

A COMPARATIVE STUDY OF THE DISTRIBUTION OF GEMMA  
GUSTATORIA IN THE DOG, CAT, SHEEP AND CHICKEN

by

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## INTRODUCTION

Animals live in a chemical world and depend on chemical senses for nutrition, reproduction, mating and many other bodily functions. Chemical senses include taste, smell and so-called common sensitivity. The gustatory and olfactory receptors usually act as monitors during the intake of food into the oral cavity and air into the nasal cavity. There are many other chemoreceptors within the organism. There is considerable overlap of gustation and olfaction since the sense of smell combines with the sense of taste in many taste sensations.

Vision and hearing lie at the top of the sensory hierarchy in richness of cognitive detail, variety of sensory experience and significance for perception. The chemical senses are classed among lower senses perhaps because of simplicity of morphology and relative paucity of information conveyed. In man taste stimulation is associated with the sensation qualities of salty, sour, bitter and sweet and one system reduces odors to four elementary classes; namely, fragrant or sweet, acid or sour, burnt or empyreumatic and caprylic or goaty.

Unlike olfaction, which may have a variety of functions including mate selection and the avoidance of predators, the function of taste is more limited. It has a role in the regulation of ingestion of nutrients and possibly the avoidance of toxic substances. The taste system in a species would be adapted through the evolutionary process to the species metabolic and dietary requirements.

Gustation, the sense of taste, is of interest because it is sensitive to different ions and molecules. The molecular basis of chemical sensitivity in the taste cell and how the selectivity gives rise to

afferent neural codes in the taste nerves has been the subject of much study. Gustation may control ingestive behavior as some tastes, like sugars, are accepted avidly by most creatures, others like quinine, are rejected while still others are relatively neutral. It initiates specific hungers of intense cravings for particular substances and contributes in some measure to the "wisdom of the body."

Naturalist observations are replete with examples of the craving for salt shown by animals in the wild. Herbivorous animals are known to travel great distances in search of a salt lick. Certain mineral deficiencies in forage grown in depleted soil may lead to other aberrations of taste preference.

Richter (1943) demonstrated that the sense of taste played a critical role in nutrition and in maintaining a constant internal environment for the animal. He demonstrated that the adrenalectomized rat, when given a choice, drank enough salt water to maintain life and growth. Similarly the parathyroidectomized animal displayed an increased appetite for food containing calcium. Such animals, desensitized by surgical removal of the taste nerves, could not make the necessary discrimination and failed to survive.

The study of the chemoreceptors presented an excellent method to determine the interaction of a chemical stimulus with biological cells. The great chemical specificity shown by taste receptors presented an interesting problem of molecular interactions with biological systems. How taste receptors differentiate and respond differently to dextro and levo amino acids is an intriguing problem since most biological systems respond to only one form.

## REVIEW OF LITERATURE

The first person who described lingual papillae as reviewed by Arey (1935) was Malpighi (1664) but he did not recognize taste buds. Peculiar bud shaped epithelia complexes were discovered in several teleosts by Leydig (1851). Schulze (1883) thoroughly examined the inner structure of these organs and differentiated two cell types, the supporting and sensory cells. It remained for Schwalbe (1868) and Loven (1868) to discover independently similar formations in the circumvallate papilla of the mammalian tongue and identify them as organs of taste.

Sisson and Grossman (1953) described the dog's tongue as marked by a median groove and thickly covered with papillae. The short, pointed, filiform papillae had free ends directed backward. On the root of the tongue were long conical papillae. The fungiform papillae were small and were scattered over the dorsum and sides of the tongue but were absent on the posterior part of the dorsum and on the root, where conical papillae occur. There were two or three vallate papillae on either side on the posterior part of the dorsum, where the conical papillae begin. Small foliate papillae were also present in front of the anterior pillars of the soft palate; they were oval and crossed by half a dozen fine fissures.

Bowne and Getty (1964) recognized five types of papillae in the dog based on the shape: filiform, fungiform, vallate, foliate and conical.

The term "filiform" or thread-shaped was really a misnomer when considering the papilla as a whole. They were distributed on the anterior two-thirds of the tongue. There was an average of four tertiary

filiform papillae on the anterior border of filiform papillae proper. The dermal cores did not extend into the keratinized papillae very far. There were two secondary filiform papillae on each side of the large filiform papilla and the former had a well-developed dermal core supporting the keratinized, stratified squamous epithelium. There were no taste buds on this papillae.

The fungiform papillae were located on the anterior two-thirds of the tongue among filiform papillae. Not all fungiform papillae had taste buds associated with them but when present, the taste buds were located on the dorsal surface.

The foliate papillae were leaf-like and were in two groups with eight to twelve papillae in each group. They were located on the dorso-lateral aspect of the tongue anterior to the anterior pillar of the fauces. Grossly, the papillae were arranged like the petals of a flower. Crypts separated the papillae and taste buds were located on the sides of the papillae facing the crypts.

The vallate (circumvallate) papillae number four to six in the dog. They were located at the junction of the anterior two-thirds with the posterior third of the tongue. They were arranged in the form of a "v" on the dorsal surface of the tongue, the apex of "v" directed posteriorly. They had a deep moat or trench around them. Taste buds were located on the sides of the vallate papilla, most numerous at the base of the papilla and decreasing in number towards the surface. The wall of the moat contained few taste buds at birth which became more numerous with the age of the animal.

The conical papillae were structurally similar to the primary

filiform papillae. The papillae just anterior to the vallate were designated as filiform and those posteriorly as the conical. The stratified squamous epithelium was thick and the outer layers were heavily cornified. The top of the papilla was very thin, heavily cornified and directed posteriorly. Taste buds were absent on these papillae.

Taste buds were located on the dorsal surfaces of the fungiform papillae, on the sides and dorsal surface of the vallate papilla, as well as the bottom and the sides of the surrounding moat. On the foliate papillae they were on the epithelial surface facing the moat and on the free surfaces of the papillae.

The facial nerve supplied the second branchial arch. Fibers of the chorda tympani branch of the facial nerve traced from the geniculate ganglion, through the middle ear to its junction with the lingual branch of the mandibular nerve. Sensory innervation to the taste buds located on the fungiform papillae in the anterior two-thirds of tongue was by the chorda tympani nerve. There was a crossing over of nerves or a dual innervation of taste buds on the fungiform papillae close to the lingual groove.

The sensory portion of the glossopharyngeal nerve passed to the second and third branchial arches and innervated the root of tongue and the pharynx. Vallate and foliate papillae were innervated by this nerve. The same nerve innervated the soft palate. In rare instances taste buds have been demonstrated on the soft palate of dog.

The vagus nerve supplied the fourth branchial arch, which formed most of the epiglottis and larynx. This nerve provided the sensory innervation to the taste buds in the region of the epiglottis and larynx.

Bradley (1959) stated that filiform papillae were small anteriorly becoming larger posteriorly, finally blending with conical papillae behind the "V" shaped rows of vallate papillae. He described the location of the fungiform papillae as being similar to the distribution of filiform papillae but found none mixed with the conical papillae. He recognized the two foliate papillae anterior to the glossopalatine arch on the lateral edge of the mucosa of the tongue. The vallate papillae were four in number but sometimes five or six.

Trautmann and Fiebiger (1952) described the filiform papillae as having mechanical function only and consisting of a connective tissue core derived from the lamina propria and covered with keratinized epithelium. In carnivores the connective tissue core extended above the surface epithelium and had papilla of unequal sizes. The caudal-most of the filiform papillae were large and most distinct in the cat. A thick caudally directed horny tooth was described on the caudal-most papillae. Large conical papillae, whose core projected beyond the surface of the tongue, occurred in all domestic mammals except the horse and donkey.

