

ACChemS V Abstracts

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1

Fine structure of odor plumes in aquatic environments in relation to chemoreception. JELLE ATEMA, BRUCE BRYANT and MARK MURRAY-BROWN (Boston University Marine Program).

To bridge the gap between physiological response properties and behavioral function of chemoreceptive organs it is necessary to have a quantified temporal-spatial picture of the chemical stimulus concentrations and compositions in the environment at the size scale of individual receptor structures. Two approaches have proven useful to describe such a stimulus field in aquatic environments: distribution of salt solution in distilled water detected with a bridge circuit and dye dispersal measured with a spectrophotometer. Important parameters are probe size, dynamic range, temporal and spatial resolution and reproducibility.

We built an AC bridge circuit with a spatial resolution of 1 mm between the platinum electrodes and temporal resolution of less than 1 ms. Its dynamic range was from 10^{-8} M to 10^{-1} M NaCl with discrimination of concentrations of half log steps. In the spectrophotometric approach we used a fiber optic probe to measure fluorescein dye in a flowing water flume. Path length -and spatial resolution- of the probe was 2 cm and its temporal resolution 100 ms. Its dynamic range was from 10^{-8} to 10^{-3} g/l fluorescein with quarter log step discrimination. The dye method can be applied in both fresh and seawater.

Measurements on turbulent dispersal in currents over different substrate types indicate significant differences in fine structure with substrate type and distance from the source. Fine structure of odor plumes may carry biologically significant information. Narrowly-tuned chemoreceptors are theoretically better capable of measuring dynamic stimulus patterns than broadly tuned receptors. This possibility is under investigation with lobsters, where several different narrowly-tuned units have been found in both antennules and legs.

Supported by grants from The Whitehall Foundation and NSF (BNS 8210434).

3

Rapid and permanent loss of memory for tastes and odors. LEWIS M. BARKER (Baylor University), CHARLES A. WEAVER, III (Baylor University).

We investigated the fate and time course of memory formation for a taste stimulus (15% sucrose solution) and for olfactory stimuli (1.33 and 5.0 PPM pyridine solution). 336 volunteers tasted 10 ml of 15% sucrose in distilled water and were given instructions to remember the strength of the solution. Subjects then ate a saline cracker and drank tap water. They were retested with either 5, 10, 15, or 20% sucrose at one of 4 delay intervals (1, 5, or 15 min, or 72 hr). They were asked to compare the 2nd stimulus with the 1st stimulus and say whether the 2nd was "less sweet," "the same," or "sweeter." Subjects reliably reported that 5% sucrose was less sweet and that 20% sucrose was sweeter. However, when the 2nd stimulus was 15% sucrose (i.e., = the standard), approximately 60% of the subjects reported that it was sweeter, regardless of the delay interval between the two stimuli. About 2/3 of those tasting 10% sucrose reported that it was = the 15% standard, regardless of the delay interval. (We then had 20 additional subjects taste 15% sucrose followed by 15% sucrose at delays of 15 and 45 sec. 8 of 10 of each group reported the second stimulus as sweeter using a magnitude estimation procedure.) The same pattern of results was also found for the olfactory memory test. When asked to match either a 1.33 or 5.0 ppm pyridine olfactory stimulus, subjects consistently picked a concentration weaker than the standard. Apparently, olfactory and taste stimuli are remembered as being substantially weaker as soon as the memory for these stimuli can be tested. These findings extend earlier observations on the phenomenon of a negative Time Order Error (TOE) when making a comparison judgment between two identical stimuli separated by only a few seconds (Guilford, 1954). The memory loss is apparently permanent, and is predicted by a "sinking trace" theory (Koehler, 1923).

2

Main and Accessory Olfactory Systems--A Reassessment. RICHARD S. BABB (Iona College, New Rochelle, N.Y.) and PAULINE JIRIK-BABB (Institute for Health Maintenance, New York, N.Y.).

The currently accepted view on the status of the main and accessory olfactory systems is that the main olfactory system is a conservative (older) structure with the accessory olfactory system being a later addition. There is mounting evidence to suggest that this is not so and that in fact the reverse may be the case. The accessory, unlike the main olfactory epithelium, does not exhibit cilia or possess Bowman's glands. In this respect the accessory olfactory epithelium is similar to that of the shark olfactory epithelium, and raises the possibility that the main olfactory epithelium of tetrapods might be a new structure adapted to receive olfactory stimuli in a non-aqueous environment, while the accessory olfactory epithelium would be an old structure common to all vertebrates. In agreement with this idea the accessory bulb is the first olfactory area of the prosencephalon to differentiate during ontogenetic development. More medially placed olfactory axons then induce the differentiation of the main olfactory bulb from the accessory bulb (Clairambault, 1976). Also in the embryonic mouse, neurons of the accessory olfactory bulb develop earlier than those of the main olfactory bulb, an order of development suggesting that the accessory is also phylogenetically older than the main olfactory bulb (Hinds, 1968). The accessory olfactory bulb projects unmyelinated fibers to the amygdala, a part of the telencephalon which is the first to appear when the human hemispheres begin to develop. The main olfactory bulbs, on the other hand, project myelinated fibers to the cerebral hemispheres. Based on this mounting evidence found at the three levels of each system, one might hypothesize that it is the main olfactory system which is the recent addition and not the accessory olfactory system. Such a theory would explain the evolution of the main olfactory system in terrestrial vertebrates as one which would have enabled the detection and discrimination of ground odors. In addition the expansion of the cerebral hemispheres would have provided the extra neural capacity required to process this new olfactory information.

4

Saliva As a Source of Gender and Genotypic Identification: We Must Remember This, a Kiss Isn't Just a Kiss. RONALD BERNHARD and CHARLES J. WYSOCKI (Monell Chemical Senses Center, 3500 Market Street, Philadelphia, PA 19104)*

Male and female mice frequently engage in snout-to-snout contact, "kissing," which may foster the exchange of chemosensory information contained in saliva. We speculated that chemosensory cues in female saliva influence the behavior of male mice and, more importantly, the male may also obtain information about the female's genotypic individuality from these salivary cues. We tested this hypothesis using the mouse ultrasonic vocalization assay: male mice generally emit vocalizations in the presence of female cues, but not male cues. We found that heterosexually experienced male mice emitted vocalizations when given access to female, but not male, saliva. Furthermore, the genotype of the female donating the saliva influenced the frequency of response. Males having experienced only one genetically distinct strain of female mouse vocalized in response to saliva from a female of that inbred strain significantly more than to saliva from a female of a genetically different strain. Finally, using inbred and congenic lines of female mice differing only at the Major Histocompatibility Complex, H-2, located on chromosome 17, we determined that selective exposure of the male to a female of one H-2 type resulted in a similar differential responsiveness to saliva from females of the same vs. different H-2 type. We conclude (i) that salivary cues influence the production of ultrasonic vocalizations, a presumptive reproductive behavior in mice, (ii) that genotypic differences at H-2 are expressed in the saliva of females and provide chemosensory information and (iii) that a male's prior experience determines, in part, his responsiveness to these salivary cues.

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5

Increased Dietary Salt Increases Preferred Level of Salt

M. BERTINO, G.K. BEAUCHAMP, Monell Chemical Senses Center, and K. ENGELMAN, Hospital of the University of Pennsylvania*

We have found that reduction in dietary sodium causes lower levels of salt in food to become most preferred. Here we attempted to determine whether increasing dietary NaCl increases salt preference?

Male and female university students were recruited as subjects and tested weekly for 11 weeks. Testing for the first 3 weeks established baseline taste perception of NaCl in soup. Subjects were then divided into 3 treatment groups. Group 1 subjects added 12 g crystalline NaCl to their food daily for 4 weeks. Group 2 ingested 12 g NaCl in enteric-coated tablets daily for 4 weeks. Group 3 ingested placebo capsules. All subjects were told they were ingesting 12 g NaCl each day. At the end of the 4 week treatment period, subjects stopped ingesting their supplemental substances. Compliance was assured by collecting 24-hour urine samples weekly and then analyzing for Na and creatinine content.

The most pleasant concentration of NaCl (breakpoint) in soup increased in Group 1 during the treatment period (when they were adding NaCl to their food) and remained high during the 4 week post-treatment period. There was no change in the concentration of the breakpoints of NaCl in the placebo group during the study.

