post-natal et la structure des papilles foliées de la langue ont été examinés chez la souris par microscopes photonique et électronique. Pour cela, les animaux ont été répartis en 8 lots en fonction de leur âge (0, J15, J30, J60, J90, J120, J150 et J180 jours post-natal, 8 souris par groupe).

Les papilles foliées étaient situées entre le corps et la racine de la langue en région caudo-latérale. Les sillons des papilles foliées de la langue commençaient à s’ouvrir à J3 et étaient complètement ouvertes à J8. Les bourgeois du goût avec des pores développés ont été identifiés à ce stade. Deux types de cellules claires ou sombres ont été distinguées au niveau des bourgeois du goût. Les papilles foliées se sont développées plus vite au cours des 30 premiers jours qu’entre les stades 30 et 90 jours et leur développement s’est achevé au stade 90 jours. La microscopie électronique a mis en évidence une structure en cryptes muqueuses des papilles foliées plutôt que des structures laminaires. L’épaisseur des cellules de bordure de l’épithélium a été évaluée avec un fort grossissement. De nombreuses microvilli ont été mises en évidence à la surface de ces cellules.

En conclusion, il a été montré que les papilles foliées de la langue achèvent leur développement au stade 90 jours post-natal et que les bourgeois du goût sont fonctionnels à partir de J8. Aucune différence de structure de surface des papilles n’a été mise en évidence de la naissance à la période adulte chez la souris.

Mots-clés Développement, papilles foliées, souris, microscopie électronique à balayage.

Materials and Methods

In this study, both male and female BALB/c mice were used. The day of birth was accepted as day zero for the age. Animals were divided into eight groups according to the age i.e., 0, 15, 30, 60, 90, 120, 150 and 180 days. Each group consisted of eight animals. In addition, another 84 mice aged
from 0 to 10 days were used to detect the opening time of taste buds. The animals were killed by gaz asphyxia. For the light microscopic examination, area of foliate papillae was fixed in 10% formalin solution. Papillae samples were processed routinely for paraffin embedding and sectioned at 5-7 µm. Sagittal sections from papillae were stained by hematoxylin eosin [15], Crossman triple staining [4] and Periodic acid-Schiff (PAS) [15]. Micrometric measurements were performed by an ocular micrometer.

The tongue samples of three mice from each group were used for the scanning electron microscopic examination. Two of these tongue samples were used as whole and one of them was divided in two parts and all samples were rinsed with the physiologic water-buffered saline. Tissue samples were prefixed in 3% gluteraldehyde (pH: 7.3) solution with phosphate buffer. After tongue fixation in the same buffer, samples were post-fixed in 1% osmium tetra-oxide at 37°C for 1.5 hours. Then, the tongue samples were treated by 3 N hydrochloric acid at 60°C for 20 minutes to remove the mucus on the surface of the tissue [9, 10]. The tongue samples were then rinsed with alcohol and amyl-acetate solutions and dried by using critical-point-dryer. Finally, they were coated with the gold by using VG Microtech Polaron SC 500 sputter-coater under 0.05 Torr pressure. All samples were examined, photographed and evaluated by the Jeol JSM 5600 scanning electron microscope between 5 and 15 kV.

Results

Foliate papillae were located as 4-6 rows between the body and the radix of the tongue at caudo-lateral sides in the mice.

The grooves of foliate papillae were not open and the taste buds were not present in the newborn mice. However, these grooves started to open at day 3 (Figure 1ab). The grooves of foliate papillae were totally opened at day 8 and pores of taste buds were first seen first at day 8. Glandula gustatoria (von Ebner glands) channel grooves of papillae were completely opened after day 15 in all animals.

It was detected that the margin, length and width of ridges of the papillae (Folium papilla); and depth and width of grooves of the papillae (sulcus papilla) developed more rapidly during the first 30 days than between day 30 and day 90 and completed their developments at day 90. The taste buds also completed their developments at day 90. No important differences were detected between days 90 and 180 (Table I).
Taste buds were seen in two of three papillae grooves and were situated vertically. The taste buds opened to papillae grooves via taste pores. Two different cell types of taste buds were distinguished, light cells (epitheliocytus sensorius gustatorius) and dark stained cells (epitheliocytus sustentans), (Figure 1c). The rate of the length to the width of taste buds was 1.50. Foliate papillae were seen as mucosa ridges rather than layer (Figure 2a) and no significant changes were detected after the birth. The thickness of the cell borders was clear by the higher magnifications and there were micro-ridges on the surface of these cells (Figure 2b).