The fungiform papillae had a connective tissue core rich in nerves and had a soft epithelium containing taste buds. Taste buds were sparse in cattle and horses; more numerous in sheep and swine; very abundant in carnivores and especially abundant in the goat. The taste buds of the cat and the dog were conspicuous for their size.

The circumvallate papillae project above the lingual epithelium only slightly or not at all. The epithelial surface facing the moat contained many taste buds. These papillae had the most taste buds in

swine and dog and fewest in cats. In carnivores taste buds were restricted to the bottom of the moat. Taste buds were absent in the surface epithelium and the peripheral wall of the moat.

The foliate papilla consisted of a series of parallel connective tissue leaves bearing secondary papillae that projected into the covering squamous epithelium. The gustatory furrows separated the leaves and serous glands opened into the gustatory furrows. These glands were abundant in horse and dog in which mucous glands were also present. Foliate papillae were absent in ruminants and rudimentary and without taste buds in the cat.

Taste buds were found in the epithelium of the fungiform, foliate and circumvallate papillae of the tongue and also found widely separated in the soft palate, the epiglottis and the free edge of the vocal folds in domestic animals in general (Trautmann and Fiebiger, 1952).

McLeod (1958) described the dorsal surface of the bovine tongue as crossed a little behind the middle of its length by the deep transverse lingual groove. Anterior to the lingual groove were horny filiform or conical papillae, and posterior to the groove on the elliptical prominence were lenticular papillae. None of the three types of papillae had taste buds. Taste buds were found on the fungiform papillae, which were located on the dorsum anterior to the groove and along the edges of the free part of the tongue, and on the vallate papillae. The vallate papillae were surrounded by a circular trench and located on the posterior part of the dorsum. Unlike many other species, the bovine does not have foliate papillae.

Parker (1922) stated that there may be four sources of innervation



to the taste buds, namely: the vagus, glossopharyngeal, facial and trigeminal nerves. He stated that the vagus was distributed to the larynx, epiglottis and posterior part of the tongue and the glossopharyngeal nerve supplied the posterior third of the tongue including the foliate and vallate papillae. He had some doubts about the innervation of the anterior two-thirds of the tongue and mentioned the trigeminal, facial and glossopharyngeal nerves as possibly communicating through a plexus of fine branches near their roots.

Cushing (1903) concluded that when the gasserian ganglion was extirpated in the human, the sensation of taste returned to the anterior two-thirds of the tongue after some time. He assumed that the chorda tympani nerve was left intact. He also believed that the degenerating trigeminal fibers had an adverse effect on the chorda tympani fibers and thus caused a temporary loss of taste impulse over the chorda tympani nerve.

Olmstead (1921) cut the lingual nerve in the dog and taste buds disappeared from the fungiform papillae on the anterior two-thirds of the tongue. Taste buds on the operated side were affected and those on the other side remained in perfect order. The taste buds disappeared by degenerative process with the aid of phagocytic leucocytes and not by dedifferentiation or metamorphosis. Epithelial cells took the place of the former taste buds and during the process there was marked proliferation by mitosis from the germinative layer.

Olmstead (1922), using two dogs, severed the chorda tympani branch of facial nerve just cranial to its junction with the lingual nerve and within eight days observed the disintegration of taste buds

and removal of the same by phagocytic action of leucocytes. All papillae which were examined from the operated side were lacking in taste buds. Taste buds were present on an equal number of papillae on the unoperated side. He also cut the mandibular nerve just cranial to the junction of the chorda tympani nerve with the lingual nerve, but it had no effect on the appearance of the taste buds on the operated side. The animal had difficulty in drinking and the left side of the tongue was paralyzed.

Guth (1957), by bilateral glossopharyngeal nerve transection, observed no significant alteration in the number of taste buds on the first post-operative day. Thereafter the number of taste buds diminished rapidly for three days and after the fourth post-operative day taste buds continued to disappear but at a lower rate. All of them had disappeared by the seventh post-operative day and were replaced by stratified squamous epithelium.

Guth (1958) observed all but a small number of taste buds on the ipsilateral circumvallate papillae of the cat disappeared after unilateral transection of the glossopharyngeal nerve. Reappearance of the taste buds occurred upon reinnervation of the papilla by either the glossopharyngeal or vagus nerve. However, reinnervation by the hypoglossal motor nerve did not induce the formation of taste buds. In higher vertebrates sensory nerve fibers not only induced epithelial cells to form taste cells but they maintained the integrity of the taste organ. Such may not be the case in lower forms, such as newt, where taste buds grafted to the liver or placed in the orbit of the eye maintained their integrity even though sensory nerves were not present (Wright, 1951; 1958). The exact nature of the influences from the

sensory nerves which result in the regeneration of the taste buds is not known, but it does not appear to be related to alterations in blood flow (Guth, 1963).

Foley (1945) showed that the chorda tympani nerve of the cat contained myelinated and unmyelinated nerve fibers ranging from 1.5 to 6 microns in diameter while the unmyelinated axons were less than 1.5 microns across. He further showed that intact nerves of the cat contained an average of 1955 combined sensory and motor axons, while in the chorda tympani of the dog a total of 3347 sensory and motor nerve fibers were found. Table 1 gives the types of fibers in each nerve.

Table 1. Sensory and motor axons in the chorda tympani of cat and dog (Foley, 1945).

Chorda tympani of	Cat	Dog
Total axons	1955	3347
Motor axons	798	1142
Myelinated	639	1073
Unmyelinated	159	69
Sensory axons	1157	2205
Myelinated	949	1698
Unmyelinated	208	507

Van Buskirk (1945) estimated that there were 1034 myelinated fibers in the chorda tympani nerve in cats and 1696 fibers in the dog. According to the same author, in man the facial nerve contained 7000 fibers, of which three-quarters were myelinated and most of them were relatively thick 7-10 microns.

Kitchell (1963) made comparative anatomical studies of the chorda tympani nerve in domestic animals and observed that the course of the nerve in cattle was aberrant. It may join the lingual nerve distal to the rostral border of the medial pterygoid muscle.

He further observed the chorda tympani nerves of dogs and cats were not fasciculated, whereas those of other domestic animals were fasciculated. Isolation of fibers in non-fasciculated was easy since the quantity of connective tissue was less. After a detailed study he arrived at the figures given in Table 2.

Table 2. Myelinated fibers in the chorda tympani nerve (Kitchell, 1963).

Animal	Average no. fibers	Fibers less than 6 microns	Mode diameter in microns	%total fibers in mode group
Cat	1555	81	4-6	43
Dog	2854	40	6-8	40
Goat	3321	49	6-8	31
Sheep	3423	55	4-6:6-8	32:32
Pig	4366	53	4-6	27
Cow	5265	35	6-8	31
Horse	5735	37	6-8	38

From Table 2 it may be observed that species with greater body mass have more myelinated chorda tympani fibers of larger diameter, a notable exception was dog whose 40% of fibers were less than 6 microns in diameter and 3% fibers larger than 10 microns in diameter.

Trautman and Fiebiger (1952) described the microscopic structure

of a taste bud as made up of supporting cells and neuroepithelial cells. The outer layer was formed by the peripheral supporting cells which were narrow cells with ellipsoid nuclei. The cells tapered distally to a fine point and the proximal end was blunt. In the interior of the bud the central supporting cells were shorter and straighter. In some species the taste bud contained basal supporting cells. Among the supporting cells were six neuroepithelial cells which were slender cells, thickened slightly in the region of nucleus and with a characteristic hairlike cuticular rod on the distal extremity.

Trautman and Fiebiger (1952) described the taste organs of the horse as melon-shaped; sheep and ox as close-set ovoid buds; and swine as spindle-shaped. The taste buds of the goat were small and irregularly ellipsoid. The dog possessed minute spherical taste buds; cat had few and poorly defined.

