Histol Histopath (1990) 5: 169-172

Histology and Histopathology

Taste bud density in circumvallate and fungiform papillae of the bovine tongue

Kaberi Dasgupta, Amreek Singh and William P. Ireland
Departments of Biology and of Anatomy and Physiology, University of Prince Edward Island, Charlottetown, PE, Canada

Summary. Taste bud quantitation may provide useful parameters for interspecies comparisons of the gustatory system. The present study is a morphometric analysis of bovine taste papillae. Circumvallate and fungiform papillae from six bovine tongues were serially sectioned and, following staining, analyzed. Circumvallate papillae were found to have a mean volume of 3.66 ± 2.82 mm^3, a mean number of taste buds per papilla of 445 ± 279, and a mean taste bud density of 155 ± 112 buds/mm^3. Values for lateral fungiform papillae for the same three parameters were 0.384 ± 0.184 mm^3, 13.2 ± 13.4, and 40.8 ± 46.6 buds/mm^3, respectively. Values for dorsal fungiform papillae were 0.438 ± 0.246 mm^3, 4.39 ± 4.78, and 14.0 ± 17.1 buds/mm^3, respectively. Circumvallate papillae were found to have a significantly greater volume, number of taste buds per papilla, and taste bud density than either type of fungiform papilla. These data should serve as background for biochemical, endocrinological, or neurological studies involving the bovine tongue.

Key words: Taste bud, Circumvallate papillae, Fungiform papillae, Bovine tongue

Introduction

Taste buds are the first structures directly involved in the gustatory process. Consequently, the morphology of taste buds and associated structures has evoked much interest. Many histological investigations have examined lingual papillae and their various components (Chamorro et al., 1986, 1987; Satoh and Selkirk, 1988; Holland et al., 1989). Others have considered taste buds alone (Kinnamon et al., 1985; Murray, 1986; Cottler-Fox et al., 1987). These studies provide insight into the gustatory process and a basis for comparison of lingual papillae between species. Another approach is the quantitation of taste buds in a given species. Work along these lines has been done in the human (Miller, 1986, 1988), the monkey (Bradley et al., 1985), and the pig (Irving et al., 1988). Relatively little attention has been devoted to the bovine tongue (Davies et al., 1979).

The cow tongue is a plentiful source of taste buds for biochemical study. It may also serve as a useful model in endocrinological and neurobiological investigations. The present investigation seeks to provide quantitative information about taste buds present on circumvallate and fungiform papillae of the bovine tongue through determination of taste bud density for these papilla types.

Offprint requests to: Dr. Amreek Singh, Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, PE, Canada C1A 4P3

Materials and methods

Six tongues from adult cows were obtained from the local abbattoir and immediately fixed in 10% buffered neutral formalin. Three circumvallate, three lateral fungiform, and three dorsal fungiform papillae were removed from each of the six tongues and stored in the fixative. These papillae were later processed by a mechanical tissue processor (Histomatic Tissue Processor™, Model 166MP, Fischer Scientific, Instrumentation Division, Pittsburgh, PA 15219), and serial sections were made from each of the 54 blocks. Forty blocks were sectioned at 15 μm and 14 were sectioned at 8 μm. Sections were subsequently stained with hematoxylin and eosin using an automatic stainer (Varistain™ 24-3, Shandon, Sewickley, PA 15143). Photographs were taken with a Zeiss Photomicroscope III on Kodak technical Pan film.

Determination of numbers of taste buds contained in each papilla was made through direct counts using a light microscope. Taste buds were traced through serial sections in order to avoid counting the same taste bud more than once. Such tracings were rendered somewhat

difficult, however, for circumvallate papillae because of the large number of taste buds present, making confusion between taste buds more probable. Consequently, in order to avoid counting the same taste bud on a given circumvallate papilla more than once, only taste buds on every fourth section were counted for 15-micrometre sections and every seventh section for 8-micromtre sections. In some cases, individual taste buds were traced backwards and forwards through the omitted sections in order to confirm their identification.

Stereology. Volume of each papilla was determined using Bioquant (Bioquant™ System IV. (1985). R & M Biometrics, Inc. Nashville, TN 37209). Images of every fourth section of each papilla were projected on a video monitor and outlines were traced on a digitizing pad. Areas bounded by the tracings were ascertained by Bioquant™. Cavalieri's direct estimator of volume from sections (Gundersen et al., 1988) was employed in calculating the volume: areas from the sections of a given papilla were summed and multiplied by the thickness between two consecutively-traced sections.

