

GUSTATION IN FISH

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Abstract

This report reviews the taste systems of teleosts as ideal animal models to investigate the neural processing of gustatory and tactile information from peripheral through to higher gustatory centers. An overview of recent physiological studies is mainly focused on to allow a better understanding of the sense of taste in fishes. Then, what should be done in this science field is discussed.

Key words

fish, taste, tactile, amino acids, facial taste system, vagal taste system, somatotopic arrangement, neuroanatomy, electrophysiology

I. INTRODUCTION

Living action of all animals including man requires a supply of energy, for which they have to obtain foods from various sources of surroundings by using chemical perceptivity. Vertebrates possess two principle systems, olfaction and taste, adapted to respond to chemical substances in environments. Although olfaction is generally termed the "distant" sense and gustation the "contact" sense, with fishes chemical stimuli are present in the aquaous medium and it is difficult to distinguish between olfactory and gustatory stimuli or to explain the development of two chemical senses in aqueous vertebrates¹⁾⁻³⁾, because taste in fish would appear to be an important distant sense having behavioral implications⁴⁾⁻⁶⁾. Fish, olfaction aside, evolve elaborately their gustatory systems of all vertebrates and are the best endowed for the detection of foods. Generally the taste sense of animals plays an important role in selection of foods and protection against noxious substances. As well-known, the mode of fish life diversifies in the aquatic environment, which affects on the structure of the hind brain in relation to fish habits of feeding⁷⁾⁻¹¹⁾. In order to adapt to a specific way of life, some teleosts, especially Cyprinidae and Siluridae (and Ictaluridae), have an enormous number of taste buds in the mouth, the palatal organ (only in Cyprinidae), the gill region, and over the whole body surface¹²⁾⁻¹⁸⁾. Especially their barbels and lips are densely studded with taste buds^{12), 16), 18)} (Fig. 1). The external cutaneous chemoreception on fish has close connection with searching, hitting and picking up behavior

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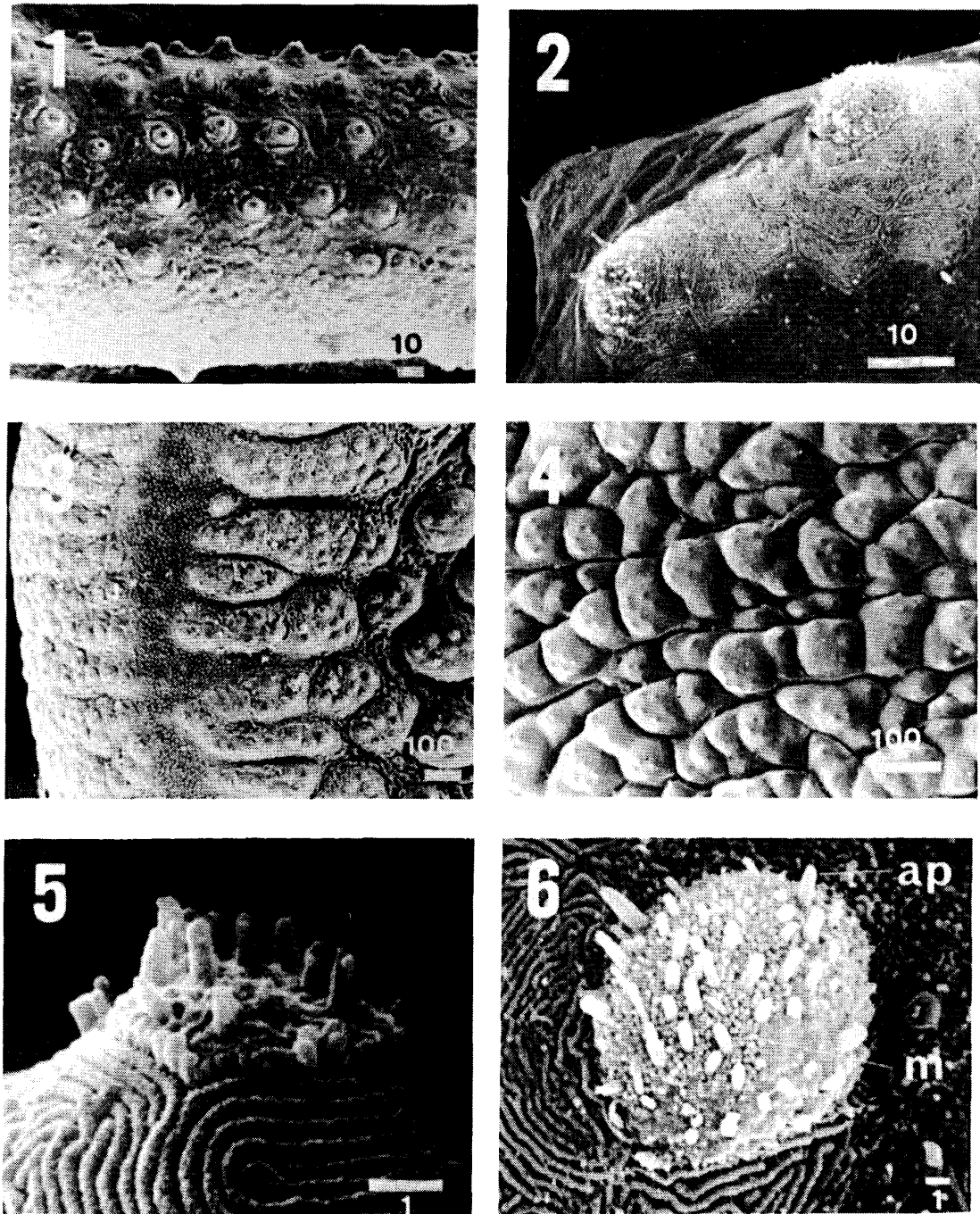


Fig. 1 Scanning electron microscopic micrographs of taste buds on carp.

1. Taste bud distribution on the maxillary barbel.
 2. Taste buds on the outer skin surface.
 3. Anterior part of the oral roof.
 4. Palatal organ.
 5. Lateral view of a taste bud swollen.
 6. Detail of the surface of the bud. The surrounding epithelium exhibits microridges, as well as in 5.
- The numbers attached the bars in each micrograph show the scale in μm . Abbreviations : ap, apical process on the surface of the bud ; m, microvilli.