These results indicate that increasing the intake of NaCl caused the most preferred concentration of NaCl in soup to increase. This preference change endured beyond the period where salt was added to food.

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7

Estrogen Treatment of Female Rhesus Monkeys enhances the Volatilization of Short-Chain Fatty Acids from Vaginal Secretions. ROBERT W. BONSALL and RICHARD P. MICHAEL (Department of Psychiatry, Emory University School of Medicine and Georgia Mental Health Institute, Atlanta, Georgia 30322.

Volatile fatty acids are present in the vaginal secretions of all Old World primates studied to date, and laboratory tests with rhesus monkeys indicate that males respond to the odor of these acids (copulin) as sex attractants in the same way as they do to the odor of estrogen-stimulated vaginal secretions. The production of short-chain acids by the microbial flora of the vagina depends upon substrate being made available by estrogen stimulation but these acids persist in the vagina, for example during the luteal phase of the cycle when the female's attractiveness declines. However, the strength of the olfactory signal depends on factors other than concentration, such as pH and the presence of protein, that influence the volatility of the acids. We have recently devised a means of measuring the vapor concentration directly. Vaginal secretions are collected on a special Teflon probe, and fatty acids volatilized from secretions are trapped quantitatively from a metered sample of air onto tris (hydroxymethyl) aminomethane coated on glass wool. The acids are subsequently desorbed at 140°C and transferred directly into the gas chromatograph for analysis. We have compared the rates of volatilization of acids from secretions collected from ovariectomized females before and during estrogen treatment (10 µg E.B. per day). Although the weight of material collected increased only 25% during estrogen treatment (30.8 ± 6.2 mg, N = 6 to 38.5 ± 5.3 mg, N = 12), the amount of C2 - C6 acids volatilized increased more than elevenfold. (67.7 ± 20.4 ng/hr to 755.6 ± 158.0 ng/hr).

Supported by Grant MH19506 from the National Institute of Mental Health. General support from the Georgia Department of Human Resources.

6

Studies of the Role of the Anomeric Centre in the Sweetness of Sugar Molecules GORDON G BIRCH

The glycopore of fructopyranose types of molecule appears to reside exocyclically presumably due to the ²C₅ conformations of such structures. On the other hand, the primary glycopore of glucopyranosides of structure, (⁴C₁ conformation) are probably located endocyclically at the 3,4 α-glycol group. The absence of involvement of the anomeric centre in these latter types of sugar is substantiated by the sweetness of 1-deoxy D-glucopyranose and 1-deoxy D-mannopyranose. Moreover, the threshold sweetness values of glucose syrups are similar to those of their hydrogenated counterparts. Recent intensity/time studies of the sweetness of freshly dissolved crystalline α- and β-D-glucopyranose demonstrate no significant differences between the anomers in either intensity or persistence of response and no differences occur during the mutarotation process. This result accords with previous reports of the sweetness of methyl α- and β-D-glucopyranoside.

8

Mammalian Facial Nerve Taste Systems. JAMES C. BOUDREAU, LATHA SIVAKUMAR, LY THI DO, JOSEPH ORAVEC, NGA KIEU HOANG, and THOMAS WHITE (Sensory Sciences Center, University of Texas at Houston).

On the basis of single unit recordings from the geniculate ganglion of the dog, cat, goat and rat, we propose that the mammalian facial nerve taste neurons can be subdivided into at least 4 distinct taste systems: a Brønsted acid system, an amino acid system, a salt (Na, Li) system, and a small fiber X system. Each system (save the X system) has well defined chemical input signals. X units are classified in large part on the basis of distinct neurophysiological characteristics, although in all species some X units discharge to alkaloids. The amino acid units of the rat are distinct in many ways from the amino acid units in the cat and dog and are not clearly distinguishable from rat X units. These four systems can be equated with neural groups described by others in the chorda tympani. Furthermore, all neural groups adequately described in the chorda tympani can be classified in one of these four taste systems. Certain systems may be lacking in some species (e.g., the dog and cat have no salt system and the goat has no amino acid system) or the acting stimulant may be somewhat different (e.g., the amino acid system of the dog, unlike that of the cat, is also responsive to sugars). We further speculate, on the basis of human whole nerve recordings and psychophysics, that the human has all four systems, with the amino acid system (and possibly Brønsted acid system) being more like the carnivore (especially the dog) than like the rat, another omnivore.

Research funded in part by the National Science Foundation.

Quantitative Comparison of Non-nutritive Sweetener Intensities. L. M. BRAND (The Procter & Gamble Company, Cincinnati, OH).

The sweet taste intensity of a non-nutritive sweetener is commonly described as the multiplier needed to give an iso-sweet concentration (w/v) of sucrose. Saccharin, for example, is considered to be "350 times as sweet as sucrose". However, this practice is inadequate for the evaluation of quantitative structure-activity relationships (QSAR) among these molecules, since sweetness potency of these agents has been observed to vary as a hyperbolic function of concentration. Sweetness potency data were obtained for a wide variety of sweeteners using a trained taste panel. Linear plots of (perceived sucrose equivalence)⁻¹ vs [sweetener]⁻¹ indicated a good fit to a modified Michaelis-Menten equation describing reversible ligand binding to a saturable receptor: $c = C_m [S] / (K_d + [S])$, where c is perceived sucrose intensity, $[S]$ is sweetener concentration, C_m is the extrapolated maximum sweetness intensity, and K_d is the apparent dissociation constant for the taste receptor-ligand interaction. C_m values did not differ greatly among the sweeteners tested (averaging 17.6% sucrose equivalence), indicating the similarity of the involved sweet taste mechanisms. Some observed K_d values were 5.7 mM (AspartameTM), 2.2 mM (saccharin) and 0.5 mM (6-chloro-D-tryptophan). The experimentally determined values K_d and C_m/K_d are concentration-independent constants for each sweetener, suitable for use in QSAR studies. The absolute magnitudes of the K_d values indicate that sweet taste perception in man involves relatively weak, freely reversible ligand-receptor interactions.

Effects of Monovalent Salts on Reduced Pyridine Nucleotide Fluorescence from Catfish Barbel. J.G. BRAND, S.J. KRON, D.M. SENSEMAN (Monell Center, 3500 Market St. and School of Dental Medicine, Univ. of PA, Phila. 19104).

Changes in the metabolic state of a tissue may be inferred by observing temporal fluctuations of intrinsic pyridine nucleotides. We observe reduced pyridine nucleotide fluorescence from a small (100 μ^2) taste-but-containing patch of the catfish barbel epithelium. The apparatus consists of a fluorescence microscope with a stabilized 100 watt Hg arc in an epi-illumination conformation. The 366 Hg line is isolated by an interference filter and reflected by a dichroic mirror through a 40X water immersion objective onto the surface of a barbel of an albino channel catfish. Fluorescent light is filtered to 450 \pm 5 nm into a blue sensitive photomultiplier. An iris restricts the epithelial field from which fluorescence is observed. The catfish is paralyzed with Flaxedil, mouth perfused and its barbel extended into an isolated stimulus chamber. The barbel is held immobile onto a Silgard base by micro-clamps that surround but do not penetrate the tissue. Blood flow through the major barbel vessels is used as an index of the viability. The basic observation is that monovalent halide salts (NaCl, LiCl, KCl at 5-200 mM) increase observed (presumably reduced nucleotide) fluorescence in a graded manner. Initiation of the increase occurs as rapidly as stimulus delivery (time constant of the instrument is ~ 0.5 sec). Flowing KCN over the barbel results in a large increase in fluorescence. This result is congruent with the assumption that CN⁻ is entering the cells and reducing available oxidized nucleotides. Adding even high concentrations (200 mM) of NaCl to the tissue previously reduced by CN⁻ results in no further increase in the fluorescence signal. The sensitivity of the system allows about 5% of the available nucleotide to be observed. The threshold for the increase in fluorescence due to NaCl is between 25 and 50 mM. These observations are consistent with the hypothesis that the salts (and CN⁻) can cross the epithelial barrier and affect metabolic activity.

Supported in part by grant from Whitehall Foundation, Inc.