Taste bud density was calculated by dividing number of taste buds per papilla by volume of that papilla. Analysis of variance was performed on papillary volume, taste bud counts per papilla, and taste bud density using the GLM subprogram of SAS Institute Inc. (SAS/STAT Guide for Personal Computers, Version 6 Edition, Cary, NC: SAS Institute Inc., 1985, 378 pp). Significance of differences between circumvallate, lateral fungiform, and dorsal fungiform papillae in terms of papillary volume, taste bud counts per papilla, and taste bud density were determined using unpaired two-tailed Student *t* tests (Steel and Torrie, 1980).

Results

Figure 1 illustrates typical taste buds present on bovine circumvallate and fungiform papillae. As indicated in Table 1, circumvallate papillae had a mean volume ± s.d. of 3.66 ±2.82 mm^3, while those of lateral and dorsal fungiform papillae were 0.384 ± 0.184 mm^3, and 0.438 ± 0.246 mm^3, respectively. Mean taste bud number per papilla ± s.d. of circumvallate papillae was 445 ± 279, that of lateral fungiform papillae was 13.2 ± 13.4, and that of dorsal fungiform papillae was 4.39 ± 4.78. Mean taste bud densities ± s.d. of circumvallate, lateral fungiform, and dorsal fungiform papillae were 155 ± 112 buds/mm^3, 40.8 ± 46.6 buds/mm^3, and 14.0 ± 17.1 buds/mm^3, respectively.

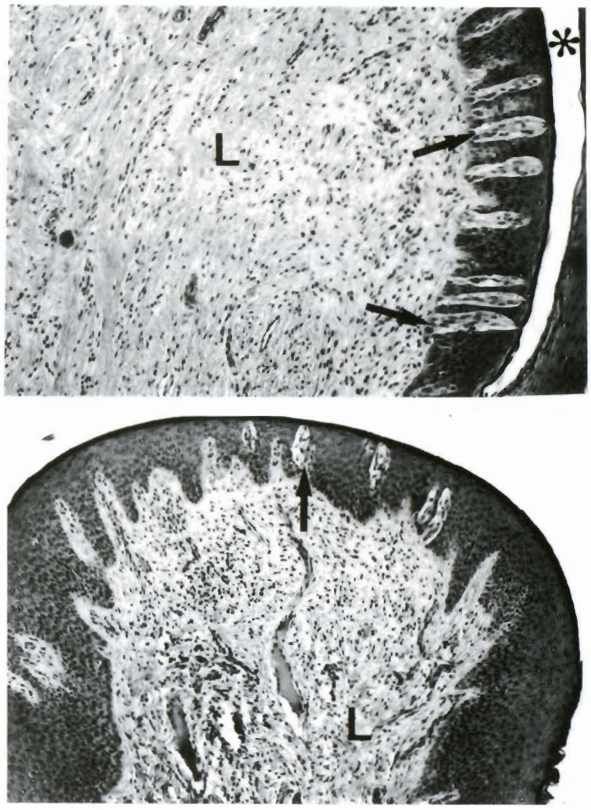

Fig. 1. Photomicrographs of portion of circumvallate (top) and fungiform (bottom) papillae from bovine tongue, demonstrating taste buds (arrows). Asterisk marks a vallum. L-lamina propria. H.E. Top × 100, bottom × 80

Circumvallate papillae were found to be significantly larger than both lateral fungiform ($t=3.56$, $p< 0.01$, $d<10$) and dorsal fungiform papillae ($t=5.01$, $p<0.001$, $df = 10$). Circumvallate papillae also had more taste buds per papilla than lateral fungiform ($t= 5.01$, $p< 0.01$, $df= 10$) and dorsal fungiform papillae ($t= 5.12$, $p < 0.001$, $df = 10$). Finally, circumvallate papillae had a greater taste bud density than either lateral fungiform ($t = 3.76$, $p < 0.01$, $df = 10$) or dorsal fungiform papillae ($t = 4.64$, $p < 0.001$, $df = 10$. No significant diferences were found between lateral and dorsal fungiform papillae.

Discussion

Traditional classifications of papillae refer to six papilla types: filiform, fungiform, circumvallate, foliate,

Table 1. Mean ± SD volume, taste bud number, and taste bud density of bovine circumvallate, lateral fungiform, and dorsal fungiform papillae.