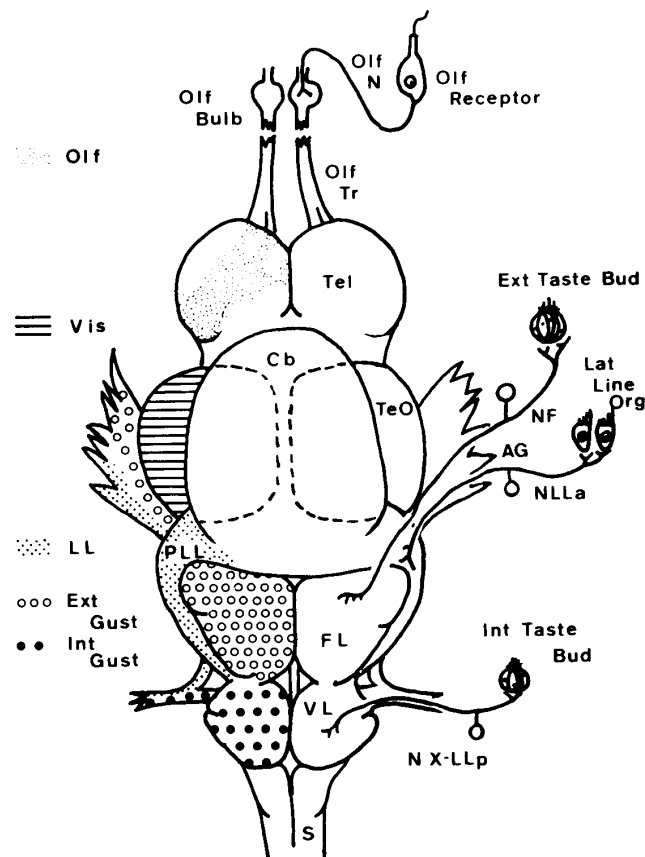


Fig. 2 Schematic dorsal view of the Japanese sea catfish ("Gonzui") brain is shown, where primary central terminal fields for various cranial nerves are represented. (Reformed from T.E. Finger, 1982, reference no. 92.)

in its feeding¹²⁾. Cyprinidae and Siluridae inhabit poorly illuminated ponds and lake bottoms, and rely less on sight than do other fishes¹⁹⁾, indicating that the families are prime exponents of such gustatory systems. Therefore, most gustatory experiments in fish have been performed using these families. Taste buds in fish are innervated by fibers from the cranial VIIth, IXth, and Xth nerves^{20)–23)}. According to those peripheral developments, the primary gustatory centers, the facial, glossopharyngeal and vagal lobes, are enlarged into prominent features, and better developed than in other fishes. Each lobe serves as the region of primary termination for one of the three gustatory fibers; facial, glossopharyngeal and vagal nerves. Their central gustatory projections show the cerebral enlargements which have a series of a few dorsal conformations^{8), 24)–30)}, the vagal and facial lobes arising from the visceral sensory column^{20), 23)}. These lobes are called "the primary gustatory centers", the nuclei gustus primus, by C. J. Herrick²³⁾. In Fig. 2, schematic dorsal view of the Japanese sea catfish brain is shown, in which only primary central terminal fields for various cranial nerves, olfactory, visual, lateral line and taste centers, are represented. Further, there are two major secondary gustatory nuclei in these species. One, the superior secondary gustatory nucleus (nGS), is situated at the isthmus region between the optic tectum and cerebellum²³⁾. This secondary nucleus should correspond to the pontine taste area in mammals^{31)–33)}. The other secondary gustatory nucleus, the inferior secondary gustatory nucleus, is situated at the medial funicular region of the caudal medulla (the nucleus funicularis medialis, nFM). The former nucleus receives input from the facial and vagal lobes. The latter does only from the facial lobe^{27), 28), 30)}. Thus, these species serve as ideal animals to study the central relation and connections

of the gustatory systems^{34)–36)}. However, there were a few report of electrophysiological experiments on their central mechanisms^{34)–39)}.

On the other hand, over the past decade, electrophysiological^{2), 3), 6), 40)–44)}, biochemical^{45), 46)} and behavioral^{41), 47)–50)} experiments clearly demonstrate that L- α -amino acids are an important class of chemical stimuli to the chemosensory systems of fishes, as well as those of numerous aquatic invertebrates^{51)–55)}. Specific amino acids are suitable, high-affinity ligands for the fish taste receptor, because electrophysiological^{3), 40), 42)–44)} and behavioral^{47), 49)} stimulus thresholds for them in the facial taste systems of a number of different teleosts fortunately approach and occasionally extend below nanomolar stimulus concentrations. Thus, using such kinds of fishes as experimental models allows us to best use our knowledge in taste physiology.

This report provides an overview of recent physiological and anatomical studies to allow a better understanding of the sense of taste in fishes closely associated with that in mammals.

II. TASTE STIMULI AND CHEMICAL ACUITY

In the 1930's, Hoagland⁵⁶⁾ recorded mechanoreceptive activity along with the gustatory activity from the barbel nerve of North American catfish, in which acetic acid, sodium chloride and meat juice evoked barely appreciable potentials in the facial nerve. The relative response magnitudes might be associated with the di-

TABLE I. GUSTATORY EFFECTIVENESS AND CONCENTRATION SERIES.

Species	Stimulus	Threshold ($10 \times M$)	References
1. Catfish <i>Ictalurus</i> <i>Punctatus</i>	L-Ala>L-Arg> L-Ser>L--Abu> L-Gln>Gly	– 8 to – 11	Caprio 1975, 1978, (40, 6)
2. Puffer <i>Fugu</i> <i>pardalis</i>	L-Pro>L-Ala> Gly	– 5 to – 6	Hidaka et al., 1976. (61)
3. Eel <i>Anguilla</i> <i>japonica</i>	L-Arg>Gly> L-Ala>L-Pro> L-Lys	– 7 to – 9	Yoshii et al., 1979 (44)
4. Minnow <i>Pseudorasbora</i> <i>parva</i>	L-Pro>L-Lys> L-Ala>L-Arg> L-CysH	– 9 to – 11	Kiyohara et al., 1980 (42)
5. Red sea bream <i>Chrysophrys</i> <i>major</i>	L-Ala>Gly> L-Arg>L-Ser> L-Lys	– 6 to – 7	Goh and Tamura, 1980.(2)
6. Mullt <i>Mugil</i> <i>cephalus</i>	L-Arg>L-Lys> L-Ala>L-Ser> L-Leu	– 6 to – 7	Goh and Tamura, 1980.(2)
7. Rainbow trout <i>Salmo</i> <i>gairdneri</i>	L-Pro>Bet> L-Hyp>L-Leu> L-Ala	– 5 to – 8	Marui et al., 1983.(43)
8. Carp <i>Cyprinus</i> <i>carpio</i>	L-Pro>L-Ala> L-CysH>L-Glu-Na Bet	– 8 to – 10	Marui et al., 1983.(3)
9. Tilapia <i>Tilapia</i> <i>nilotica</i>	L-His>L-Arg> L-Try>L-Ser> L-Gln	– 8 to – 9	Marui et al., unpublished