DeLorenzo (1958) examined the fine structures of the taste buds in the rabbit with phase contrast and electron microscopy. The bud contained two types of cell: the gustatory receptors and the sustentacular cells. The receptors were characterized by a dark nucleus and densely granular cytoplasm. The apical process bore numerous microvilli which extended into the taste pore. Imbedded between the microvilli was a dense substance. The sustentacular cells contained a large pale nucleus and less dense cytoplasm. The basal surfaces rested on a basement membrane.

The sub-epithelial nerve plexus comprised the fibers which innervated the gustatory receptors. The nerve fibers were ensheathed by Schwann cells and their diameters ranged from 500  $\text{A}^\circ$  to 0.3 micron. The

intragemmal fibers entered the taste bud between adjacent cells and were ensheathed by the plasma membranes of the supporting cell until they synapsed upon the gustatory cell. The synaptic terminals contain synaptic vesicles.

Murry and Murry (1960) observed the fine structure of the taste buds of Rhesus and Cynomolgus monkey and no evidence of two distinct sustentacular type cells was found. These observations were at variance with the generally accepted view (Ham and Loeson, 1961). Although De Lorenzo (1958) supported two cell types, Engstrom and Rytzer (1958) were not certain about two distinct types. No desmosomes were present between the taste cells and the surrounding epithelia. The nuclei of the gustatory cells had dense nuclear membranes and did not show double contours. The nuclei were sharply defined and showed distinctive internal structure. Mitochondria were numerous in gustatory cells, were widely distributed and showed well defined cristae. The Golgi complex was not numerous or extensive. Dense spherical bodies were scattered through the cytoplasm but were most numerous in the apical parts. Concentric bodies 500-3000Å in size and having an amorphous or densely granular core surrounded by many layers of clearly defined membranes in concentric array were in close association with the nucleus of the gustatory cell in monkeys. No taste hairs or cilia were found and microvilli, though present, were not so prominent as in the rabbit.

Murry and Murry (1960) observed two types of nerve endings within and around the taste buds, namely intragemmal and perigemmal, but no intergemmal endings. The nerve fibers never branched after entering the taste bud. The nerve endings had empty axoplasm and a scattering of

vesicles of 200-600 A° in size. Motor synapses on the ends of efferent fibers were also commonly found.

DeLorenzo (1963), from electron microscopic studies, confirmed the absence of sustentacular cells in the mammalian taste bud. He observed that many of the cells were transitional and represented different stages of the same basic cell type. In addition various degenerating cells have been observed in the taste bud. About six receptor cells were seen near the region of the taste pore. The apical tips demonstrated cell surface specialization--microvilli--which were extensions of the plasma membrane. Microvilli were 0.1 micron to 0.2 micron wide and 2.0 micron or more long. There were no taste hairs and the structures seen in the light microscope appeared to be artefact. Cytoplasm had large clusters of vesicles and fibrillar material. Between the cells in the region of the taste pore were large amounts of dense osmiophilic material. The apical cytoplasm contained accumulations of mitochondria and a large number of dense granules. Those granules were enclosed in membranes and resembled neurosecretory or "catechol" amine granules. The cells interdigitated and showed desmosomes. An endoplasmic reticulum, RNP granules, mitochondria and Golgi apparatus were the other cell-components.

DeLorenzo (1963) observed myelinated nerve fibers about 1-6 micron in diameter in the connective tissue underlying each taste bud. Myelin was lost in the nerve plexus near the basement membrane and unmyelinated fibers entered the bud and established synaptic contiguity with the receptors. Two kinds of nerve fibers one with a diameter of 0.5 micron to 1.0 micron and the other below 0.5 micron were

observed. Usually about thirty nerve fibers, consisting of both the large and small types, pushed into a single receptor cell. The large fibers terminated in or innervated two or more receptor cells and consistently showed synaptic vesicles. Smaller fibers terminated in individual receptor cells and did not contain large vesicles but contained a few mitochondria and punctate fibrils. Large fibers appeared as "free endings."

According to De Lorenzo, whenever two kinds of endings were seen in a receptor system the possibility of efferent ending should be considered. The "synaptic vesicles" were on the post-synaptic side of the synapse and that location suggested that they were efferent endings.

De Lorenzo (1963) also noted that the turnover of taste cells in the foliate papillae was rapid and labelled cells migrated into taste buds about one cell every thirty hours.

There was a discrepancy between the expected and the observed conduction velocities of the gustatory afferent fibers. For approximately 2000 myelinated fibers in the canine chorda tympani, Kitchell (1963) found that the most common diameter (40% of total) was six to eight microns. Using the common assumption that a multiplication factor of six existed between a fiber diameter and the conduction velocity, gustatory fibers were expected to have conduction velocities of 36 m/sec. (Hursh, 1939). The conduction velocities of chorda tympani gustatory afferent fibers in the dog, measured by Iriuchijima and Zotterman (1961), were found to range from 1.6 m/sec. to a maximum of 17.8 m/sec. If fibers larger than three microns in diameter reached the vicinity of the taste buds of the dog, these larger fibers could represent an



efferent pathway (De Lorenzo, 1963). The hypothetical efferent system would help explain the many synaptic vesicles reported at the termination of the large intragemmal fibers (De Lorenzo, 1963; Murry and Murry, 1960).

Esakov (1961) showed the influence of the interoreceptors of the stomach on the taste receptors in the frog. He observed that mechanical stimulation of the stomach (distension by filling with water) had a marked action on the character of the flow of afferent impulses from the receptors of the tongue. Stimulating the mechanoreceptors of the stomach (distension) led to an increase in the electrical activity of the receptors of the tongue (to water and salt); stimulation of the chemoreceptors (peptone) led to a decrease in the activity of the receptors of the tongue. This effect was reflex in nature, for it was abolished by the action of cocaine on the interoreceptors of the stomach and by extirpation of the medulla. The processes of the efferent control of the lingual chemoreceptors were affected by medulla; it had a feed back character and was meant to adjust the receptors to a definite level of activity in accordance with the requirements of external and internal environment of the organism.

This hypothesis also found support in the study of Zaiko and Lokshina (1962) who observed in man that the presence of food in the stomach appeared to alter the sensitivity of the fungiform papillae. With the stomach empty, the degree of mobilization of the taste receptors was high i.e., the taste sensitivity was acute. After food had been taken through a gastric fistula, without any stimulation of the taste receptors, the mobilization level was reduced. Stimulation of the gastric interoceptors produced a reflex alteration in the taste

apparatus.

Taste receptors themselves played a part in the change of taste sensitivity after food was taken. Before the food was introduced into the stomach through the fistula, it was chewed by the subject and, in this case, the reduction of taste sensitivity after the food had been introduced into the stomach was somewhat greater. In man, according to Zaiko and Lokshina, signals from the gastric receptors acting through the central nervous system played an important part in regulating the reaction of the gustatory apparatus.

Chernetski (1964) observed that the basis for the modifications of gustatory responses might be the sympathetic effects on gustatory neural responses in frog. Cathodal stimulation of the first sympathetic ganglion enhanced the ongoing gustatory activity from small branches of the glossopharyngeal nerve during application of gustatory stimuli to the frog's tongue. The delay between the sympathetic stimulus and the time of maximum enhancement of gustatory activity may be several seconds and was a linear function of the logarithm of the initial rate of response to the original gustatory stimulus.

The taste buds of the circumvallate papillae region of the posterior portion of the mammalian tongue were generally considered to be innervated only by the glossopharyngeal nerve. The chorda tympani may sometimes innervate taste buds in the circumvallate zone. Gustatory responses have been recorded by Bernard (1964) from the calf chorda tympani upon chemical stimulation of either the anterior or the posterior portion of the tongue.