PAPILLA TYPE	VOLUME (mm^3)	TASTE BUD NUMBER (buds/papilla)	TASTE BUD DENSITY (buds/mm^3)
CIRCUMVALLATE	3.66 ± 2.82	445 ± 279	155 ± 112
FUNGIFORM			
lateral	0.384 ± 184	13.2 ± 13.4	40.8 ± 46.6
dorsal	0.438 ± 0.246	4.39 ± 4.78	14.0 ± 17.1

conical, and lenticular (Stinson and Calhoun, 1987). The bovine tongue, however, does not possess any foliate papillae, and, as is also true for other species, its filiform, conical, and lenticular papillae do not contain taste buds. Consequently, all taste buds of the tongue in bovines are present in circumvallate and fungiform papillae.

Our results indicate that bovine circumvallate papillae are significantly larger, comprising a greater volume than either lateral fungiform or dorsal fungiform papillae, which do not differ significantly from one another. Irving et al. (1988) determined that porcine circumvallate papillae were larger than either porcine fungiform papilla type, as found in the present study of the bovine tongue, but that porcine lateral fungiform papillae were, in turn, significantly larger than their dorsal counterparts.

Davies et al. (1979) reported that the number of taste buds per circumvallate papilla in bovines varied greatly, maximum found on single taste bud being 1786, with an average of 612; standard deviation values were not provided. In the present study there was variation in the number of taste buds per circumvallate papilla although this was less than the variation for the number of taste buds per lateral or dorsal fungiform papilla. The maximum number of taste buds recorded on a single circumvallate papilla was 1094, with a mean of 445 ± 279. It should be noted that we considered a total of 18 circumvallate papillae derived from six tongues while Davies et al. (1979) examined a total of 54 circumvallate papillae derived from two tongues.

Trautmann and Fiebiger (1957) stated that taste buds on bovine fungiform papillae were sparse. Davies et al. (1979), however, noted a range of 1 to 15 taste buds per fungiform papilla, with a mean of 6.8 buds per fungiform papilla on the first tongue and 8.9 on the second tongue. In our study, a much greater range of values for number of taste buds per fungiform papilla was found (0 to 56). This may be partly explained by the fact that a greater number of animals were used than in the study of Davies et al. (1979). However, in the present study, the mean number of taste buds per papilla was 13.2 ± 13.4 for the lateral fungiform papillae and 4.39 ±4.78 for the dorsal fungiform papillae. The mean for both fungiform types combined was 8.8 taste buds per fungiform papilla, a value in agreement with that of Davies et al. (1979).

In the porcine tongue, lateral fungiform papillae have a greater number of taste buds per papilla and a greater taste bud density than dorsal fungiform papillae (Irving et al., 1988; Singh and Ireland, 1988). The data in Table 1 suggests that this may also be true for lateral and dorsal fungiform papillae in the cow tongue. Nonetheless, *t* tests have indicated that these differences are not significant; this lack of significance may be a result of the large error variance that was observed.

Large error variances have been found in other studies concerned with taste bud quantitation (Miller, 1986, 1988). It has been suggested that such variability reflects age-dependent differences in taste bud number. However, Miller (1988) discovered a more than a 100-fold range in taste bud density to be evenly distributed among age groups and sexes in the human tongue. In their study of rat tongues, Mistretta and Oakley (1986) found significant differences in the average percentages, per age group, of papillae that contained a taste bud; in reality, however, few taste buds were lost in the oldest rats. Other studies have indicated that there is no age-related difference in numbers of taste buds in humans (Arvidson, 1979), rhesus monkeys (Bradley et al., 1985), and Wistar-derived rats (Mistretta and Baum, 1984).

Irving et al. (1988) found no significant difference between taste bud density of porcine lateral fungiform and circumvallate papillae though circumvallate papillae were significantly larger than lateral fungiform papillae and had a significantly greater number of taste buds per papilla. It appears that the greater number of taste buds in a porcine circumvallate papilla relative to that of a lateral fungiform papilla corresponds to the former's greater volume.

Bovine circumvallate papillae are significantly larger and have significantly more taste buds per papilla than either lateral or dorsal fungiform papillae, but they also have a greater taste bud density than either fungiform papilla type. Similarly, porcine lateral fungiform papillae are larger, have more taste buds per papilla, and a greater taste bud density than dorsal fungiform papillae. Thus, in both of these instances, the number of taste buds of the larger papillae in question is greater than that increase which would be expected to accompany an increase in volume relative to the smaller papillae in question.

Taste buds may be used by investigators concerned with the gustatory process in particular or membrane chemosensitivity in general (Roper, 1989). The present work on morphometric analysis of cow taste papillae, we believe, will provide a firm basis for biochemical, endocrinological, or neurological studies employing the bovine tongue.