ameter of nerve fibers : the large Gasserian ganglion cells supply the tactile receptors and small size of the geniculate ganglion cells send axons to taste buds. Since then, there are a number of reports concerning fish taste physiology. However, most investigations had been performed by using the classical taste stimuli until Caprio⁴⁰⁾ found with electrophysiological techniques that the taste system of the channel catfish is sensitive to extremely dilute concentrations of amino acids ($< 10^{-9}\text{M}$). After the report on the catfish taste, taste systems of many fishes^{2), 3), 42) - 44)} were shown to parallel that of the catfish taste sensitivity to amino acids. Being highly responsive to amino acids allows the determination of the molecular characteristics required for stimulatory effectiveness, which is an advantage of gustatory receptors of fish. Electrophysiological recordings from the taste system of fishes support the contention that the amino acid spectrum is highly species-specific across species tested so far (Table I). Gustatory effectiveness of basic or acidic amino acids is strongly dependent on species, suggesting species differences of receptor sites^{2), 3)}. Generally, taste responses to amino acids for fish were highly stereospecific ; the L-isomer of an amino acid was stimulatory, but the enantiomer was not (Fig. 3). The gustatory receptors were responsive to amino acids containing 3 to 4 carbon atoms having unbranched and uncharged side chains (Fig. 4). Further, it seems that α -amino group and α -hydrogen were essential for maximal effectiveness, and that ionically charged primary carboxyl group was generally unnecessary for a maximal receptor response (Fig. 5). Representative examples as mentioned above for the carp taste system are shown in Figs. 3, 4 and 5. Consequently, the effectiveness of an amino acid molecule depends on its size, the arrangement of its atoms and the chemical properties of substituents in the side chain^{3), 6), 57)}.

As to the possible reasons why amino acids are potent chemosensory stimuli to fish, it is conceivable that those chemicals serve as a source for orientation to potential food organisms. However, the integrated facial

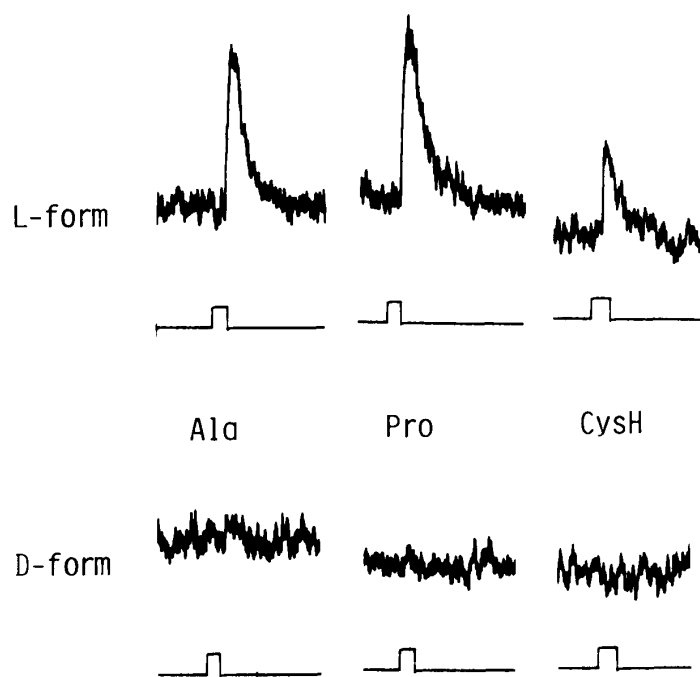


Fig. 3 Typical integrated responses upon stimulation of gustatory receptors with amino acids, showing that L-isomer of an amino acid is stimulatory, but its antiomer is ineffective. The results indicate that the receptors involved possess a corresponding chirality.

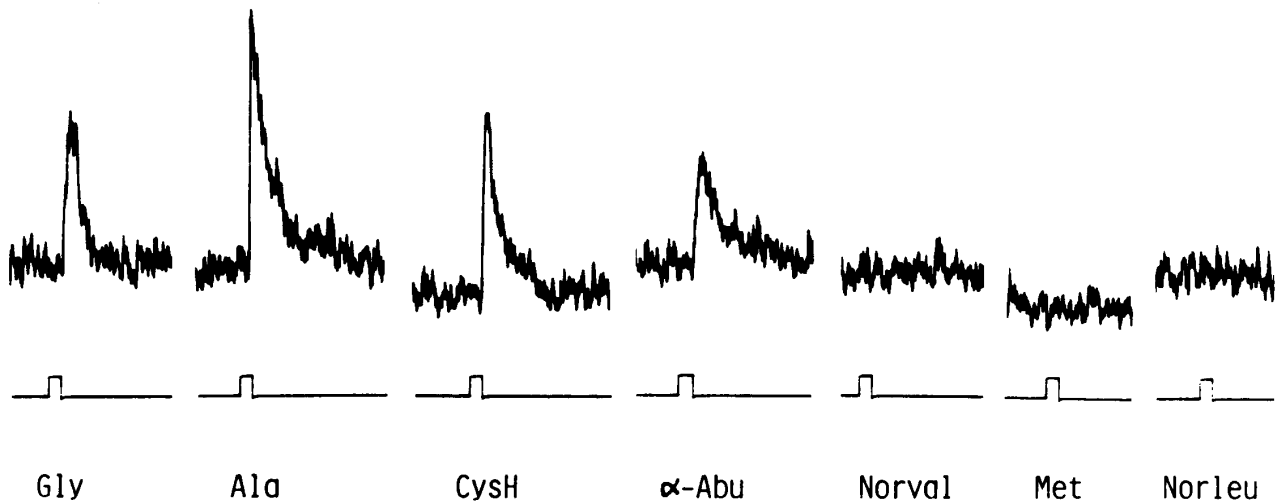


Fig. 4 The carp gustatory receptors are stimulated best by amino acids containing 3 to 4 carbon atoms and having unbranched and uncharged side chains.