Bernard (1964) studied the multiunit neural activity from the

gustatory nerves of twenty-eight calves following chemical and mechanical stimulation of the tongue. The chorda tympani nerve was found to have a posterior, in addition to the classical anterior, receptive field. The anterior field responded to NaCl and acetic acid starting at 0.001 M. and KCl and propionic acid starting at 0.01 M. Fructose, glucose, xylose and sucrose had higher thresholds and were less effective than equimolar concentrations of the salts and acids studied. Quinine sulfate, Na saccharin, distilled water and mechanical stimulation produced small responses. In contrast, the posterior field yielded large responses to distilled water and to mechanical stimulation. On the receptive field of the glossopharyngeal nerve mechanical stimulation was more effective than chemical stimulation, which produced small responses from high stimulus concentrations. This indicated the calf relies mainly on the chorda tympani for fine gustatory discrimination and uses the glossopharyngeal pathway for chemical discrimination at higher concentrations only.

There was no simple relationship between behavioral and multi-unit neural taste responses in the calf. Strong behavioral responses existed where little or no neural response was recorded as in the case of the sugars at the lower concentrations and for quinine. Weak behavioral responses, such as neither preference nor rejection, occurred with neural responses that were large for NaCl and small for Na Saccharin. Only in the case of the acids were behavioral and neural responses correlated and co-extensive. Thus, taste-dependent behavior could not be predicted on the basis of the multi-unit neural response nor could one assume that a measurable neural response would always accompany behavioral

responses. However, when neural responses exist there was a possibility of behavioral responses.

Bohm and Strang (1962) indicated in clinical data that the human glossopharyngeal nerve also may not be of great importance in taste. They suggested that the chorda tympani nerve also innervated the posterior part of human tongue and gustatory loss did not consistently occur following human glossopharyngeal nerve section.

Moon and Pullen (1963) reported that when the chorda tympani nerve was sectioned during middle ear surgery in humans, there was loss of taste sensation and probably loss of pain and touch to the anterior two-thirds of the tongue. When the chorda tympani nerve was sectioned the sub-maxillary and sub-lingual glands ceased to function and probably atrophied. The symptoms experienced by the patients was of a bitter and/or metallic taste and numbness localized to the same side of the tongue. Those symptoms occurred initially in 28 percent of the patients and persisted after six months in 17 percent of the patients. Two patients out of 262 reported mouth dryness as a result of elimination of the function of one maxillary and one sub-lingual gland.

Schwarz and Weddell (1938) observed that in some cases surgical section of the chorda tympani had not been followed by loss of the sense of taste in the anterior two-thirds of the tongue. It has been assumed that the taste fibers in these cases, after having followed the distal part of the chorda tympani, entered the greater superficial petrosal nerve via anastomoses near the otic ganglion.

Nelson (1953) observed that the facial nerve contained sensory fibers, whose cell-bodies lay within the geniculate ganglion of the

facial nerve. The sensory fibers which innervated the taste buds on the anterior two-thirds of the tongue in mammals were special visceral afferent fibers, coursing along the chorda tympani nerve. He described the sensory components of the glosso-pharyngeal nerve as being derived from neuron bodies within the superior and petrosal ganglia and that the portion that supplied the taste buds on the posterior third of tongue was functionally classified as special visceral afferent. The sensory components of the vagus nerve took their origin from neuroblasts of the neural crest which ultimately incorporated in the jugular and nodosal ganglia. The cell bodies of the special visceral fibers of the vagus were in the nodose ganglion.

Beidler (1954) proposed a theory of the mechanism of taste stimulation. An equation was developed from the mass action law, assuming adsorption of sapid molecules or ions by a finite set of equivalent, independent sites on the membrane of the gustatory receptor cell:

$$1. \quad K = \frac{Z}{(N-Z) C}$$

Where C was the concentration of the stimulus used, N was the total number of available receptor sites, Z was the number of receptor sites that have combined with the sapid chemical at concentration C, and K was the equilibrium constant. Neither N, the total number of sites, nor Z could be directly measured. However, it was assumed that the gustatory response R produced at concentration C would be proportional to Z, and that the maximum obtainable gustatory response  $R_s$  for the chemical involved, would be proportional to N. Inserting this information into Eq.1,

$$2. \quad K = \frac{R}{(R_s - R) C}$$

which could then be rearranged into

$$3. \quad \frac{C}{R} = \frac{C}{R_s} + \frac{1}{KR_s}$$

which was the fundamental taste equation of Beidler's theory. If the theory was correct, the plot of the experimental data in the form of  $C/R$  versus  $C$  should and did produce a straight line. The derived equation was similar to that of Langmuir adsorption isotherm or to many enzyme-substrate equations. The good agreement between data and theory did not necessarily mean that all the assumptions used in the theory were correct, but it did prove that the taste equation derived was sufficient to describe the experimental data. Beidler's concept was that the stimulus was adsorbed to the cell surface involving weak bonding of the reacting chemical to the site and not an enzymatic one.

Nejad (1961) proposed a multi-step gustatory receptor process, with only the initial step directly described by Beidler's theory. At least some of the steps were probably enzymatic. Several experiments indicated that sulfhydryl (SH) groups may be involved in a secondary step in the response of the gustatory receptor cell to sapid chemicals. In one such study a one-minute flow of the SH-group inhibitor  $HgCl_2$  over the rat tongue was found to decrease irreversibly the chorda tympani response to tongue stimulation with NaCl. Iodoacetic acid, which can combine with SH-groups, also irreversibly blocked responses to NaCl. The inhibition by iodoacetic acid did not occur if the tongue was first soaked in the SH-group supplier cysteine.

Kimura and Beidler (1961) observed, by using a micropipette electrode thrust into the cells of a single taste bud located on a fungiform papilla of the rat and hamster, slow potential changes of the

receptors elicited by a number of salts or several kinds of chemical stimuli. One taste cell responded to as many as four major taste substances: sucrose, NaCl, HCl or quinine, although the ratios of the responses to these substances were different in each cell. Each taste cell differed in overall sensitivity as well as its relative magnitude of response to series of select stimuli. Thus, highly specific taste receptors that respond to but one type of stimulus were not found.

Data obtained from microelectrode studies were similar to those derived from the measurements of the integrated response of the chorda tympani or single fiber data by Pfaffmann (1955).

Beidler (1953) introduced a quantitative measure of whole-nerve activity and a flow system for the tongue in his study of taste reception in the rat. He found that summated chorda tympani responses could be grouped into three general categories. The first comprised the responses to acids and salts, which rapidly gave rise to a large response. The next included the sugar responses, which developed more slowly and had a lower magnitude. Saccharin belonged to the first rather than second group. The third category included responses to quinine, strychnine etc., which were barely perceptible. These categories were thought to be correlated with the "four" human taste qualities, with the acid and salt combined into one. The lack of a large response to quinine and similar compounds was believed to be due to the mediation of most responses to "bitter" substances by the glossopharyngeal pathway. An extensive series of sodium and chloride salts was examined to obtain indirect indication of receptor properties.

In an examination of NaCl and KCl electrophysiological taste

responses in several species, Beidler, Fishman and Hardiman (1955) found that in carnivores--cat, dog and raccoon--the KCl response was greater than NaCl response; whereas the reverse was true for the rodents--rat, hamster and guinea pig. They also found a correlation between the Na and K content of the red blood cells and neural activity from the taste receptors (Table 3).

Table 3. Relationship between Na and K content of red blood cells and taste response (Beidler, Fishman and Hardiman, 1955).