### Tableau 1: Average measurements taken from foliate papillae and taste buds in the postnatal period.

<table>
<thead>
<tr>
<th>Age of Animals (Days)</th>
<th>0</th>
<th>15</th>
<th>30</th>
<th>60</th>
<th>90</th>
<th>120</th>
<th>150</th>
<th>180</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Length of papilla ridge (µm)</strong></td>
<td>87.2 ± 3.5</td>
<td>122.1 ± 3.6</td>
<td>155.8 ± 5.2</td>
<td>164.2 ± 4.0</td>
<td>178.9 ± 5.6</td>
<td>179.5 ± 5.3</td>
<td>178.1 ± 6.8</td>
<td>178.3 ± 4.2</td>
</tr>
<tr>
<td><strong>Width of papilla ridge (µm)</strong></td>
<td>75.0 ± 6.2</td>
<td>115.8 ± 4.8</td>
<td>138.0 ± 6.1</td>
<td>153.2 ± 5.5</td>
<td>157.3 ± 6.7</td>
<td>158.2 ± 5.5</td>
<td>159.2 ± 6.4</td>
<td>158.3 ± 5.8</td>
</tr>
<tr>
<td><strong>Depth of papilla groove (µm)</strong></td>
<td>-</td>
<td>82.4 ± 3.3</td>
<td>128.2 ± 3.9</td>
<td>129.6 ± 4.2</td>
<td>131.8 ± 4.1</td>
<td>132.8 ± 4.5</td>
<td>132.5 ± 4.2</td>
<td>131.5 ± 6.2</td>
</tr>
<tr>
<td><strong>Width of papilla groove (µm)</strong></td>
<td>-</td>
<td>14.2 ± 0.7</td>
<td>19.5 ± 1.4</td>
<td>22.5 ± 1.5</td>
<td>24.4 ± 1.3</td>
<td>25.6 ± 1.1</td>
<td>25.6 ± 1.4</td>
<td>25.3 ± 1.5</td>
</tr>
<tr>
<td><strong>Length of taste bud (µm)</strong></td>
<td>-</td>
<td>34.0 ± 1.3</td>
<td>37.4 ± 0.5</td>
<td>39.2 ± 0.7</td>
<td>41.5 ± 0.8</td>
<td>41.9 ± 1.2</td>
<td>42.2 ± 0.9</td>
<td>41.9 ± 0.9</td>
</tr>
<tr>
<td><strong>Width of taste bud (µm)</strong></td>
<td>-</td>
<td>22.2 ± 0.8</td>
<td>25.0 ± 1.0</td>
<td>26.0 ± 0.6</td>
<td>26.3 ± 0.6</td>
<td>27.0 ± 0.9</td>
<td>27.3 ± 1.2</td>
<td>27.0 ± 1.1</td>
</tr>
</tbody>
</table>

**Discussion**

The number of foliate papillae ridges were reported to be 7-8 rows [13] in Japanese dormouse, 20 rows [14] in porcupines, 34 rows [5] in flying squirrels, 15-20 rows [12] in rabbits and 7-8 rows [19] and 4-5 rows [20] in mice. In the present study, the number of the rows of foliate papillae was shown to be 4-6.

HARADA et al. [8] detected that the size of the papillae of rats increased until the 4th week and at this time, the depth of foliate papillae grooves increased and reached 264 µm. KOBAYASHI [12] reported that the width of papillae ridges was 220 µm in rabbits and that the width of each groove was about 30 µm. ROYER and KINNAMON [20] observed that the length of papillae grooves was 150 µm. In the present study, the ridges of foliate papillae increased and reached 178 µm in length and 157 µm width. The depth of grooves (131 µm) and the width of the grooves (24 µm) were evaluated at day 90.

MILLER and SMITH [18], and SMITH and MILLER [21] have shown that there was no taste buds in newborn hamsters, immature taste buds were seen at day 2 while mature taste buds were evidenced at day 4. STATE et al., [22] stated that 2 days old albino rat had immature taste buds and that taste buds with pores (mature) were seen in 10 days old animals. In the former study, papillae grooves started to open at day 14 and took the best shape between the 14th and the 21st days. In our study, we observed that newborn animals have no taste buds, immature and developed taste buds and grooves being firstly seen at day 3 and day 8 days, respectively.

ROYER and KINNAMON [20] identified three different type cells of taste buds, dark, light and intermediate, in mice by the electron microscopic examination. STATE et al. [22] evidenced five different types of cells in the taste buds by light microscopic examination. In our study, only two type cells of taste buds, dark and light stained were evidenced in developed foliate papilla.

MILLER and CHAUDRY [16] stated that the length and width of foliate papillae taste buds of hamster were 100-125 µm and 50-80 µm, respectively. HARADA et al. [8] reported that the length of taste buds in rats was 38.9 µm at the second week of the age. In the present study, the mean length of taste buds was 27.28 µm, i.e. values lower than that observed in hamster and rats.

Regarding the structure of foliate papillae by scanning electron microscopic examination, papillae were seen to show several lamina on the postero-lateral surface of the tongue of calomys callosus mouse [24], few lamina with deep grooves in the same animals [25]. The papillae were
digitiform in cats but showed layers in rabbits [3], and several grooves which were rowed parallel to each other and vertical to the tongue axis [11]. In the present study, foliate papillae which consisted from 4-6 rows of ridges and grooves on the postero-lateral sides of the tongue were detected.

It has been reported that the borders of surface cells were clear [23], micro-ridges were seen on the cell surfaces [1, 24] and became functional during the first 8 days of the postnatal period. No differences in the surface structure of the papillae and taste pores were opened to the deep grooves between papillae layers on the foliate papillae surface [2]. In this study, likewise the other workers, it was detected that the borders of the epithelial cells on the papillae were clear and that there were many micro-ridges but no taste pores that opened to the ridges of papillae. Our results appear to be consistent with those of AGUNGPRIYONO et al. [1] and UTIYAMA et al. [24].

In conclusion, it was detected that foliate papillae developed at day 90 and that taste buds in the foliate papillae developed and became functional during the first 8 days of the postnatal period. No differences in the surface structure of the papillae were seen from the birth to the maturity.

References