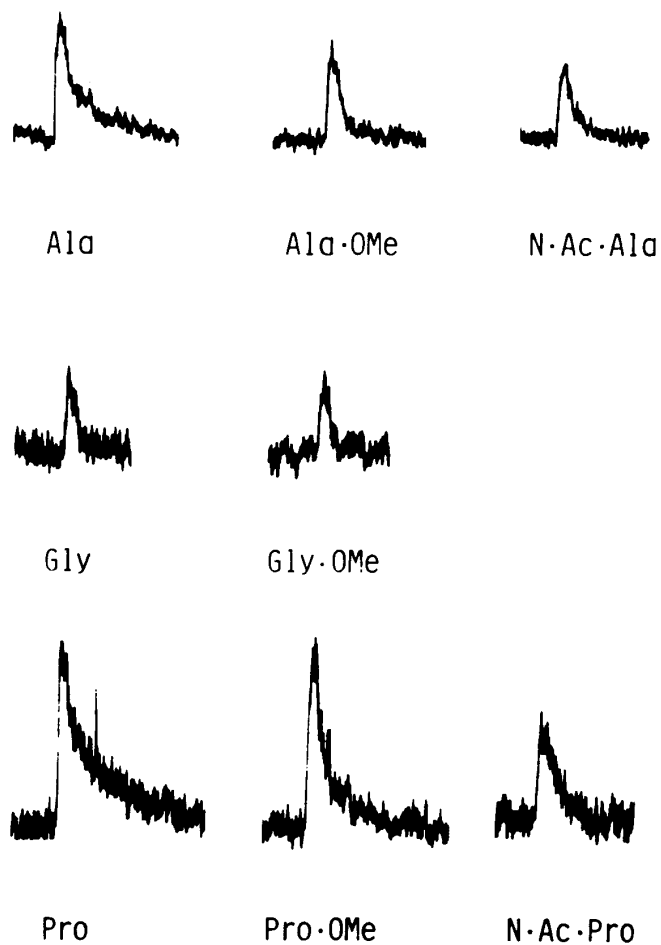


Fig. 5 Effects on gustatory neural responses due to structural changes in amino acids. Esterification of the primary carboxyl group resulted in little change in response magnitudes. However, replacements of the α -amino group significantly reduced the taste responses.

taste activity of some fishes^{44), 57)} is greater to carboxylic acids than to amino acids, and the concentration-response curves for some carboxylic acids were steeper than for the corresponding amino acid with increases in concentration (unpublished data in carp). Therefore, further attempts are desirable to know what chemical is the most adequate stimulus. On the other hand, it has been shown that certain mixtures of L-amino acids or extracts of prey animals were stimulatory in experimentally eliciting feeding behavior, while individual components of mixtures or extracts were inactive or weak stimulants^{50), 58)}. Also, for some fish, there is evidence that amino acids and betaine interacted synergistically to elicit feeding behavior^{50), 59), 60)}. Synergistic effects were obtained with a mixture of betaine plus amino acids for the gustatory system with electrophysiological techniques^{2), 44), 57), 61)}. Since betaine and some amino acids are contained in the extracts of prey animals, it is likely that betaine acts strongly as a feeding stimulant in conjunction with L-amino acids. Although most effective chemicals for the taste system in fishes are of lower molecular weight as amino acids, the mixtures like food extracts show highly stimulatory effectiveness for the system⁵⁷⁾.

III. TASTE PATHWAYS

C. J. Herrick^{23), 62)} confirmed essentially the primary afferent projections of the VIth, IXth nerves in fish. During the past decade, the primary and secondary gustatory connections of the facial and vagal taste systems in fish have precisely been reported by new anatomical neural tracing methods^{24)–30)}. According to Finger²⁵⁾, the sensory and recurrence roots of the facial nerve of the catfish (*Ictalurus nebulosus* and *I. natalis*) project together to the facial lobe via the anterior ganglion. Terminal degeneration of the sensory roots of the carp²⁴⁾ and the crucian carp²⁷⁾ was exclusively in the ipsilateral half of the facial lobe forming the so-called tuberculum impar^{20), 23)}, but differing from the catfish, the two roots of the carp and crucian carp enter the brainstem separately. In Siluridae (Ictaluridae), the facial lobes are paired structures with fiber fascicles dividing each lobe into several lobules^{25), 26), 30)}, which means more developed manner of the primary gustatory nucleus. The topographic organization of the facial lobe to each of the body regions have been reported in the catfish^{25), 35), 37)} and goldfish⁶³⁾ as presented in the carp^{34), 36)}. Taste and touch neurons in the medio-anterior part of the lobe in the carp are arranged, from dorsal to ventral, in order of head, upper jaw and upper lip, and those in the latero-posterior part are in the order of fin, gill-cover, lower jaw, barbels, and lower lip (Fig. 6). It has been revealed with electrophysiological techniques that taste and tactile neurons in the facial lobes of channel catfish were organized in a somatotopic manner³⁵⁾ as well. These results confirm the anatomical reports of their lobes^{24)–26)}. The secondary taste neurons in these fishes tend to be arranged generally in the more dorsal regions of the tactile sensitive areas of the facial lobe. In Fig. 7, typical gustatory unitary discharges in the facial lobe of the channel catfish are shown. The secondary gustatory neurons in the facial lobe responded with highest frequency to L-alanine or L-arginine applied to taste buds along the external body surface (Fig. 7), which is similar to that obtained from the peripheral neurons in the same species^{6), 40)}. However, there are some discrepancies among their somatotopic arrangements of these species. Although these discrepancies may be due to a real species difference or may merely reflect a difference in techniques, the differences of somatotopic manner within the facial lobe might be dependent on a developmental (specialized) degree of their gustatory centers. On the other hand, there is only one report concerning somatotopy and characteristics of visceral neurons in the vagal lobe of the channel catfish^{38), 64)}. The majority of the units observed in the vagal lobe were excited by mechanical stimulation and taste activity was generally restricted to the superficial region ($< 600 \mu\text{m}$ deep) of the lobe. Vagal unit responses to mechanical stimulation of the oral epithelium revealed that the anteroposterior axis of each lateral half of the oral cavity is represented ipsilaterally in an anterior to posterior direction in the vagal lobes. Specific structures, such as the palatal organ, gill arches and tongue region, are represented in somewhat diffuse and