Inhibition of Behavior and Settlement of Barnacles by Substances from Gorgonians. E.S. BRANSCOMB, D. RITTSCHOF, I. HOOPER and J.D. COSTLOW (Duke University Marine Laboratory, Beaufort, NC 28516)

Cyprids, the larval settling stage of barnacles, were used in behavior and settlement bioassays to screen for antifouling activities in molecular preparations of soft corals (*Leptogorgia virgulata*). Assays compared responses of cyprids of the Balanoid barnacle *Balanus amphitrite* in both the presence and absence of barnacle settlement factor, and of soft coral preparation.

Soft coral was homogenized, centrifuged, lyophilized, resuspended in distilled water and dialyzed. Molecules from the dialysate soluble in 80% methanol were tested and further fractionated. Prior to testing or further fractionation, methanol was removed under vacuum. For testing all material was redissolved in seawater at a concentration reported as mg/ml of original homogenate.

Starting material (that in 80% methanol, see above) inhibited both investigative behavior and attachment at 2mg/ml. Passage of starting material through a C18 Sep-pak (octadecyl silane-bonded silica) separated the two inhibitory activities. Unadsorbed material affected investigative behavior of cyprids. A 100% methanol eluate of the material adsorbing to the Sep-pak inhibited attachment. Both activities were observed in the presence of and the absence of settlement factor at an inhibition concentration of 4mg/ml. Tests of inhibition at 8 mg/ml failed to detect crossreactivity of the two activities. Settlement inhibition was significantly reduced in the presence of settlement factor. Inhibition of settlement appears to be via a non-toxic surface alteration.

This was supported by ONR Contract #N00014-78-C-0294.

The Nature of Residual Associative Taste Processing Capacities in Rats Lacking Gustatory Neocortex. JAY BRAUN and NANCY BOWMAN (Arizona State University, Psychology Department, Tempe, AZ 85287)

In previous work we have shown amnesic taste disturbances and disruptions of taste learning to be ubiquitous consequences of gustatory neocortex (GN) ablation, at least for taste avoidance habits. The effects are specific to GN damage because equivalent damage to adjacent cortical areas has no effect on taste learning or memory. Furthermore, the effects following GN ablation are specific to taste because there is little disruption of identically instated odor habits. Our current work focuses on the capacities displayed by rats lacking GN for learning or relearning discriminatively specific avoidance responses to taste stimuli. First, while the olfactory system seems little involved in learned taste habits normally, it becomes profoundly involved in absence of GN. Second, rats lacking both olfactory bulbs and GN display a coarse residual associative taste capacity. Third, and most interesting, evidence will be presented supporting the idea that the residual taste avoidance learning capacity may be based upon a "bitter" rejection reflex.

Unilateral Odor Deprivation: Differential Effects Due to Time of Onset. P.C. BRUNJES & M.J. BORROR. Dept. of Psychology, Univ. of Va. Charlottesville, Va. 22901

Considerable attention has been directed at describing the anatomical consequences of unilateral odor deprivation. The general procedure is to occlude (usually by cautery) single nostrils of animals within a few days after birth. Approximately 30 days later, subjects are sacrificed and the structure of the "deprived" olfactory bulb (ipsilateral to occlusion) compared with that of the "control" bulb. Typically, deprived bulbs are 25-30% smaller than control bulbs, due to changes in the volume of nearly all bulb layers. The present study was an attempt to determine if occlusion-induced changes are peculiar to the preweanling period. Rats were unilaterally occluded on either 1, 30 or 60 days postpartum and sacrificed 30 days later. Brains were horizontally sectioned at 30 μ m and stained with thionin. Volume of the glomerular layer (GLM), external plexiform layer (EPL) and inner granule layer (IL) of both deprived and control bulbs was determined by measuring serial microprojector images with a computer-controlled graphics tablet system. Findings from animals occluded on Day 1 and sacrificed on Day 30 mirror those reported previously: approximately 25% reduction in bulb size with the EPL exhibiting most change, followed by the IL and GLM. However, only very small changes were detected in animals occluded on Day 30 and sacrificed on Day 60: deprived bulbs were less than 2% smaller than controls. Percentage differences from control bulb size were 4.8, -7.8 and -1.6 for GLM, EPL, and IL, respectively. As expected, preliminary results from animals occluded on Day 60 and sacrificed on Day 90 also show only a small (3.3%) difference in deprived vs. control bulb size. Therefore, unilateral deprivation does not consistently result in atrophy of the olfactory bulb; deprivation induced changes only occur during early life.

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Temporal Integration in the Common Chemical Sense
WILLIAM S. CAIN, J. ENRIQUE COMMETO-MUNIZ* (John B. Pierce Foundation Laboratory and Yale University).

Four experiments explored possible temporal summation in olfaction and the common chemical sense. In the first experiment, participants judged the perceived magnitude of various concentrations and durations (1.25 - 3.75 sec) of the pungent odorant ammonia and the nonpungent odorant isoamyl butyrate. The perceived magnitude of ammonia increased with duration of inhalation whereas the magnitude of isoamyl butyrate did not. Time-intensity trading relations for ammonia indicated nearly perfect temporal summation. In the second experiment, modulation of the concentration of ammonia during an inhalation led to assessments of perceived magnitude that confirmed the high degree of temporal summation seen in the first experiment. That is, approximately equal time-integrated mass of inhaled ammonia led to approximately equal perceived intensity. The third experiment indicated that temporal summation for ammonia arose from its pungency rather than from its odor, the fourth that trigeminally-mediated reflex apnea in response to ammonia also exhibits temporal summation. The degree of temporal summation measured with the reflex came very close to that assessed psychophysically. When stimulated with ammonia for short durations, the common chemical sense behaves more like a total-mass detector than a concentration detector.

*Fellow of the Consejo Nacional de Investigaciones Científicas y Técnicas, República Argentina, on leave from the Laboratorio de Investigaciones Sensoriales (CONICET-Fac. de Medicina, UBA), Buenos Aires, Argentina.

Conspecific Chemical Stimuli in Catfish: Responses in a Social Context. BRUCE BRYANT and JELLE ATEMA (Boston University Marine Program, MBL, Woods Hole, MA 02543).

The brown bullhead catfish (*Ictalurus nebulosus*) exhibits territorial/dominance behavior. Earlier work has implicated but not demonstrated the functional significance of conspecific chemical stimuli in this behavior. This work seeks to extend others which have studied chemosensory social behavior outside of a social context.

To measure unconditioned responses to chemical cues, body odor (tank water) from single males and females was siphoned into the tanks of male-female pairs of fish. The behavior of these pairs was measured during control and stimulus introduction periods, by recording frequencies of stereotyped behavioral acts. Generally, fish oriented first to the odor source, then to its tankmate. This was frequently followed by aggressive interactions initiated by the males.

A source of body odor from donors of both sexes was highly attractive to both fish; attraction to heterosexual odor was the strongest. After this initial attraction both fish displayed the solitary behavior 'scratch'. This occurred more to male than to female donor odor and may be substrate marking behavior. Male odor elicited more aggressive interaction between the fish than female odor and the aggressive response of a given fish was stronger during the introduction of odor from a donor of the same sex. In a few instances, fish displayed aggressive behaviors directed at the odor stimulus or source rather than at the other fish.

This study shows that gender information is available in body odor and that it is used. It also shows that responses to body odor alone are consonant with observed patterns of territorial behavior.

Interactions between Sucrose-caffeine and Coffee-sugar beverage. AMALIA MIRTA CALVIÑO and MARIA ROSA GARCIA-MEDINA (Laboratorio de Investigaciones Sensoriales, CONICET, Facultad de Medicina, UBA, C.C.53, 1453-Buenos Aires, Argentina).

The following data show suppression for sweetness and coffee flavor intensity when they are mixed with caffeine and sugar respectively.

First, unmixed caffeine (0.5 and 1.0 % W/V) and sucrose solutions (5, 10, 20, and 40 % W/V) and their mixtures were presented and rated for their sweetness and bitterness. Second, nine mixtures at three different ratios of sucrose/caffeine molar concentrations were evaluated similarly.

It was found that the increase of caffeine in the mixture decreases the sensitivity for sweetness. In the other hand, for a given value of caffeine bitterness was negatively correlated with the increase of sweetness intensity.

When 3 or 6 % W/V of sugar were added to five different coffee levels (5, 10, 20, 40, and 100 % V/V) flavor coffee intensity decreases in the same manner as in taste-taste mixtures.