	Na/K	
	Red cell content	Relative taste response
<u>Carnivores</u>		
Raccoon	19.3	0.74
Cat	17.0	0.67
Dog	12.0	0.44
Average:	16.1	0.62
<u>Rodents</u>		
Rat	0.12	2.0
Hamster	0.11	2.8
Guinea pig	0.14	2.6
Average:	0.12	2.5

The Na/K ratio in the red blood cells was  $>1$  whereas the Na/K ratio of the relative neural taste response was  $<1$  in the carnivores. In rodents the red blood cell Na/K was  $<1$  and the neural taste response Na/K ratio was  $>1$ . Although one could correlate the Na and K content of the red cell with the neural activity of the taste receptors to the same salts,



it was not implied that there was necessarily a causal relationship between the events in the red cell and those in taste. Differences in taste responses were probably a result of slight difference in the physicochemical structure of the receptor surface. The correlation between taste response to NaCl and KCl and the red cell content which existed for many species broke down when individual sheep were considered. Some sheep have red cells of high sodium and low potassium content, whereas others have the opposite (Evans, 1957). The response to sodium and potassium was recorded electrophysiologically on nine sheep and the red cell content of the same individuals determined for sodium and potassium. The response of all sheep tested to 0.5 M. sodium chloride was twice the magnitude of that of 0.5 M. potassium chloride even though seven sheep had red cells of high sodium content (90 M. equiv.) and two of low sodium (37.6 M. equiv.) and high potassium (72.2 M. equiv.) content (Beidler, 1962).

Tateda and Beidler (1961) observed the electrical responses of the taste cell of the rat to chemical stimuli by means of microelectrode techniques. The steady voltage of the cells of the taste buds, as recorded with microelectrodes, could be deflected positively by application of a chemical stimulus to the tongue, the magnitude of the voltage change increasing with the strength of the stimulus. If however, this steady voltage was raised to much more positive level with previous application of 1 percent cocaine or 0.06 M. ferric chloride to the tongue, then the stimulus produced a negative change in voltage, as if it always approached to some equilibrium level. The study suggested that application of a chemical stimulus caused an increase in cell membrane perme-

ability at some point distant from the microvilli. This change in permeability or cell voltage was associated with the excitation of the innervating nerve fibers.

Beidler and Smallman (1965) investigated the functional anatomy of taste buds using the techniques of colchicine inhibition of mitosis, electrophysiology and radioautography. Colchicine blocked mitotic division of the epithelial cells surrounding the taste bud of the rat tongue. Response to chemical stimulation decreased 50 percent three hours after colchicine injection as measured by the electrical activity from the taste nerve bundle and no response after 8-10 hours. Radioautography using tritiated thymidine showed that those epithelial cells surrounding the taste bud divided and some of the daughter cells entered the taste bud and slowly moved toward the center. The life span of the average cell was about  $250 \pm 50$  hours. The tagged cells disappeared from the taste bud logarithmically, which suggested that the cells may die as a result of indiscriminate injury and not as a result of age. Mature taste cells do not undergo mitotic division.

Beidler (1966) described the physiological basis of taste sensation. The individual taste cell within the bud was like a living battery with the source of electrical energy being an uneven distribution of potassium and sodium ions across the cell surface. Each cell had finger-like projections, taste microvilli, that extended into the saliva coating the tongue where contact was made with various food chemicals. These chemicals were adsorbed at specific sites on the microvilli, slightly changing the spatial arrangement of the molecules making up the surface of the taste receptor. In some unknown manner, the influence of

the filled sites was projected further down the surface of the receptor cell where minute holes were created in the  $75 \text{ \AA}^0$  lipid-protein cell membrane. The  $\text{Na}^+$  and  $\text{K}^+$  ions leaked through these holes and exchanged with similar ions in the thin layer of solution encircling the body of the receptor cell. This increase in ion permeability of the receptor cell membrane resulted in a decrease in the electrical voltage normally present across the receptor cell membrane. The magnitude of the voltage change was proportional to the number of microvilli receptor sites filled by the taste molecules, the maximum change being about 30 to 40 mv as measured by microelectrodes inserted into the cells. This voltage change was associated with a change in current which crossed and thereby stimulated the nerve endings that were closely encircled by the receptor cell membrane. The nerve then developed a series of brief (1-3 m/sec) electrical impulses that were carried to the brain, which in turn acted as a computer to determine whether the food was salty, sour, bitter or sweet.

Pfaffmann (1941) studied the distribution of taste sensitivity on the cat's tongue by recording nerve impulses in the chorda tympani and glossopharyngeal nerves. The apex and anterior lateral margins of the tongue were found to be most sensitive to salt, the base and posterior lateral margins to quinine and all regions except the mid-dorsum were sensitive to acid. Response to sugar rarely occurred. He showed that there were three types of gustatory fibers in the cat: one type that responded to acid, one that responded to both acid and sodium chloride and one to both acid and quinine.

Zotterman (1935) studied the cat using a brush border applicator.

He found responses mainly from chemical stimulation when recording from the chorda tympani. Sodium chloride, quinine sulphate and acetic acid were effective stimuli, whereas sucrose was generally ineffective. Quantitative measures of whole nerve activity were not made but response from single fibers were analyzed.

Pfaffmann (1955) observed that the taste solutions applied to the anterior tongue surface of the rat, the cat and the rabbit elicited a maintained asynchronous discharge of impulses in the chorda tympani nerve. For all species the basic taste stimuli could be ranked from low to high in order of thresholds as follows: quinine, HCl, NaCl and sucrose. The magnitude of the total nerve response was typically an increasing sigmoid function of the logarithm of the stimulus concentration. The responses to NaCl and HCl were typically of larger magnitude than those to quinine and sucrose. Recording from single nerve fibers of the chorda tympani nerve in the three species showed that thresholds for any one substance were different from one fiber to another. Frequency of discharge during the first second approximated a sigmoid function of the logarithm of the stimulus concentration. Each single fiber preparation was characterized by a different pattern of sensitivity to the four basic taste stimuli. Every element isolated responded to more than one of the four basic taste stimuli but to varying degrees. No simple classification of receptors by types was obvious; chemical specificity appeared to be relative so that any one element might be characterized by the stimulus or stimuli for which it had the lowest threshold.

Anderson, et al. (1950) attempted the localization of the different tastes on the dog's tongue. Acid elicited impulses from the whole

of the tongue supplied by fibers running in the peripheral part of the lingual nerve. In the cat there were no fibers responding to sweet solutions but in the dog an area beginning just behind the tip of the tongue was sensitive to sweet. This area differed from that in man in that the very tip of the tongue was fairly insensitive to sweet. The sensitivity for sweet increased posteriorly and it was found to be maximal at the most posterior part of the tongue supplied from the chorda tympani. The tip was most sensitive to salt while the response to acid seemed to be evenly distributed over the whole tongue.

Cohen, Hagiwara and Zotterman (1955) analyzed the taste receptors in the cat's tongue by recording from single afferent chorda tympani nerve fibers while applying various sapid solutions to the tongue. Single fibers were observed which responded to the flow of distilled water over the tongue. The "water" fibers were also stimulated by quinine hydrochloride and mineral acids below pH 2.5. Single "salt" fibers were observed which responded to various salts above 0.1 M. concentration. These fibers were also stimulated by acid. Another fiber type responded primarily to quinine chloride and little if at all to strong acid. The fourth fiber type responded only to acids below pH 2.5.