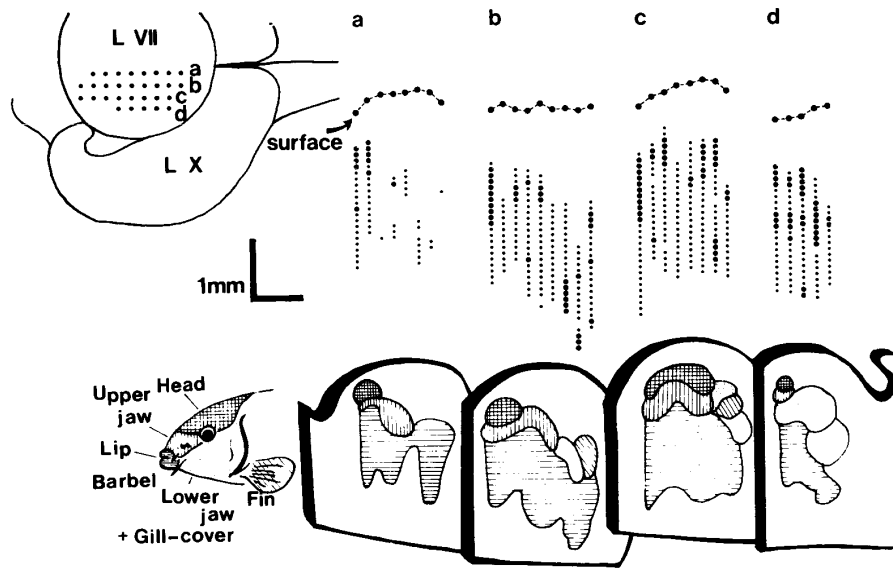


Fig. 6 Localization of taste and tactile neurons in the facial lobe and their topographical relation.

A: dorsal view of the facial lobe with 30 inserted electrode points, B: sagittal sections of one-half facial lobe at a, b, c, and d levels being at intervals of 0.25mm. Small closed circles indicate touch responding points in the lobe and large circles indicate taste points. C: division of the face with different marks. D: distribution of the 5 areas in the facial lobe. The marked regions correspond to those in the face.

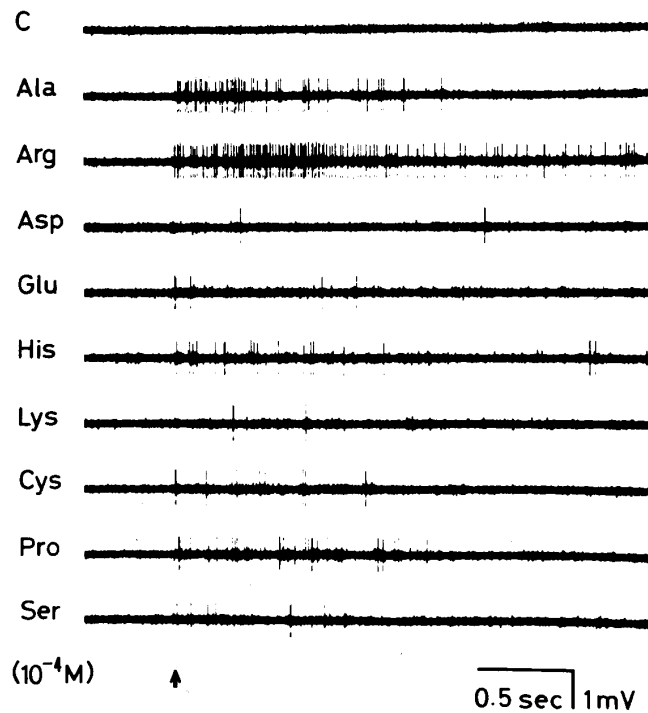


Fig. 7 Taste responses in a neuron of the facial lobe of the channel catfish to $10^{-4}M$ amino acids. This unit responded with highest frequency to L-Ala and L-Arg-HCl among several amino acids tested.

overlapping domains. Thus, the vagal lobe preserves, in general, the spatial organization and electrophysiological nature of the visceral sensory input, as observed in the facial lobe of the same species³⁵⁾.

From the facial lobe and the vagal lobe in siluroids and cyprinoids, a path made up of the axons of the secondary gustatory cells, runs ventralward and ascends ventral and medial to the descending root of the trigeminus²⁵⁾⁻³⁰⁾. As the secondary gustatory tract approaches the rostral end of the medulla, the tract breaks up into a number of fascicles, turns medially and enters the superior secondary gustatory nucleus (nGS)^{23), 28), 30)}. It has been revealed with anatomical^{26), 27), 30)} and electrophysiological⁶⁵⁾ techniques that the ascending secondary gustatory tract from the facial lobe projects bilaterally to the nGS^{26)-28), 30), 65)} in agreement with the description by Ariens-Kappers²⁰⁾. Judging from the extent of responses obtained electrophysiologically, projections of the ascending secondary gustatory or tactile axons to the nGS were ipsilateral, contralateral or bilateral. Most neurons in the nGS responded only to mechanical stimulation such as stroking the skin surface with a small brush or a galss probe, or with flowing water. The taste neurons were located in the tactile sensitive areas and approximately 95% of them gave tactile responses as in the facial lobe³⁴⁾ of the same species. A segregation of the facial taste system from the vagal one in the nGS was reported in these fishes^{26), 29), 30)}. It is also noted in the catfish by the anatomical observations that some degree of somatotopy exist in the second order projection to the nGS, because the vagal lobe projects most heavily to the lateral portion of the nGS and the facial lobe to more medial regions³⁰⁾ (Marui, Kanwal and Caprio, unpublished on the Japanese sea catfish). In the carp, some degree of somatotopy was confirmed in the nGS⁶⁵⁾, but a separation of terminal areas between the facial and the glossopharyngeal-vagal taste systems, which was also found in the crucian carp²⁷⁾, was not clarified in the electrophysiological experiments of the carp⁶⁵⁾. The topographical relation to each of the body regions in the nGS was ambiguous because of their wide receptive fields. The receptive fields were organized such that the most anterior fields are found in the most ventral portion of the nGS in the same manner as in the facial lobe. The size of the area of tactile sensitive neurons in ithmic region is about 6 mm wide, 1 mm long in front and rear, and 3 mm deep, while that of taste responsive neurons is about 2 mm deep centered within the tactile area⁶⁵⁾. However, the precise construction of the nGS in the carp is not appeared yet but Herrick's description²³⁾.