Behavioral Responses to Adenosine 5'-Monophosphate and Analogs Suggest a Marine Shrimp has External Chemoreceptors Related to Internal Adenosine Receptors of Vertebrates. WILLIAM E.S. CARR (Whitney Marine Laboratory of the University of Fla., Rt. 1, Box 121, St. Augustine, FL 32084), HILARY W. THOMPSON (Dept. of Zoology and Physiology, Louisiana State University, Baton Rouge, LA 70803).

A vast literature exists on the role of adenosine and its nucleotides in vertebrate animals as activators, inhibitors of modulators of many activities in the brain, heart and elsewhere. Recently we have found that the nucleotide adenosine 5'-monophosphate (AMP) is a potent chemoattractant for the marine shrimp, *Palaemonetes pugio*. The AMP dose-response curve is biphasic with response increasing with dose up to about 10 μ M and then declining at higher concentrations. The response to AMP is antagonized by theophylline and adenosine. Bioassays of substances structurally related to AMP reveal that the integrity of both the adenine and the ribose phosphate moieties are required for maximal activity. However, the integrity of the ribose phosphate is of special importance since most substances having modifications here are completely inactive as attractants. The relationships of structure and activity, and antagonism by theophylline, suggest the shrimp has external chemoreceptors with marked similarities to the R-type adenosine receptors found coupled to the enzyme adenylate cyclase in certain vertebrate tissues. These findings provide further support for the notion that internal receptors in higher organisms evolved from external chemoreceptors of more primitive organisms.

Effects of hemodialysis on chemosensory function and zinc metabolism. F. CATALANOTTO, N. RYAN, L. ALLEN, M. OSTROM, J. SCHLITZER & A. SOLINSKY; Departments of Pediatric Dentistry and Nutritional Sciences, Connecticut Chemosensory Clinical Research Center, University of Connecticut Health Center, Farmington, CT.

Thirty-nine patients with end stage renal disease (ESRD) receiving 2X or 3X weekly hemodialysis and 36 age- and sex-matched control subjects (CONS) participated in the study. Blood for several zinc parameters was collected before and after dialysis from ESRD and once from CONS. For the taste test, all subjects were given 5 concentrations each of NaCl (N), sucrose (S), citric acid (C), and quinine hydrochloride (Q) and a water blank, in a suprathreshold scaling, magnitude estimation task. Taste tests were given just before and immediately after dialysis from ESRD subjects and several hours apart for CONS. Taste data were analyzed with 2 different scoring systems developed in our laboratory as well as by plotting standard psychophysical functions.

Patients undergoing hemodialysis demonstrated decreased plasma zinc values but increased RBC zinc concentrations. Patient plasma zinc values increased after hemodialysis with most, but not all, of the increases accounted for by hemoconcentration. There was a weak, but marginally significant, negative correlation between taste scores and zinc levels.

The results of the chemosensory tests demonstrated mixed results. Standard psychophysical functions of the ESRD and CONS suggested no significant difference between the two groups. The scoring systems developed in our laboratory suggested a significant impairment of taste function in the subjects undergoing hemodialysis. We are currently analyzing these data in more detail to determine the significance of these disparate results. Further studies in these subjects are being pursued to more clearly define the chemosensory status of patients with ESRD as well as any potential interaction with zinc metabolism and/or nutritional status.

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Taste sensitivity and zinc metabolism in patients with multiple sclerosis. F. CATALANOTTO, P. DORE-DUFFY, J. DONALDSON, K. M. OSTROM, M. TESTA, M. PETERSON, Depts. of Pediatric Dentistry & Neurology, Connecticut Chemosensory Clinical Research Center, University of Connecticut, Farmington, CT.

64 patients with Multiple Sclerosis (MS) and 59 age & sex matched control subjects (CONS) were studied. Fasting blood samples for zinc were collected between 8 a.m. and 12 noon from MS and CONS. Subjects were given 6 concentrations each of NaCl (N), sucrose (S), citric acid (C), and quinine (Q) hydrochloride and a water blank in a suprathreshold scaling, magnitude estimation taste test. Taste data were analyzed with a scoring system developed in our laboratory as well as by plotting standard psychophysical functions.

Patients with MS demonstrated plasma zinc levels that were within normal limits but also showed a significant increase in the serum zinc carrier protein, α_2 -macroglobulin, a significant decrease in α_2 -bound zinc and a significant increase in red blood cell bound zinc; in addition, RBC bound zinc seemed correlated with disease severity.

Analyzing MS taste sensitivity using our scoring system and the standard psychophysical method gave strikingly similar results. Results of the scoring system were analyzed with an ANOVA by group (MS & CONS) with age as a covariant and demonstrated a significant alteration of N & Q sensitivity for 5 of 6 derived measures. The scoring system demonstrated a significant effect of age but not length or severity of disease on taste function. In addition, MS patients with craniofacial lesions were not significantly different than other MS patients. The standard psychophysical analysis of the data demonstrated a significant flattening of the slope of the function for N in MS patients ($p < 0.01$) but a significant steepening of the function for Q ($p < 0.01$).

Further studies are being pursued to more clearly define the chemosensory defect in MS and to determine the locus of the lesion.

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Patterns of 2-Deoxyglucose Uptake in Snail Tentacle and CNS during Olfactory Stimulation. RONALD CHASE and IAN CHIN-YEE (Dept. of Biology, McGill University).

The snail's peripheral olfactory apparatus is served by a population of about 100,000 neurons in each tentacle. In order to understand the functional organization of this system, patterns of metabolic activity were examined with an autoradiographic method using [14 C]2-deoxyglucose. Isolated tentacles, or tentacle plus cerebral ganglion preparations, were incubated with 2-DG for periods of 1-3 hrs. Odor stimuli (typically amyl acetate or octanol at -1.0 log saturation) were delivered by means of a dilution olfactometer. Surface conditions on the epithelium were monitored by microscopic observation; the epithelium was kept moist by microperfusion. Following incubation, the tissue was frozen, freeze-substituted in acetone, and processed according to the procedures of Sejnowski *et al.* (1980) and Lancet *et al.* (1982).

The autoradiographs provide resolution of labelled cells approximately 10 μ m in diameter. In the tentacle, label is dense and extensive over neuropile, even in preparations not intentionally stimulated. The consequences of wind or odor stimulation are subtle and require further investigation. Under all sensory conditions, local regions of contrasting high activity are frequently seen, both in the neuropile and in lobules containing sensory neuron parikarya. Cerebral ganglia were examined as bilateral pairs, after only one of the two tentacles was stimulated with odor. The cerebral ganglion ipsilateral to stimulation consistently showed the greatest density of label. Cell bodies and neuropile were both labelled, sometimes with pronounced local variations.

Since the snail olfactory epithelium constitutes a flat surface, has a simple geometric shape and is completely accessible for controlled sensory stimulation, these preliminary results suggest that the snail preparation is suitable for testing the hypothesis of spatial coding in the olfactory system.

Specific Influence of Olfactory Bulb on Olfactory Marker Protein Synthesis in Olfactory Receptor Cells.
M.I. CHUAH and A.I. FARBMAN (Northwestern University).

Previous studies have shown that specific immunohistochemical staining for the olfactory marker protein (OMP) can be demonstrated in organ cultures of olfactory mucosa from E15 rat embryos 6 days after explantation. Explants were cultured on collagen coated Millipore filters supported by a stainless steel grid platform in an atmosphere of 5% CO₂ at 35°C. The culture medium was Waymouth's medium 752/1 supplemented with 0.30 mg/ml L-ascorbic acid and 0.10 mg/ml Gentamicin. Eight groups of explants were cultured: olfactory mucosa (OM) and presumptive olfactory bulb (POB) explanted *en bloc*; OM and POB separated and recombined; OM alone; OM and heart; OM and cerebellum; OM and cerebellum; OM and cervical spinal cord; OM and 3 pieces of POB separated by a 25 μ m thick Millipore filter of 0.45 μ m pore size. Seven days after explantation, cultures were pooled, homogenized and the amounts of OMP determined by radioimmunoassay (RIA). The first two groups of explants consistently showed about twice as much OMP as the rest of the groups. In the last group of cultures, where the mucosa was separated from the POB by a Millipore filter, no enhancement of OMP synthesis was observed. This suggests that the POB probably increases synthesis of OMP by a mechanism that requires direct contact between the receptor neurons and POB. Explants of olfactory mucosa alone and those cultured with POB *en bloc* were also processed for light microscopy and serially sectioned at 7 μ m. The peroxidase-antiperoxidase immunohistochemical technique was applied and alternate sections examined. Receptor neurons demonstrating OMP immunoreactivity in both the perikarya and dendrites were counted. The cell counts showed that the enhanced OMP synthesis shown in RIA of explants of mucosa and bulb *en bloc* was due to about twice as many receptor neurons synthesizing this protein.