Zotterman (1956) studied the species differences in the water taste including in his study the rabbit and the white rat. He observed that the white rat lacked taste fibers discharging on the application of water to the tongue but that it had "salt" fibers which gave a prompt and lasting response to the application of NaCl solutions as weak as 0.02 M. The rabbit displayed a delayed response to water while the cat

and the pig responded promptly to the application of water upon the tongue. In the cat, pig or rabbit the "salt" receptors were almost completely adapted to the saliva and adapted quickly to the application of Ringer's solution. After a water rinse they all produced a phasic response to even as diluted NaCl solutions as 0.002 M. The high discrimination of salty solutions in the rat must rest upon its highly sensitive "salt" fibers. There was no information about salt discrimination in the cat, dog and pig. These species possess water fibers that may play a part in discrimination between water and weak salt solutions.

Zotterman (1959) observed that the chorda tympani of the Rhesus monkey contained a large number of fibers that respond specifically to sweet-tasting substances such as sucrose, glycerol, ethylene glycol and even to saccharin. The monkey has been the first animal possessing specific fibers responding to sucrose in which the same fibers respond positively to saccharin. The monkey had specific fibers for salt, sweet, bitter and acid solutions as well as water. In the dog, pig and rat, all of which possess specific sweet fibers, saccharin did not produce any positive effect on the sweet receptors. In the dog strong concentrations of saccharin seemed to stimulate the bitter-taste fibers (Anderson, et al., 1950).

Zotterman (1961) reported that the calf and the lamb either lacked or had very few fibers in their chorda tympani that responded to sweet testing solutions. Sweet-tasting solutions elicited positive responses in the rat, dog and pig but very sweet-tasting saccharin solution 0.02 M. failed to elicit any response in those animals. In the rat and man, the

application of water to the tongue was followed by a reduction in the spontaneous activity in the nerve. He found the human and the rat did not have fibers that responded specifically to the application of water in contrast to the cat, the dog and the monkey.

Beidler, Fishman and Hardiman (1955) observed that the rabbit, dog and cat were not as responsive to inorganic chloride salts as were the rat, hamster and guinea pig. NaCl was effective in stimulating the chemoreceptors of the rat, hamster and guinea pig but was the least effective salt when applied to the cat, dog and rabbit.  $\text{NH}_4\text{Cl}$ , on the other hand, was a very effective stimulus for all preparations. The rat responded better than the cat to sugar and the hamster and guinea pig better yet. Quinine was more effective in the cat than in the rat or rabbit (Pfaffmann, 1955).

Pfaffmann (1961) observed, as a result of species comparisons and extensive single fiber analyses, basic receptor types analogous to "four" taste qualities could be modified into a concept of differential chemical sensitivity.

Baldwin, et al. (1959) studied the electrical response of the chorda tympani and glossopharyngeal nerve of sheep, goat and calf on stimulation of the tongue with sapid solutions. All three species reacted strongly to acid and salt solutions and less strongly to quinine solutions. Reaction to sugar solutions was difficult to elicit by simple irrigation of the tongue but definite responses could be evoked from the glossopharyngeal nerve, especially when the circumvallate papillae were moved at the same time as the solution was applied. Saccharin produced no response although ethylene glycol and glycerine did.

The goat, sheep and calf did not appear to have "water" fibers, for the spontaneous activity seen in the chorda tympani was never increased but decreased when the tongue was irrigated with water.

Kitchell (1961) elaborating on the previous data reported that in the goat the application of water simultaneously with stroking of the tongue did not produce a response other than due to stroking the tongue. Responses appeared following the application of 0.7 M. NaCl, 0.02 M. quinine dihydrochloride, 0.3 M. sucrose, 0.2 M. acetic acid and 0.75 M. Na bicarbonate while stroking the tongue. Difficulty was experienced in observing any response at all in the glossopharyngeal nerve if the vallate papillae were not stroked.

Zotterman (1935) found responses to both chemical and mechanical stimulation of the cat tongue when recording from the glossopharyngeal nerve. Pfaffmann (1941) reported responses to mechanical stimulation in the cat and found the posterior area of the tongue responsive to 0.1 M. HCl and 0.02 M. quinine hydrochloride.

Appelberg (1958) studied the rat, cat, dog, rabbit and monkey. Responses to NaCl, quinine and acetic acid were found in all the species studied. All except the cat responded to sucrose and glycerin. Only the monkey responded to saccharin and to water. A tactile response was found in all species. The tactile receptive field of the glossopharyngeal nerve extended from the epiglottis to a border line a few millimeters anterior to the circumvallate papillae and was strictly ipsilateral. The row of large fungiform papillae at the border of the caudal part of the tongue were very sensitive to touch. There was a certain overlap between the tactile receptive fields of the glosso-



pharyngeal and the lingual nerve, which was restricted to a few millimeters.

Kare and Ficken (1963) gave the mean number of taste buds in a variety of species as follows: chicken, 24; pigeon, 37; bullfinch, 46; starling, 200; duck, 200; parrot, 350; snake, 0; kitten, 473; bat, 800; human, 9,000; pig and goat, 15,000; rabbit, 17,000; calf, 25,000; and catfish, 100,000. The cow with 25,000 taste buds did not respond behaviorally to chemicals that evoked strong rejection in the fowl having only a few dozen buds. The correlation between the gross anatomy of the receptors and taste behavior was difficult to explain. The numerical enigma paralleled many others in taste which so far affected no pattern of function.

Bell (1963) observed the variation in taste thresholds of ruminants. Herbivorous animals in general, ruminants in particular, often existed on a quite precarious mineral balance because the vegetative diets contained little sodium and relatively more potassium. He described the variation in preference thresholds for sodium in monovular twin calves. He adopted Denton's (1957) method of inducing sodium deficiency in sheep by exteriorization of the parotid duct so that continuous secretion of saliva caused a loss of sodium from the body. When the parotid duct of a calf was exteriorized the salivary Na:K ratio was reversed from 145 M. equiv: 4.5 M. equiv to 5.6 M. equiv: 132 M. equiv but that it could be readily restored by feeding a supplement of NaCl or NaHCO<sub>3</sub>. When an experimental calf was allowed continuous access to either 1 percent NaCl or 2 percent NaHCO<sub>3</sub> it would take a drink sufficient to maintain its salivary Na/K ratio at normal levels, whereas its

control twin took only minimal quantities.

The behavior provided support for Richter's view and indicated that the sense of taste played an important part in the "environmental homeostasis" of the ruminant herbivores. Although the adrenal gland was the main source of the controlling mechanism of electrolyte control in sodium depleted sheep, it was possible that other areas, perhaps some part of the central nervous system might also be concerned. In ruminants there was a close interrelationship between taste, metabolism and nutrition.

#### Taste pathways

The afferent fibers of the VIIth, IXth and Xth cranial nerves after entering the medulla formed a well-defined common descending tract, the tractus solitarius. In this respect taste fibers behaved like the pain and temperature fibers of the trigeminal nerve, which descended in the neighboring spinal trigeminal tract. Anatomical studies have pointed to the cephalic portion of the nucleus of the tractus solitarius as the medullary terminus for the primary gustatory neurons (Terrik, 1956).

Pfaffmann, et al. (1961) recorded electrical activity in the rostral portion of the nucleus of the solitary tract in the medulla of the rat and activity was recorded in the thalamus from the medial tip of the ventro-basal complex. The discharges in the medulla and thalamus were similar to the discharges recorded peripherally in the chorda tympani nerve.

The fibers of the three nerves terminated at different levels in the nucleus. Fibers from the facial and glossopharyngeal nerves termi-

nated in the rostral part of the nucleus only, and vagal fibers entered the caudal portion (Ranson and Clark, 1955). Consequently, secondary taste neurons were concentrated in the rostral part of the nucleus.

The axons of the secondary neurons pursued a course up the brain stem in close relation to the medial lemniscus and the ventral secondary quinto-thalamic tract. The gustatory fibers relayed in the most medial part of the nucleus ventralis postero-medialis (arcuate nucleus) (Rush and Patton, 1965).