The tertiary gustatory tract projects ipsilaterally to the nucleus glomerulosus and the nucleus diffusus lobi inferioris of the diencephalon^{23), 26), 27), 66)} which might correspond to the tertiary gustatory nucleus. However, there is no agreement with the terminology of the thalamic regions in fish among investigators. Nobody either reported about the tertiary gustatory nucleus with electrophysiological techniques so far.

The descending secondary gustatory tract from the facial lobe is closely related to the trigeminal complex. This tract projects mostly to the nucleus of the spinal trigeminal tract and sparsely to the nFM^{23), 27), 62)}. However, there is no projections to these regions from the vagal lobe in the catfish³⁰⁾. The descending projections from the vagal lobe were recognized to occur to the nucleus commissure of Cajal and so on. It is indicated by Herrick⁶²⁾ that terminations of the secondary descending visceral tract were recognized within both lateral and medial funicular nuclei in the carp and *Ameiurus* (= *Ictalurus*), and these nuclei are regarded as gustatory-tactile correlation centers. Actually, tactile responses can easily be recorded there, where the latencies obtained by electrical stimulation to the peripheral receptive fields show rather short; i.e. the tactile information may terminate to the funicular regions directly through the trigeminal root (Marui, unpublished data). The two components concerning the gustatory sense, facial and vagal gustatory system, are dissociable and parallel within the vertebrate central nervous system²⁹⁾. The former system is to be used for food selection and the latter is to be involved in swallowing²⁹⁾, which confirms the earlier findings by Atema¹²⁾. It was also demonstrated by neural tracing methods^{29), 30)} that the primary gustatory nucleus for the facial system projects to the reticular formation and to trigeminal nuclei other than the main gustatory tract, and the nucleus for the vagal taste system does to the motor neurons (nucleus ambiguus) mediating swallowing. The

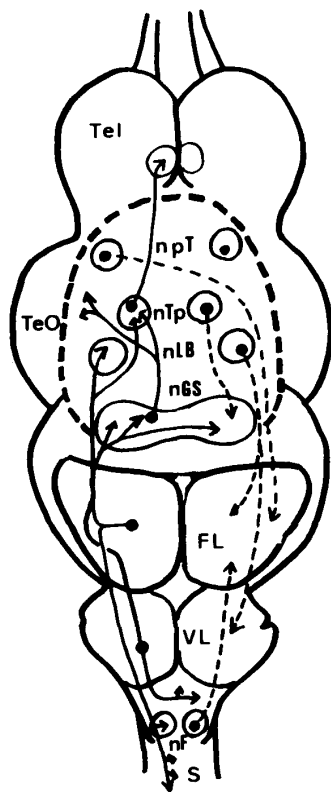


Fig. 8 Schematic diagram of a dorsal view of the catfish brain, showing the central fiber connections of the different gustatory centers. Solid lines represent ascending gustatory pathways from the facial and vagal lobes, and broken lines show descending gustatory pathways to the two lobes. Abbreviations: FL, facial lobe; nF, funicular nuclei; nGS, secondary gustatory nucleus; nLB, nucleus lobobulbaris; npT, pretectal nucleus; nTp, posterior thalamic nucleus; Tel, telencephalon; TeO, optic tectum; VL, vagal lobe.

whole projections of the gustatory systems of the Japanese sea catfish are simply summarized in Fig. 8, schematically showing different brainstem connections. Further neurophysiological experimental works will be necessary in parallel with anatomically neural tracing to test and confirm these taste pathways in detail.

IV. TACTILE INPUT TO THE GUSTATORY NUCLEI

It is believed that certain fishes developing their external gustatory system find their foods ordinarily by the simultaneous use of taste and touch^{5), 22), 62)}. In fish, the trigeminal and facial nerve components are intimately complexed peripherally as mentioned before so that it is difficult to discriminate one from the other, anatomically and functionally. However, the two senses belong to entirely different system of neurons, visceral and somatosensory, respectively. Thus, it becomes of great interest to solve a question of their connections. The lips, barbels and other facial skin of fish are highly sensitive to chemical and tactile stimulation^{43), 56), 67) - 72)}. Functional properties of mechanoreception in the carp and the channel catfish coincided with those of barbel mechanoreceptors in catfish, *Ictalurus nebulosus*⁷³⁾ and *Plotosus anguillaris*⁷⁴⁾. Position type neurons responded to a slight bend a more distal point on the barbel, so it is considered to be involved in stretch-sensitive receptors as in the catfish. This might indicate the connection for reflex movements of barbels. It is well-known that these regions are controlled by the cranial Vth and VIIth nerves. Taste buds on the facial skin surface have been thought to be innervated by the latter^{22), 23)} and the former is regarded as a tactile nerve carrying impulses from the same regions as innervated by the facial nerve⁶⁸⁾. Clarification is needed as to where and in what way the taste and tactile messages from the outer skin come into physiological relation to facilitate the ordinarily reflex feeding movements of fish. Several attempts were also made to shed light on the properties of the tactile input and tactile pathways³⁶⁾.