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Odor Responses of Hamster Olfactory Bulb Units Following Recovery From Olfactory Nerve Transection.
RICHARD M. COSTANZO (Medical College of Virginia, Richmond, VA 23298).

Following damage to mature olfactory receptor cells, there is a neurogenesis and replacement of neurons in the olfactory epithelium and these replacement neurons grow axons that can establish morphological connections with cells in the olfactory bulb. To determine whether or not these newly formed connections are functional, single unit bulb recordings were made following recovery from transection of the olfactory nerves. A total of 23 units from hamsters with recovery times of 4, 35, 60, 90, 120, 180, and 270 days were recorded. At day 4, although there was spontaneous activity recorded from the bulb, units did not respond to stimulation of the olfactory epithelium with odors. At longer recovery times, odor driven responses were obtained for 15 units. Most units responded to Amyl Acetate, and in several cases an increase in response with increasing concentration was recorded. Response to other odors (1-Butanol, Ethyl Acetate, and Ethyl Butyrate) was variable. Based on recording depth and histological verification of nerve transection, the location of units was well above a teflon shaving used to mark the lowest extent of the transection. Nerve connections observed above this point were from newly reconnected receptors and presumably provided the pathway for stimulus information to reach bulb units.

These data indicate that, following injury, olfactory neurons are capable of reestablishing connections with the olfactory bulb, and furthermore, that these connections make functional synapses that transmit sensory information to second order cells. These findings in the olfactory system suggest that regeneration and repair of neural pathways may be possible in other parts of the nervous system.

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Vomeranasa Organ Removal Eliminates Male Hamster Copulatory Responses to a Macromolecular Fraction of Vaginal Discharge.
A. N. CLANCY, F. MACRIDES, A. G. SINGER and W. C. AGOSTA (Worcester Foundation for Experimental Biology, Shrewsbury, MA 01545 and The Rockefeller University, New York, NY 10020)

The importance of the accessory olfactory system for male copulatory responses to an active, macromolecular fraction of hamster vaginal discharge was determined following bilateral removal of the vomeronasal organ (VOX). A surgical approach through the palate minimized damage to the main olfactory system. Completeness of vomeronasal deafferentation and sparing of the main olfactory afferents were verified using nasal lavage with HRP after the behavioral testing. The macromolecular fraction (15,000 to 40,000 mol. wt.) was obtained by gel permeation chromatography followed by filtration through XAD resin. We examined the behavior of VOX and sham-operated males toward female surrogates (anesthetized males) scented with the fraction, unfractionated discharge, and a solvent control solution. The shams exhibited intense investigation and repeated copulatory attempts toward both the fraction- and discharge-scented surrogates. The VOX males displayed somewhat reduced investigation but significant copulatory behavior toward the discharge-scented surrogates; in contrast, their behavior toward the fraction-scented surrogates was not significantly different from that toward solvent-scented surrogates. Because the VOX males did respond to the unfractionated discharge, their failure to respond to the macromolecular fraction cannot be attributed to general effects of the surgery. Thus, the accessory olfactory system clearly is involved in the copulatory responses to the active material in this fraction. We cannot conclude that other constituents of the discharge elicit copulatory responses via the main olfactory system or that the active material in the fraction cannot also act via this system, i.e., there likely is loss during fractionation and only the unfractionated discharge may have contained the active material in sufficient quantity to have reached the main olfactory system without a specialized pumping mechanism like that of the vomeronasal organ.

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Direct Scaling of the Intensity of Basic Tastes: A Life Span Study.
BEVERLY J. COWART (Clark University)*

Several recent studies have employed ratio-scaling procedures to assess age-related changes in suprathreshold taste sensitivity. None, however, has included a comparison task to provide a measure of possible changes in nonsensory performance factors. Moreover, all have relied solely on the slopes of group psychophysical functions to describe scaling responses. This strategy provides no clear basis for statistical comparisons among groups and delimits the information about sensory functioning that is obtained.

In the present investigation, estimates of the intensity of solutions representing each of the four basic taste qualities were obtained from 72 individuals ranging in age from 4 to 93 years. A cross-modal matching, ratio-scaling procedure was employed. All subjects first used this technique to scale the heaviness of a series of weights.

Statistical evaluations of intensity scaling responses addressed two distinct aspects of suprathreshold sensitivity. First, ratings were analyzed to determine how well subjects were discriminating among stimuli. Specifically, were repeated ratings of stimuli within each series consistent and distinctive, and did mean responses increase with increases in physical stimulation? Second, the median responses of age groups were compared to determine if there were significant differences in the absolute magnitude of intensity estimates assigned to any stimulus.

The age groups did not differ significantly on any of these measures when scaling the heaviness of weights. Analyses of responses to the taste tasks indicate that young children may be poor discriminators of taste stimuli. Significant declines in both discriminative ability and sensation magnitude may occur with aging. However, the nature of the decrement and the point in the life span when decrements are first observed vary with taste quality. A few older individuals appear to suffer marked, quality-specific losses in discriminative ability.

*This research was conducted while the author served at the National Institute of Dental Research.

A comparison of olfactory sensitivity with odor pleasantness perception in a large sample of a general population. RICHARD G. DAVIS (VAMC)

Ratings of odor pleasantness based on microencapsulated odor delivery systems yield very reliable characterizations of odor perception by individual persons. These measurement systems can be inexpensive and administered quickly. However, altered perception of odor pleasantness is not necessarily related to olfactory acuity or sensitivity. Persons (N=453) recruited from passersby at a state fair were given a test of odor sensitivity (four alternatives, forced choice, binary scale, single series of ascending intensities to a criterion of two consecutive correct responses) and an odor pleasantness scale (20 odors, 4 judgements per odor). The results indicate that the sensitivity of an individual to cyclohexanone is not related to the ability of that person to provide consensually valid judgements of pleasantness. Specifically, people distribute themselves on pleasantness perception in generally the same way, independent of their absolute sensitivity to cyclohexanone. Thus, it appears that pleasantness perception is a robust property of olfaction that is largely immune to effects of sensory deficits such as anosmia.

Evidence for a Na Current in Taste: A Multidisciplinary Approach. J.A. DESIMONE (Medical College of VA, Richmond, VA. 23298), S.S. SCHIFFMAN (Duke University, Durham, N.C. 27706), F.W. MAES (Duke University), G.L. HECK (MCV), S.K. DESIMONE (MCV), SHEELA MIERSON, (MCV).

In vitro preparations of the dorsal lingual epithelium of dog and rat give evidence of active ion transport across the epithelium. When the mucosal concentration of NaCl is increased beyond adaptation levels a two-component increase in transepithelial potential difference is observed. This potential and the associated short-circuit current can be largely eliminated by ouabain. This fact and the indifference of the resistance to ouabain indicates that the current is mainly transcellular. However a paracellular pathway having different cation specificity can be demonstrated. Amiloride (0.1 mM) added to the mucosal medium reduces the inward current due to NaCl and to various sugars in a low Na milieu. These results indicate the presence of specific Na-pathways accessible from the mucosal side. This concept is further supported by whole nerve and single fiber studies in the rat which show that amiloride diminishes the NaCl response relative to KCl. Human psychophysical studies show that the tastes of Na and Li salts and various sweet-tasting compounds are diminished by amiloride. K, Ca, sour, bitter and non-sweet amino acids are unaffected. The effects are selective and reversible. The overall results suggest that an amiloride-sensitive Na-flux may be important in NaCl and sweet taste.