Acknowledgements. The authors are grateful to Ms. Susan Hughson for preparing most of the serial sections, and to Professor Robert Campbell and Dr. James Amend for critically reviewing the typescript. We would also like to thank the Natural Sciences and Engineering Research Council for awarding K. Dasgupta an Undergraduate Summer Research Studentship.

References

Arvidson K. (1979). Location and variation in number of taste buds in human fungiform papillae. Scan. J. Dent. Res. 87, 435-442.

Bradley R.M., Stedman H.M. and Mistretta C.M. (1985). Age does not affect numbers of taste buds and papillae in adult rhesus monkeys. Anat. Rec. 212, 246-249.

Chamorro C.A., de Paz P., Sandoval J. and Fernández J.G. (1986). Comparative scanning electron-microscopic study of the lingual papillae in two species of domestic mammals *(Equus caballus* and *Bos taurus)* 1. Gustatory Papillae. Acta Anat. 125, 83-87.

Chamorro C.A., Sandoval J., Fernández J.G., Fernández M. and de Paz P. (1987). Compared studies of the lingual papillae of the cat *(Felis catus)* and the rabbit *(Oryctolagus cunuculus)* by

scanning electron microscopy. Anat. Histol. Embryol. 16, 37-47.

Cottler-Fox M., Arvidson K. and Hammarlaund E. (1987). Fixation and occurrence of dark and light cells in taste buds of fungiform papillae. Scand. J. Dent. Res. 95, 417-427.

Davies R.O., Kare M.R. and Kagan R.H. (1979). Distribution of taste buds on fungiform and circumvallate papillae of bovine tongue. Anat. Rec. 195, 443-446.

Gundersen H.J.G., Bendtsen T.F., Korbo L., Marcussen N., Moller A., Nielsen K., Nyengaard J.R., Pakkenberg B., Sorensen F.B., Vesterby A. and West M.J. (1988). Some new, simple and efficient stereological methods and their use in pathological research and diagnosis. Acta Pathol. Microbiol. Immunol. Scand. Suppl. 96, 379-394.

Holland V.F., Zampighi G.A. and Simon S.A. (1989). Morphology of fungiform papillae in canine lingual epithelium: location of intercellular junctions in the epithelium. J. Comp. Neurol. 279, 13-27.

Irving A., Ireland W.P. and Singh A. (1988). Analysis of taste buds distribution on porcine tongue. VIII International Symposium on Morphological Sciences, Rome, Italy, Abstracts p 190.

Kinnamon J.C., Taylor B.J., Delay R.J. and Roper S.D. (1985). Ultrastructure of mouse vallate taste buds: I. Taste cells and their associated synapses. J. Comp. Neurol. 235, 48-60.

Miller I.J. Jr. (1986). Variation in human fungiform taste bud densities among regions and subjects. Anat. Rec. 216, 474-482.

Miller I.J. Jr. (1988). Human taste bud density across adult age groups. J. Gerontol. 43, B26-B30.

Mistretta C.M. and Baum B.J. (1984). Quantitative study of taste buds in fungiform and circumvallate papillae of young and aged rats. J. Anat. 138, 323-332.

Mistretta C.M. and Oakley I.A. (1986). Quantitative anatomical study of taste buds in fungiform papillae of young and old Fischer rats. J. Gerontol. 41, 315-318.

Murray R.G. (1986). The mammalian taste bud type III cell: a critical analysis. J. Ultrastruct. Mol. Struct. Res. 95, 175-188.

Roper S.D. (1989). The cell biology of vertebrate taste receptors. Ann. Rev. Neurosci. 12, 329-358.

Satoh Y. and Selkirk L.W. (1988). Taste threshold, anatomical form of fungiform papillae and aging in humans. J. Nihon Univ. Sch. Dent. 30, 22-29.

Singh A. and Ireland W. (1988). Stereology of the porcine taste buds. Anat. Histol. Embryol. 17, 376.

Steel R.G.D. and Torrie J.H. (1980). Principles and Procedures of Statistics. A Biometrical Approach. 2nd ed. McGraw-Hill Book Company. New York.

Stinson A.W. and Calhoun M.L. (1987). Digestive System (Tongue). In: Textbook of Veterinary Histology. 3rd ed. Dellmann H.-D. and Brown E.M. (eds). Lea & Febiger. Philadelphia. pp 212-215.

Trautmann A. and Fiebiger J. (1957). The Digestive System (Tongue). In: Fundamentals of the Histology of Domestic Animals. Translated and revised from 8th and 9th German editions by Habel R.E. and Biberstein E.L. Comstock Publishing. Ithaca. New York. pp 164-169.

Accepted October 26, 1989