Fujiya and Bardach⁷²⁾ considered that taste cells in the yellow bullhead, *Ictalurus natalis*, are sensitive to very light touch. Electrical responses of the facial nerve induced by tactile stimulation of the facial skin have been reported in catfish^{56), 73)}, carp^{36), 69)} and salmon⁷⁰⁾. It is unlikely that the taste fibers themselves of the facial nerve carry impulses of tactile sensation, since Johnstone⁷⁵⁾ reported in an anatomical study in fish that the facialis contains general tactile fibers, apart from the taste fibers. There was also an electrophysiological study in the puffer, *Fugu paradalis*⁷⁶⁾ which showed that taste messages from the facial regions are transmitted by the communis only and tactile ones by the trigeminus mainly. The communis root does admittedly contain a small amount of tactile fibers. The sensory part of the trigeminus is regarded exclusively as a tactile nerve by Herrick²³⁾. Primary nerve cell bodies are densely distributed among axon bundles in catfish⁷⁷⁾ and carp²⁴⁾. Therefore, it was easily possible to obtain electrical activities of primary neurons in fish by tactile and taste stimulation with the microelectrode techniques. The electrical amplitude of the trigeminus is 10 times higher than that of the other primary nerves³⁶⁾. This may be attributed to a mixture of large and small cells originating from the Gasserian ganglion, while the geniculate is composed of small ones^{56), 78), 79)}. Therefore, the spikes evoked by mechanical stimulation are easily obtained as rather large responses from the trigeminofacial complex nerve under the metal hook electrode techniques. Tactile responses have been reported in the facial lobes of catfish^{35), 37)}, carp^{34), 36)} and goldfish⁶³⁾. Unit discharges in the facial lobe of catfish are generally situated at a depth of 0.5–3.2 mm. This range agrees with the results in the carp^{34), 36)} and in the channel catfish³⁵⁾ in which taste and tactile areas in the lobe were found at an average depth of 0.4–3.2 mm below the surface. The sensory trigeminus root, upon entering the oblongata, turns caudal to constitute the spinal V tract^{23), 26), 27), 30), 62), 80)}. There is no chief sensory nucleus at the level of entrance of the nerve^{24), 81)} and the funicular nucleus has been regarded as a tactile center in fish^{62), 68)}. The latency measurements of the tactile units obtained from nFM with electrical stimulation suggest mono-synaptic pass to this area (Marui, unpublished). Although purely taste loci were found in the facial lobe of the carp, almost all taste sensitive areas gave tactile responses^{34), 35)}. It has been considered that crossmodal responses were attributed to convergences of neural input in the central nervous system. Luiten²⁴⁾ verified by means of the technique of anterograde axon degeneration in the carp that the trigeminal fibers terminate in several areas of the brain, not in distinct areas. Bimodal responses in the facial lobe are consistent with the anatomical findings that the solitary nucleus in rat⁸²⁾ and cat⁸³⁾ receives trigeminal fibers, and the facial lobe of catfish, *Ameiurus nebulosus*, receives trigeminal collaterals from fibers passing to the spinal V nucleus⁶²⁾. Recently, it is revealed with neural tracing methods of HRP (horseradish peroxidase) that in the Japanese sea catfish, some part of the trigeminal root passing to the spinal V nucleus turn medially towards the facial lobe and then terminate directly in the lobe, which may be collaterals⁸⁰⁾. These anatomical findings support the observation that the facial lobes of the carp and the channel catfish fit in with the somatotopic arrangement of the tactile nuclei of trigeminal medullary components^{34)–36)}. Therefore, there must be a fibrous connection between the spinal V tract and facial lobe to correlate the diverse modalities. The facial lobe of fishes, then, is conceivable to be a primary center not only for the gustatory root of the VIIth nerve but also for the tactile root of the Vth nerve, and must be an important correlation station for simple short reflexes. Although latency measured by electrical stimulation of the end organs is not equivalent to stimulation of the nerves, the results of the two methods agree³⁶⁾. The latencies obtained in the carp³⁶⁾ of up to 59 msec (Fig. 9) are larger than Biedenbach's³⁷⁾ observation in the catfish, in which latencies of the facial lobe neurons were of 4–34 msec. The long latencies of electrically driven neurons suggest that some tactile input from the trigeminal root might project to the facial lobe via multisynaptic path. This supports partly the anatomical finding that direct fibers from the descending root of the trigeminus also pass into the deeper portion of the facial lobe²⁰⁾ and thus are correlated with tactile and gustatory impulses from homologous body regions.

The superior secondary gustatory nucleus of the carp is situated in the dorsal isthmus region^{20), 23), 65)} which

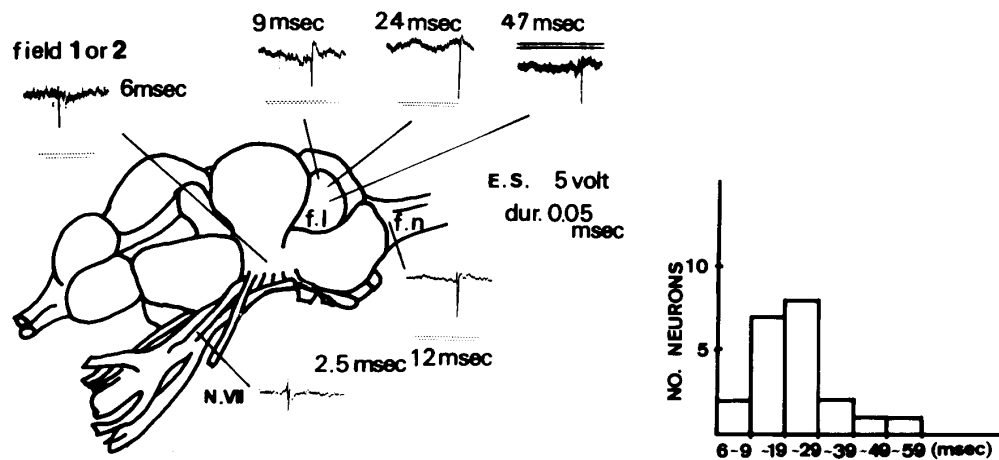


Fig. 9 The latency measurements in several regions of the brainstem of the carp by electrical stimulation to the trigemino-facial complex nerves at the base of a mandible barbel. The right graph shows distribution of latencies of evoked potentials measured in the facial lobe tactile neurons; 20 neurons evoked by electrical stimulation to the trigemino-facial complex nerves.

Abbreviations: f.l. facial lobe; f.n., funicular nuclei; N. VI, facial nerve.

is in front of motor nucleus of the trigeminal nerve and is in synaptic relation with this nucleus through the dendrites of motor neurons of the trigeminal nerve. This might be related to the extent of the tactile input in the isthmus region⁶⁵). The bimodal chemosensitive neurons converged by other inputs in the facial lobe may reflect on the tertiary gustatory neurons.