Odor Quality Coding in a Simple, Glomerular-Type Brain. CHARLES D. DERBY and BARRY W. ACHE (C.V. Whitney Laboratory, University of Florida)

To understand odor quality coding, it is necessary to describe patterns of convergence of chemoreceptor cells onto secondary neurons. We are using a simple preparation to decipher coding mechanisms at two contiguous neuronal levels: olfactory receptor cells and low-order interneurons. This preparation, the brain and olfactory organs of the spiny lobster, has many parallels with the vertebrate olfactory pathway; e.g. a large number of primary receptor cells with peripheral somata, and receptor axons that synapse onto interneurons in a glomerular-type neuropile. Yet its relative simplicity makes the problem of understanding coding mechanisms more tractable. We are using a defined mixture and its components to describe the specificity (breadth of responsiveness) and across-neuron response patterns of receptor cells and low-order interneurons. Most receptor cells responded to only 1 of 8 components tested, and the response to this compound equaled the response to the mixture. Cells with slightly broader response spectra were also found. Overall, receptor cells showed very low breadth of responsiveness and across-neuron correlations. On the other hand, low-order interneurons showed much greater breadth of responsiveness, with most responding to all 8 of the individual components. Although there are some differences in response spectra of interneurons grouped according to receptive field- or modality-sensitivity, the greater homogeneity of these cells relative to receptor cells is reflected in their much higher values for breadth of responsiveness and across-neuron correlations. There is also evidence for interaction (suppression) of components in the mixture at both the receptor and interneuronal levels. These data suggest that extensive convergence of narrowly-tuned peripheral channels onto low-order interneurons occurs at early synaptic levels. Experiments to correlate structural complexity (branching patterns in glomeruli) and response complexity of these interneurons are in progress.

Topographic distribution of taste responsiveness in the hamster medulla. J. DAVID DICKMAN and DAVID V. SMITH (Dept. Psychol., Univ. Wyoming).

Multi-unit activity in many portions of the nucleus tractus solitarius (NTS) in the rat can be elicited by stimulation of both the anterior tongue and posterior oral cavity (Halpern & Nelson, 1965). There is also some evidence for a differential distribution of sensitivities to sucrose and QHCl within the NTS (Halpern, 1967). Chemical stimuli (0.1 M sucrose, 0.03 M NaCl, 0.003 M HCl and 0.001 M QHCl) were delivered with equal velocity to the anterior tongue (anterior field) and posterior oral cavity (posterior field) of the hamster. Multi-unit activity was recorded throughout the taste-responsive portion of the NTS (60 penetrations in 10 hamsters). These responses were integrated and the measure of response magnitude was the area under the integrated response over a 5-sec period. Responses at all locations were expressed relative to the response to 0.03 M NaCl evoked by stimulation of the anterior field. Although the rostral and caudal extremes of the gustatory-responsive area were sensitive only to anterior and posterior field stimulation, respectively, most locations within the nucleus could be driven from both fields. Preliminary analysis of these data suggests that the relative responsiveness of the nucleus to the various stimuli is differentially distributed. Sucrose responsiveness is greatest in the anterior and lateral portions of the nucleus when the anterior field is stimulated. Responsiveness to QHCl appears to be best on the lateral edge of the NTS, whether the response arises from the anterior or posterior fields. Sucrose stimulation of the posterior field evokes activity throughout the NTS (except in the extreme rostral portion), as does HCl stimulation. Several studies have demonstrated that separate peripheral fields in the rat contain taste receptors with different patterns of sensitivity (Frank, 1975; Oakley, 1967). Perhaps the differential distribution of responsiveness in the NTS of the hamster reflects the pattern of peripheral innervation of this medullary nucleus from differentially sensitive receptor populations.

Taste Responses of Parabrachial Units Conditioned with Intravenous LiCl. PATRICIA M. DI LORENZO and JOHN GARCIA (Department of Psychology, University of California at Los Angeles)*

Unit activity in the parabrachial nucleus was recorded in response to "taste aversion" conditioning in paralyzed rats. To identify taste-responsive units, a solution of NaCl (.1M) was bathed over the tongue through a plastic flow chamber and the resultant responses were recorded. These units were then conditioned by pairing a presentation of NaSaccharin (.1%) with an intravenous injection of isotonic LiCl (.15M). After at least 5 minutes, extinction trials of NaSaccharin were presented until the unit response appeared identical to the response before conditioning. The response to NaCl was also recorded before and after the conditioning procedure.

Preliminary analysis of both single and multiunit records suggests that the effects of this conditioning procedure are restricted to the initial phasic component of the excitatory response to NaSaccharin. Most often the initial burst of activity was eliminated by conditioning and gradually returned to a pre-conditioning level over several extinction trials. The tonic excitatory phase of the response was unchanged. Those elements that did not show a prominent phasic excitatory response to NaSaccharin did not appear to alter their NaSaccharin response after conditioning. Conversely, those that showed only a phasic response to NaSaccharin appeared unresponsive after conditioning. These responses gradually recovered over extinction. Units that did not respond at all to NaSaccharin before conditioning were also unresponsive after conditioning. In every case the response to NaCl was unchanged by the conditioning procedure.

These data point to the initial burst of activity in response to NaSaccharin as a possible behavioral correlate of a behavioral and/or hedonic decision. Further studies on the role of the parabrachial nucleus in this process are now in progress.

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Further Development and Application of the University of Pennsylvania Smell Identification Test. RICHARD L. DOTY, PAUL SHAMAN, DONNA DEWEY, RONITA GIBERSON & LENORE SIKORSKI (Smell and Taste Center, University of Pennsylvania, Philadelphia)*

The University of Pennsylvania Smell Identification Test (UPSIT), a 40-item forced-choice test incorporating micro-encapsulated odorants, has now been administered to over 2500 subjects. The results of the administration of this test to specific patient groups known to have disorders of the olfactory system will be presented, along with a discussion of studies associated with its development. For example, multiple regression analyses indicating the relative influences of age, gender, race, and smoking habits on UPSIT scores will be presented, as will data related to the test's reliability and relationships to other commonly-presented tests of olfactory function. This test provides, for the first time, an easy-to-administer and efficient instrument for screening smell function in non-laboratory situations, such as doctor's offices, patient's rooms, and other settings.

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Aversive Responses to Glucose. RICHARD J. DIROCCO (Monell Chemical Senses Center)

Ingestion of a sweet-tasting substance is usually followed by its oxidation and the generation of energy in the form of ATP. By examining the response of rats to a sweet taste before and after its pairing with an energy deficit, we were able to ask whether the normal relationship between taste-elicited responses and metabolism is subject to modification by learning. Chronically-indwelling oral cannulas and vena cava catheters were surgically implanted in rats. On a test day, the cannula and catheter were connected to delivery syringes by silicone tubings and the rat was placed in a clear plexiglass chamber which permitted videotaping of its orofacial responses to orally administered glucose, as well as remote sampling of blood (1). Rats were given 50 µl oral injections of glucose (0.8 M) every 5 seconds for 1 minute. Six minutes after the beginning of oral glucose injections, the glucose analogue 2-deoxy-D-glucose (400 mg/kg) was, injected into the vena cava catheter. 2DG blocks glycolysis thereby inhibiting subsequent oxidation of glucose. Blood samples were withdrawn over the next 3 hours. Plasma glucose determinations revealed the expected compensatory sympathoadrenal hyperglycemic response to 2DG, indicating the challenge was recognized by the CNS. By the third test day clear cut aversive responses to glucose, including gaping and chin-rubbing, were observed. Future studies will deliver 2DG directly to the liver, in order to determine whether similar effects can be observed at lower doses delivered directly to a putative site of metabolic receptors. This will help confirm or reject our working hypothesis that it is a metabolic signal, and not malaise, that is being paired with the sweet taste of glucose to produce the affects we have observed.

1) Grill, H.J. and R. Norgren. Brain Res. Bull. 143: 263-279, 1978.

University of Pennsylvania Clinical Smell and Taste Research Center: Analysis of Patients Admitted from 1980 to 1983. RICHARD L. DOTY, PAUL SHAMAN, R. GREGG SETTLE, VERNON J. BRIGHTMAN, JAMES B. SNOW, JR., RONITA GIBERSON, LENORE SIKORSKI, DONNA DEWEY, AMY SISLEY & IRIT NORDIN (Smell and Taste Center, University of Pennsylvania, Philadelphia)*

Since its founding in 1980, our Clinical Smell and Taste Research Center has evaluated the smell and taste function of over 3000 individuals. In the clinic alone, over 300 patients have been tested. Based upon this latter group, the Center has developed a computerized patient registry containing (i) clinical records (submitted with the permission of the patient), (ii) the information patients provide on standardized intake questionnaires, and (iii) the results of otorhinolaryngologic, oral medicine, and smell and taste sensory evaluations. Included in this data base are detailed demographic data and various types of psychological information, including scores on the Moos Menstrual Distress Questionnaire and the Beck Depression Scale. The tabulation of information from this data set will be presented. Answers to questions such as the following will be addressed: (1) Do more men or women seek help for dysfunctions of the chemical senses? (2) What are the most common etiological factors associated with disorders of the chemical senses? (3) Do persons with olfactory problems differ from normal persons in regards to basic endocrine function (e.g., menstrual cycle length)? (4) What psychological manifestations are present in persons with smell and/or taste loss? (5) At what ages are specific types of smell disorders most likely to occur? (6) How common is the belief that taste dysfunction is present in persons suffering olfactory disorders? Answers to these and other questions emphasize the fact that disorders of the chemical senses are clearly of major significance to persons experiencing them.