Halpern (1959) plotted bulbar responses for various chemical substances and observed that they were almost identical to those obtained from peripheral nerves.

Frommer (1961) recorded multi-unit responses from the thalamus, which added to the data furnished by Halpern (1959), provided a comparison of responses in the central nervous system (Table 4).

Table 4. Response magnitude to various chemical stimuli (Frommer, 1961).

Chemical stimuli	Relative response magnitude		
	Thalamus	Medulla	Chorda tympani
0.1 M. NaCl	100	100	100
0.1 M. KCl	39.8	27.0	36.5
0.005 M. HCl	52.0	35.5	53.5
0.01 M. Quinine HCl	21.8	22.8	15.5
1.0 M. Sucrose	33.8	30.0	44.0

Makous, et al. (1963) observed the gustatory relay in the medulla of rats and cats. Multi-unit recording in the medulla in the anterior

solitary tract and its nucleus yielded responses to the application of taste stimuli to the anterior tongue. Histological analysis verified the location of the taste reactive areas in the anterior solitary tract or associated nucleus. The taste areas of the medulla were contiguous to and often overlapped areas sensitive to tactile and temperature sensitivity of the tongue and general mouth areas.

Benjamin and co-workers (1963) mapped the thalamic projections of the various taste nerves in the rat and the squirrel monkey. In both animals taste had an exclusive territory independent of other tongue modalities in the most medial extension of the ventral nuclear mass. Non-gustatory neurons responding to either or both thermal or mechanical stimulation of the tongue were situated more laterally. Taste was ipsilateral. In the rat and the squirrel monkey thalamus the taste system was spatially separated from other tongue modalities. In the cat, taste, touch and temperature units were all intermingled.

Anderson and Jewell (1957) studied the thalamic relay for taste in the goat and observed stimulation of the posteromedial ventral thalamic nucleus in the unanaesthetised goat evoked licking, chewing and "rejection" movements which suggested that the animal was experiencing an unpleasant taste. After bilateral ablation of this nucleus, thirsty goats, which normally refuse to drink distasteful solutions, readily drank such solutions and appeared to have lost all sense of taste.

In the cortex the taste receiving area lay in close association with the representation of other modalities from the tongue. Benjamin (1963) observed two small areas on the surface of the rat brain, one for each chorda tympani nerve, that evoked response with a gross microelec-

trode. The gustatory function of this taste nerve area was established by ablation experiments. Bilateral removal of the composite area produced impairment of taste discrimination. Conversely, removal of the rest of the neo-cortex, sparing the composite area, left discrimination normal. In the squirrel monkey electrical stimulation of the chorda tympani stimulated two areas. Removal of one area in each of five monkeys produced only a small deficit in one and removal of both the areas as well as the fronto-parietal operculus and part of the insula produced only a slight deficiency in one monkey. The studies established that in the squirrel monkey the claustrum may be involved in taste perception.

Elliot (1937) studied taste bud distribution on the tongues from four neonatal kittens and observed that all taste buds were restricted to the dorsal surface and in association with the fungiform and circumvallate papillae. An average of 473 taste buds exclusive of those found in the circumvallate papillae were counted. A small percentage of those were found in the anterior third and about 50 percent of the total number in the middle third and almost as many in the posterior third. The number of taste buds in the circumvallate papillae were not determined. The taste buds in the fungiform papillae were in both the tops and sides of such papillae. The taste pore was absent in the taste bud in the top and those buds were probably the first to show retrogression. The number of taste buds did not exceed four in the fungiform papillae. The taste buds were found in the tops and sides of the circumvallate papillae, as in the fungiform papillae. Around each circumvallate papillae was a shallow trench with an outer trench wall. In the adult,

the taste buds were described as being present in the outer trench wall; but none were in the kitten at birth.

Hayes and Elliot (1942) observed the distribution of the taste buds on the tongue of the kitten, with particular reference to those innervated by the chorda tympani nerve and noted the average of 575 taste buds, innervated by the lingual nerve of one side and approximately 1150 for the entire tongue. This indicated an early post-natal rise in the number of buds on the tongue of the kitten as there were 473 buds on the tongue of the kitten at birth (Elliot, 1937).

In the kitten at two months, taste buds associated with the fungiform papillae were found on the tops of the papillae and some papillae were found to contain no buds. The average number in a single papilla was four. Taste buds were found on the tops and sides of the circumvallate papillae and began to appear in the outer wall by the time the kitten was two months old. They were not present in the outer trench wall at birth (Elliot, 1937).

Holliday (1940) studied the total distribution of the taste buds on the tongue of the pup and noted all the taste buds were limited to the dorsal surface associated only with the fungiform and the circumvallate papillae. An average of 1706 buds per tongue was observed, composed of 262 buds in the circumvallate papillae and 1444 buds in the fungiform papillae. The taste buds were found only on the tops of the fungiform papillae and the greatest number to occur in any fungiform papilla was six. There was a gradual increase in the number of taste buds from the tip of the tongue toward the circumvallate papillae, but none were found caudal to the circumvallate papillae. The circumvallate

papillae varied from four to six and contained the greatest number of taste buds. Taste buds were on the top and sides of the papillae and in the floor of the trench which surrounded the circumvallate papillae. Buds were most numerous on the sides of such papilla. No taste buds were present on the outer trench wall.

Busono (1966) studied the distribution of the taste buds in sheep (3 foeti and 2 adults) and observed that the circumvallate papillae always bore taste buds on the side walls in an adult sheep and also on the oral surface in a fetus or newborn lamb. Taste buds were found in fungiform papillae but not in all of them. The apex linguae fungiform papillae had more taste buds than those papillae on the dorsum linguae. The pharynx, the epiglottis, the anterior region of the esophagus and the aditus laryngis had scattered taste buds during fetal life, but a few months after birth those buds atrophied and vanished. Taste buds were absent on the soft palate, the hard palate, papilla incisiva, lips, cheeks and conical papillae.

Bell and Kitchell (1966) observed that the goat, sheep and calf had gustatory chemoreceptors which responded to salt, sweet, sour and bitter solutions, from which afferent fibers passed centrally in the chorda tympani and glossopharyngeal nerves. Action potentials in the afferent gustatory nerves could be detected when the tongue was irrigated with sodium bicarbonate, ethylene glycol, glycerine and saccharin. There were no fibers which responded to distilled water. The responses from the circumvallate papillae were enhanced if the papillae were gently moved at the time of irrigation with sapid solutions. The chorda tympani was most responsive to salt and acid solutions while the glosso-

pharyngeal was more responsive to sugars, quinine and acid.

Moore and Elliott (1946) studied the distribution of the taste buds on the tongue of the pigeon. Papillae resembling mammalian filiform papillae were present on the tongue folds and on the cranial portion of the tongue wings. Taste buds were not found in connection with these, but were scattered through the epithelium, making their identification rather difficult. Taste buds were found on the dorsal and lateral surfaces of the soft portion of the tongue and on the lateral, medial and ventral surfaces of the tongue wings. The greatest concentration of taste buds, 70.7 percent of the total, was found on the dorsal portion of the tongue caudad of the tongue fold. The maximum number of taste buds present on any one tongue was 59, and the minimum was 27, with an average of slightly less than 37 for the six tongues which were plotted.

Kare, et al. (1957) by the methodical testing of hundreds of potential stimuli showed that the chicken had a sense of taste. The response to dilute solutions of flavor based on the limits of man's perception or to small alterations in concentration supported the contention that this sense in the fowl is more than rudimentary. The response to a variety of sweet and bitter flavors suggested that the broad classifications of taste recognized by man were not applicable to the fowl. The chicken's chemical response spectrum was different from that of the human.