V. BEHAVIORAL IMPLICATIONS

A number of studies have shown dexterously that amino acids acting singly and in combination elicit feeding behavior in fishes^{84) - 87)}. Behavioral and electrophysiological results available for comparison indicate considerable overlap between effective gustatory stimuli determined for the channel catfish^{6), 47)}, the sea bream⁴¹⁾, eel^{44), 84)} and puffer⁵⁰⁾ (Table II). However, in the rainbow trout, the results obtained by both methods make us confusing. L-proline, which was in highest concentration in the synthetic squid extract, was inactive as feeding stimulant⁵⁸⁾, but was a highly stimulatory taste chemicals with electrophysiological test⁴³⁾; lysine, tyrosine and histidine were inactive physiological stimuli (Table II). Therefore, it is risky to apply the electrophysiological data to behavioral bioassay in a hurry; i.e. there is a possibility that olfaction may relate to the eliciting feeding behavior, because both gustatory and olfactory systems are capable of detecting amino acids^{1), 88)-91)}. Behavioral experiment is very important to the fish chemosensory field in confirming earlier information obtained electrophysiologically on amino acid sensitivity of the other fishes. Accomplishment of behavioral evaluation for stimulatory chemicals obtained from electrophysiological investigations could provide valuable insights and speculations into the difference and similarity of the effectiveness between results from both experimental procedures. Then, definitive statements concerning the correlations of both behavioral and physiological results should be made after accumulation of such data in numerous other species. If not so, our objectives will be misled to out of the truth. Thus, further experiments are required to determine if the gustatory sense is involved primarily in eliciting feeding behavior. Details concerning the role of gustation in feeding behavior should be referred to other reviews⁸⁵⁾⁻⁸⁷⁾.

TABLE II. POTENT AMINO ACIDS DETERMINED BY ELECTROPHYSIOLOGY AND BEHAVIOR.

Species	Electrophysiology	Feeding Behavior
1. Channel catfish (<i>Ictalurus punctatus</i>)	Ala, Arg, Pro ^a	Arg, CysH, Met ^b
2. Eel (<i>Anguilla japonica</i>)	Arg, Gly, Ala, ^c Pro	Arg, Ala, Gly ^d
3. Puffer (<i>Fugu pardalis</i>)	Gly, Pro, Bet, ^e Ala	Pro, Bet, Ala, ^f Gly
4. Rainbow trout (<i>Salmo gairdneri</i>)	Pro, Hyp, Leu, ^g Ala	(Tyr, Phe & His) ^h or (Tyr, Phe & Lys)
5. Sea bream (<i>Chrysophrys major</i>)	Ala, Gly, Arg, ⁱ Ser	Ala, Gly, Ser ⁱ

a. Caprio, 1975(40)

b. Holland & Teeter, 1981(47)

c. Yoshii et al., 1979(44)

d. Hashimoto et al., 1968(84)

e. Hidaka et al., 1976(61)

f. Hidaka et al., 1978(50)

g. Marui et al., 1983(43)

h. Adron & Mackie, 1978(58)

i. Goh & Tamura, 1980(2)

j. Goh & Tamura, 1980(41)

VI. GENERAL DISCUSSION

The objectives of our research are to obtain the critical information needed for describing how taste quality and quantity are represented in the peripheral nerves and to know how these neural inputs are processed in the taste centers of the brain. However, due to the complexity and time-consuming nature of these objectives, it has the advantage of improving the efficiency of the taste research that specific aims should be focused on some organisms specializing gustatory systems. Then the general meaning of the results obtained is to be drawn for our better understandings of the research. On the other hand, there is another difficulty in making rapid progress of the taste physiology. Differing from the other senses like vision or audition, no continuum of the stimulant for the chemical senses is found, which throws us into confusion. The significant parameters of the stimulus in the chemical senses are not understood yet, although a certain parameter is confirmed in the other senses like wavelength in the visual sense. Nevertheless, it is quite possible to come to considerable understanding of the neural message without knowledge about the significant aspects of the taste stimulus.

Indeed it appears to be many difficulties in the field of chemical senses, but specific advantages for using fishes as the appropriate model for the taste research include :

- (1) In certain fishes such as catfishes, the quantity and distribution of taste buds have increased and facially innervated extraoral taste buds are found on lips, head, barbels, fins, and flank^{12), 67)}.
- (2) Peripheral taste neurons are easily separated into small bundles in which highly responsive and durable individual taste fibers can be easily isolated and studied electrophysiologically.
- (3) In the taste system of fish, amino acids are known to be highly stimulatory and behaviorally relevant compounds.
- (4) Since the gustatory brainstem nuclei and other higher taste centers are specialized and consequently enlarged, electrophysiological investigations are readily accessible.
- (5) Somatotopic and overlapping taste-tactile neural maps of the external body surface (facial taste system) and oro-pharyngeal regions (glossopharyngeal-vagal taste system) onto medullary taste nuclei are known

are an aid in quickly defining chemosensory receptive fields.

Teleosts are probably the best developed of all vertebrates for detecting chemicals in their surroundings, using at least three different chemosensory systems (olfaction, gustation and the common chemical sense). They can identify chemical compounds in their environment with such chemosensory systems. At present, gustation is thought to have a close relationship to fish feeding behavior, the relative importance of olfaction may primarily be related to conspecific chemical communication concerning social behavior, and the common chemical sense detects the presence and location of various chemicals like noxious substances.

Problems concerning processing of the taste information in the central nervous system are as follows:

- (1) What is a pathway for reflex movements of the lips, barbels, tongue, jaws, gills and pharynx in response to gustatory stimulation ?
- (2) How does the taste pathway come into relation with the most complex reflexes of which the fish is capable ?
- (3) What are the roles of different cell types⁹²⁾ of gustatory nuclei ?
- (4) How are connections among gustatory nuclei constructed anatomically and functionally ?
- (5) How does the tactile information via the trigeminus play a role with the gustatory one via the facialis in feeding behavior ?

Thus, experiments should be planned at first to answer following questions concerning the processing of amino acid taste information across peripheral, medullary, pontine, thalamic and telencephalic levels of neural organization.

- (1) Do taste sensitive neurons that respond with highest frequency to certain amino acids belong to distinct classes at each synaptic level ?
- (2) Is taste input to taste nuclei of the brain modified by synaptic processing ? If so, how ?
- (3) How prevalent are bimodal (taste and tactile) units at all synaptic levels ? Is there evidence for taste-tactile interaction ?
- (4) Is there evidence for taste vs the other senses interaction in these taste pathway?

These experimental protocols are to be performed with both neuroanatomical and neurophysiological techniques, which might provide the basic concepts of the neural structures on taste pathways and processing of taste information in vertebrates including man.

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