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Olfaction and Taste: Independent Yet Interacting.

MELVIN P. ENNS and DAVID E. HORNING (St. Lawrence University, Canton, New York 13617)*

To study the influences of tastants on the scaling of odorants, and vice versa, we developed a device that permits the simultaneous and independent varying of the gustatory and olfactory components of a flavor compound. This device uses two glass scintillation bottles, the cap of one which is glued to the bottom of the other. A glass straw is inserted through a hole in the side of the lower bottle and extends so that the contents of the bottom bottle can be tasted through the straw while the contents of the top bottle are being smelled. A polyurethane sleeve conceals and insulates the contents of both bottles. Thus, the concentrations of the stimuli delivered to the nose can be varied independently from the concentration delivered to the tongue.

The first experiment compared the results obtained using our device to those obtained using a more traditional psychophysical delivery system. With instant coffee and the method of magnitude estimation, the slopes of the olfactory, taste, and flavor curves produced by our device were similar to the slopes produced by an open-cup delivery system. To then study the interaction of taste on smell and vice versa, subjects were asked to scale five concentrations of olfactory stimuli against the background of each of three concentrations of tastants, to scale five tastants against the background of each of three concentrations of odorants, and finally to scale overall intensity (flavor) when presented with all possible combinations of four concentrations of odorants and tastants. For instant coffee, olfactory backgrounds did not significantly alter the scaling of taste and, similarly, taste backgrounds did not significantly alter the scaling of olfaction. The scaling of flavor was determined by a combination of the taste and smell components. However, changes in the olfactory stimulus did not have as much effect on the perceived magnitude of flavor as did changes in the gustatory stimulus.

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Fine Structure of Taste Buds in Monkey Foliate Papillae.

A. I. FARBMAN (Northwestern University, Evanston, IL) and G. HELLEKANT (University of Wisconsin, Madison, WI).

Particular attention was paid in this study to the possible relationship between taste bud cell structure and stage of cell differentiation. The results make it clear that, based on such criteria as cell shape, cell polarity, nuclear morphology and distribution and amount of cytoplasmic organelles, there are at least three distinct cell types. It is also clear that the "dark cell-light cell" nomenclature is not a useful means for distinguishing among cells; within an individual taste bud both dark and light cells may be seen that, based on the above criteria belong to the same cell type. The three cell types are: 1) Type I or secretory cell, characterized by a narrow, very irregular cell body, oval nucleus with patchy heterochromatin, abundant rough endoplasmic reticulum, supranuclear Golgi bodies and dense secretory granules that collect near the apex of the cell. Dark and light versions of this cell type are present within the same taste bud and probably represent different stages of its development; 2) Type II cell, largest (in width) of the cells in the taste bud with a spherical nucleus, many mitochondria, supranuclear and infranuclear Golgi bodies, much smooth endoplasmic reticulum particularly in the apical cytoplasm. This cell type, too, may be light or dark; 3) Type III cell, characterized by an irregularly shaped nucleus containing patchy heterochromatin. The apical and basal regions of the cytoplasm are highly organized and strikingly different from each other. In the apical region there are longitudinally oriented microtubules, mitochondria, smooth endoplasmic reticulum, and few ribosomes. The basal cytoplasm is virtually filled with vesicles of varying size. A characteristic vesicle type is spherical, about 80-120 nm in diameter and contains a single eccentrically placed dense particle. In the perikaryon region a whorled array of smooth endoplasmic reticulum is frequently seen. This cell type forms synaptic connections with intragemmal nerves.

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Similar Neural Responses from Regions Rich in Ciliated and Microvillous Cells, respectively, in the Catfish Olfactory Epithelium. JAY ERICKSON and JOHN CAPRIO (Louisiana State University).

Anatomical evidence has indicated that microvillous cells may be olfactory receptor neurons in fishes. In salmonids it has been proposed that ciliated cells primarily respond to bile salts, while microvillous cells are responsive to amino acids (Thommesen, 1982). The purpose of this study was to compare olfactory neural responses from regions of predominantly microvillous and ciliated cells, respectively, in the channel catfish, *Ictalurus punctatus*.

SEM observations support the findings of Cancalon (1982) that the two cell types are segregated into two portions of the sensory region on the face of each olfactory lamella. Microvillous cells are predominant in the dorso-medial portion of each sensory region, while a greater density of ciliated cells is located ventro-laterally. Metal-filled glass capillary microelectrodes tipped with Pt-black were used to record multiunit responses from these two epithelial regions. 10^{-4} M solutions of five amino acids and three bile salts were used as test stimuli. The responses were integrated and standardized to the L-alanine response. In both regions, L-cysteine, L-alanine and L-norleucine were highly stimulatory, while the sodium salts of cholic acid, taurocholic acid and tauroolithocholic acid (i.e. bile salts) were the least stimulatory compounds tested. No differences were detected in the response specificities of the two lamellar regions for the compounds tested.

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Developmental Time Course of Myelination in Chorda Tympani Nerve of Postnatal Rat. FAY FERRELL, RICHARD CHOLE and TRACY TSUETAKI (Departments of Nutrition and Otorhinolaryngology, University of California, Davis, CA 95616).

Studies of the postnatal rat gustatory system show that neurophysiological changes in monochloride salt responses occur with age. These functional changes might be related in part to developmental changes in properties of the chorda tympani nerve fibers. To determine the time course of myelination of CT fibers over the period during which marked changes occur in neurophysiological response characteristics, we examined the CTs of rats aged 4-30 days, taken at 3-5 day intervals, and of adult rats. Numbers of myelinated fibers were counted on TEM montages of cross-sections of the nerves. For each myelinated fiber, total X-sec area (μm^2), X-sec area of myelin (μm^2), and % of total X-sec area occupied by the myelin sheath [$\text{myelin}(\mu\text{m}^2)/\text{total}(\mu\text{m}^2)$] were calculated using a Zeiss videoplan. Means are reported for four ages below:

	4-Day	21-Day	30-Day	Adult
# Myelinated Axons	39	414	597	763
Total Area (μm^2)	800	5,834	8,264	8,738
Myelin Area (μm^2)	28	770	1,151	2,266
Myelin/Total	0.035	0.132	0.139	0.259

By 30 postnatal days the young rat had 80% of the total number of myelinated CT fibers observed in the adult, but the total cross-sectional area of myelin and the percent it comprised of the total cross-sectional area of the nerve fiber were only slightly over 50% the value found for the adult. These differences may contribute to longer latencies reported for younger rats in single fiber recordings (Hill, Mistretta and Bradley, 1982). Developmental changes in unmyelinated fibers are now being analyzed.

The Effect of Duration of Anosmia on Nutritional Status. A. M. FERRIS (Nutritional Sciences, Univ. of Connecticut, Storrs) and J. L. SCHLITZER, M.J. SCHIEBERL, M. PETERSON & F. CATALANOTTO (Pediatric Dentistry & Connecticut Chemosensory Clinical Research Center, Univ. Connecticut Health Center, Farmington, CT).

One of our early observations at the Taste and Smell clinic was that patients with recently developed anosmia were very upset by their loss of flavor perception while life-long anosmics seemed to enjoy food. We felt that these perceptions could be quantified through an assessment of nutritive status and food behavior. In addition, we wondered if anosmia placed patients at nutritional risk.

Anosmics were divided into three groups: a) lifelong anosmics (n=9) b) anosmics of greater than 5 yrs. but not lifelong (n=22) and c) anosmics of less than 5 yrs. (n=18). Each patient was compared to an age and sex matched control subject. We measured standard anthropometric, biochemical, and nutritional parameters (24 hour recall and 3 day food intake), and developed a measure for food complexity.