Lindenmaier and Kare (1959) showed the specialized receptors on the tongue of the chicken were in reduced numbers and restricted in distribution. The taste buds were found only on the base of the tongue and floor of the pharynx, caudal to the row of large horny papillae. In



day-old chicks the number of buds averaged eight; in the three-month-old cockrel there were 24. Morphologically, the chicken taste buds resembled, but were not identical to, those of mammals. The fasciculus solitarius in the medulla transmitted taste impulses in mammals. This structure was highly developed in birds and it is possible that it greatly reinforced the impulses which it relayed from the small number of taste buds to the cortex.

Kare and Medway (1959) found that the chicken could discriminate between carbohydrates. It was indifferent to dextrose and sucrose but rejected xylose. The pattern of discrimination suggested that sweetness, as recognized by the human, was of no consequence to the fowl. Viscosity, refractive index, concentration, osmotic pressure and density were not factors determining the reaction of the fowl to various sugars. There was no clear-cut evidence for explaining the rejection or indifference to sugars, instead there was a suggestion that the discrimination was based on absolute specificity for the sugar involved.

Kitchell, et al. (1959) conducted electrophysiological studies of thermal and taste reception in chickens and pigeons by applying adequate stimuli to the tongue. Receptors were found in the tongue and pharynx which responded, in general, like those found in mammals. The thermal and taste receptors discharged through two peripheral branches of the ninth nerve only. In the chicken positive responses from the nerves followed the application of distilled water, salt, glycerine, ethylene glycol, quinine and acetic acid to the tongue but not of sucrose and saccharin. In general, a good correlation was seen between the presence or absence of a taste response and the behavior of the bird in relation

to a particular substance. As a result of the studies, a taste mechanism in domestic fowl was convincingly demonstrated.

#### SUMMARY

In the dog five types of lingual papillae were described and named--filiform, fungiform, conical, circumvallate and foliate papillae. Taste buds were found in the fungiform, circumvallate and foliate papillae. The number of circumvallate papillae ranged from four to six.

The mammalian taste bud degenerated and completely disappeared after transection of the innervating taste nerve and reappeared upon reinnervation of the papilla by the sensory nerves.

The chorda tympani nerve contained not only taste nerves but also other sensory and motor nerves.

The chorda tympani nerve of the dog and cat was not fasciculated and a single nerve fiber was in functional relation with more than one taste bud and many more receptor cells. Gustatory responses were recorded from the calf chorda tympani upon chemical stimulation of either the anterior or posterior portion of the tongue.

The division of the taste bud into sustentacular cells and neuro-epithelial or receptor cells was questioned for some years. The absence of sustentacular cells was confirmed by recent electron microscopic studies. No taste hairs were seen in the bud by use of the electron microscope but microvilli were consistently found.

The synaptic vesicles found at the termination of the larger of the unmyelinated nerve fibers indicate an efferent pathway.

Beidler's analytical theory of the taste mechanism indicated that

the initial step in gustatory stimulation was an adsorptive one. Enzymatic processes in taste were probably located further downstream at secondary stages. The life span of the average taste cell was  $250 \pm 50$  hours.

Species differences in responses to four types of chemical stimuli namely sweet, sour, bitter and salty was discussed as was the absence of water receptors in ruminants, rats and rabbits.

The gustatory fibers of the seventh, ninth and tenth cranial nerves ran into the tractus solitarius, with its nucleus in the medulla. The second link in the pathway was formed by axons of the cells of the solitary tract which ascend in the brain stem. The taste pathway in animals was ipsilateral and the taste receiving area in the cortex lay in close association with representations of other modalities of tongue.

In the kitten at birth an average of 473 taste buds, exclusive of those found in circumvallate papilla, were found. There was an early post-natal rise in the number of taste buds to 1150 for the entire tongue. No taste buds were found in the outer trench wall at birth but they began to appear at the age of two months. In each fungiform papilla the number of taste buds was four.

In the pup an average of 1706 buds per tongue was observed--262 buds in the circumvallate and 1444 buds in the fungiform papillae. The greatest number of fungiform papillae was six. Circumvallate papillae were four to six in number. Taste buds were most numerous on the sides of each papilla but were absent on outer trench wall.

In the sheep, not all fungiform papillae showed taste buds. Circumvallate papillae had buds on the side walls in the adult and also

on the oral surface in a fetus or new-born lamb. The pharynx, epiglottis, the anterior region of the esophagus and aditus laryngis had scattered taste buds during fetal life and up to a few months after birth; but in the adult those buds atrophy and vanish.

The pigeon has 27 to 59 with an average of 37 taste buds, with the greatest concentration on the dorsal surface caudad to the tongue fold.

Day old chicks had eight taste buds and three-month-old cockrel, twenty-four. The fasciculus solitarius was highly developed in birds. Electrophysiological study to the neural responses in the afferent nerves confirmed the presence of taste buds in the domestic fowl. There was no chorda tympani branch of the facial nerve so all afferent impulses discharged through ninth nerve only.

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A COMPARATIVE STUDY OF THE DISTRIBUTION OF GEMMA  
GUSTATORIA IN THE DOG, CAT, SHEEP AND CHICKEN

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AN ABSTRACT OF A MASTER'S REPORT

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This report deals with the distribution of taste buds in the dog, cat, sheep and chicken. The mechanism of taste receptor stimulation, species' differences to taste, electrophysiological studies with a special emphasis on anatomical study and ultrastructure of the taste buds and nerve fibers and their central pathways are discussed.

In the dog, five types of lingual papillae were described, namely: filiform, fungiform, conical, circumvallate and foliate. Taste buds were found in the fungiform, circumvallate and foliate papillae. In the cat foliate papillae were rudimentary and without taste buds.

The mammalian taste buds degenerated after transection of the innervating taste nerve and reappeared upon re-innervation of the papilla by the sensory nerve. The chorda tympani nerve contained besides taste nerves other sensory and motor nerves. In dog and cat this nerve was not fasciculated. A single nerve fiber was in functional relation with more than one taste bud and many more receptor cells. The division of taste bud cells into sustentacular and neuroepithelial cells was modified. Electron microscopic studies confirmed the absence of sustentacular cells and taste hairs at the end of receptor cells and the consistent presence of microvilli. Synaptic vesicles at the termination of the larger unmyelinated fibers could indicate efferent pathway. The initial step in gustatory stimulation was adsorptive and secondary steps were probably enzymatic. The life of an average chemical taste cell was  $250 \pm 50$  hours. Species' differences to several chemical stimuli were discussed. The gustatory fibers of the seventh, ninth and tenth cranial nerves ran into the tractus solitarius in medulla; the second link was in the brain stem ending in the thalamus. The taste pathway was ipsi-

lateral in animals and in the cortex the taste receiving area was in close association with terminations of other tongue modalities. Gustatory responses were recorded from the calf chorda tympani upon chemical stimulation of either the anterior or posterior portion of the tongue.

The kitten at birth averaged 473 taste buds exclusive of those in circumvallate papillae. There was an early post-natal rise in the number of taste buds to 1150. In the pup an average of 1706 buds per tongue--262 buds in circumvallate and 1444 buds in the fungiform papillae were counted. In the sheep not all fungiform papillae showed taste buds. The circumvallate papillae had buds on the side walls in the adult and also on the oral surface in a fetus and a new-born lamb. Taste buds were scattered in the pharynx, epiglottis and the anterior region of the esophagus and aditus laryngis during fetal life but after birth atrophied until they vanished in the adult.

In the pigeon an average of 37 taste buds were observed. The chicken's taste ability was well developed. The day old chick had eight taste buds and a three-month-old cockrel, twenty-four. The fasciculus solitarius in the medulla was highly developed. There was no chorda tympani branch of the facial nerve so all afferent impulses discharged through the ninth nerve only.