Anosmics and controls did not differ in a number of anthropometric measures including body weight or body mass index. Anosmics of more than five years showed significant decreases in serum albumin and hemoglobin but not serum total protein, plasma zinc or RBC zinc. The controls did not have a more complex diet nor did they eat more frequently. All anosmics studied rated their enjoyment of eating significantly lower than the controls. Analysis is now proceeding to examine if either the lesser enjoyment of food or differences noted in biochemical indices are reflected in nutrient intake.

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Comparison of Gustatory and Fin Ray Chemosensory Pathways in the CNS of the Sea Robin, *Prionotus carolinus*. THOMAS E. FINGER (Univ. Colorado School of Medicine and Marine Biological Laboratory).

In addition to the usual gustatory and olfactory chemical senses, sea robins possess spinally-innervated fin rays which are capable of sensing relatively low concentrations of various amino acids (see Silver and Finger, this vol.) The animals use these fin rays to locate food despite the absence of taste buds on the fin rays. Thus it is of interest to compare the organization of the gustatory and fin ray chemosensory systems within the CNS of this species. HRP was used as a neuronal tracer and was injected into the primary gustatory nucleus (facial-vagal lobe), as well as the primary and secondary nuclei mediating the fin ray chemosense. The results show no overlap in the ascending projections of these systems through diencephalic levels. The facial-vagal lobe projects bilaterally to the superior (isthmus) secondary gustatory nucleus and ipsilaterally to a posterior lateral thalamic nucleus. Central afferents to the facial-vagal lobe arise from cells of the posterior lateral thalamic nucleus as well as larger neurons of n. lobobulbaris and the cells surrounding the lateral recess of the third ventricle in the inferior lobes. In contrast, ascending fin ray chemosensory afferents project ipsilaterally to a lateral funicular nucleus, and after synapse there, to the contralateral preglomerular nucleus of the thalamus. Thus the thalamic representations of these two feeding chemical senses are quite distinct. The thalamic gustatory representation is predominantly ipsilateral whereas the thalamic fin ray representation is contralateral. There is, however, a projection from large, deep cells of the facial lobe to the spinal accessory lobes mediating the fin ray chemosense. Thus the gustatory system can provide input to the primary fin ray chemosensory nuclei but not vice versa.

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Sniffing Patterns and Main Olfactory Bulb EEG during Open Field Behaviors in the Rat. WM. FORBES (Worcester Foundation for Experimental Biology, Shrewsbury, MA 01545)

In the open field, rats exhibit a variety of behaviors, many of which have an obvious olfactory component. In the present study, patterns of sniffing, measured with an indwelling nasal thermocouple, and main olfactory bulb EEG, analysed using power spectral procedures, were evaluated during grooming, eating, walking, resting, sniffing an object, and scanning (head extended; fast, regular, moderately deep sniffing). The EEG consistently was found to contain 5 major frequency components with center frequencies as follows. I: 1-10 Hz (at the frequency of sniffing); II: 15-22 Hz; III: 43-53 Hz; IV: 70-90 Hz; and V: 140-175 Hz. Components II and III tended to covary in frequency and amplitude. Both these components were highest in frequency and amplitude during grooming and eating and lowest during resting and object sniffing. Since grooming and eating were characterized by slow, irregular, deep inhalations in proximity to an odor source, object sniffing by very fast, regular, shallow inhalations, and resting by slow, regular, moderately deep inhalations, it was felt that the amplitude and frequency of components II and III were primarily dependent on the magnitude of the sensory volley which accompanied each sniff. Component IV did not exhibit state-dependent variations in frequency but increased in amplitude slightly during resting, when the animal seemed inattentive to sensory input. The frequency of component V varied directly, though somewhat loosely, with the frequency of sniffing. Components III and IV correspond with Adrian's "induced" and "intrinsic" waves, respectively, while components II and V have not been well documented previously. Thus, frequency analysis of main olfactory bulb EEG in the freely behaving rat reveals a greater number of oscillatory phenomena than has been evident through time domain analyses of the EEG in more constrained behavioral paradigms.

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Temporal Relationships among Sniffing, Slow Waves in the Main Olfactory Bulb, and the Limbic θ Rhythm in Freely Behaving Rats. WM. FORBES and F. MACRIDES (Worcester Foundation for Experimental Biology, Shrewsbury, MA, 01545)

During acquisition of an odor discrimination task rats tend to entrain their sniffing to the limbic θ rhythm such that the onsets of sniffs exhibit a preferred latency relationship to the θ pacemaker activity (Macrides, Eichenbaum and Forbes, J. NEUROSCI., 2:1705-1717, 1982). The present study characterized this temporal relationship during open field behavior and compared it with that between sniffing and slow waves in the main olfactory bulb (MOB). Across 1-second analysis epochs, the mean frequency of sniffing was slightly lower than that of the θ rhythm though the two rhythms tended to assume the same frequency within epochs significantly more often than would be expected by chance. The variability of sniffing frequency was consistently greater than the variability of θ frequency, strengthening our impression that the covariance of the two rhythms is attributable to modulation of sniffing behavior rather than of the θ rhythm. Based upon analyses of frequency-dependent changes in the phase relationship of the two rhythms we verified that sniffs tend to occur approximately 145 msec prior to a particular phase of the θ rhythm. Using the same analytic procedure we found that the inhalation-evoked slow wave potential in the MOB lagged inhalation by about 45 msec. The differential of 100 msec between the occurrence of the bulbar evoked potential and the latency at which sniffing and the limbic θ rhythm become entrained is substantially greater than would be expected if the sniff/ θ entrainments merely reflected an inhalation-related limbic evoked response. These results are thus supportive of our previous interpretation that sniffing tends to become entrained to endogenous limbic rhythmic activity. The present results also indicate that the preferred latency relationship between sniffing and the limbic θ rhythm is not restricted to discrimination learning situations but may occur during a variety of behavioral states in the open field.

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Comparative Anatomy of the Olfactory Bulb of the Sooty Shearwater, Northern Fulmar and Rock Dove. FOX, V. SUSAN, DAVIDIAN, HENRY, WENZEL, BERNICE M., AND HUTCHISON, LARRY V. (Dept. of Physiology, UCLA School of Medicine, Los Angeles, CA 90024)*

It is known that, like other procellariiforms, the Sooty Shearwater (*Puffinus griseus*) and Northern Fulmar (*Fulmarus glacialis*) 1) have a specialized nasal morphology that accommodates extensive olfactory mucosae, 2) have olfactory bulbs that are very large compared to those of other avian species, and 3) respond to food-related odors when foraging. Nothing is known, however, about their central neural olfactory tissue nor have quantitative comparisons been made with the single avian species whose brain has been described, the Rock Dove (*Columba livia*). Gross and microscopic examination has been carried out for these three species. In proportion to bulb size, the diameter of the olfactory nerve in the procellariiforms is about double that of the Rock Dove. The bulbs of all three species are similar to those of mammals, in that the same cellular layers are represented. In microscopic anatomy and cellular organization, the procellariiforms differ from the Rock Dove. Mitral cells in the Sooty Shearwater and Northern Fulmar are larger and more numerous, and their morphological distribution suggests greater functional coupling, compared to the Rock Dove. Differences in relative sizes of cerebellum and cerebrum in the three species will be discussed in terms of possible differences in olfactory foraging strategies.

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Altered Neural Taste Responses in the Hamster Following Treatment of the Tongue with Acid. MARION E. FRANK & THOMAS P. HETTINGER (University of Connecticut Health Center, Farmington, CT 06032).

Gustatory stimuli are thought to interact with receptor sites that are relatively specific for a given class of compounds. In addition, taste receptors may be affected differentially by substances which are not specific stimuli, but which in some way can modulate receptor activity. A strongly acidic solution of hydrochloric acid (0.1 M) applied to the tongue of a hamster for 10-30 seconds not only produces an electrophysiological response of its own, but also causes dynamic changes in subsequent stimulations with moderately strong solutions of HCl (0.01 M), NaCl (0.1 M), sucrose (0.3 M) and KCl (0.3 M). The response to sucrose, measured by the integrated activity of the chorda tympani nerve 5 minutes after the acid had been washed off, was reduced to less than 20% of the value before acid treatment, while the response to KCl was increased by about 30%. Lesser effects were seen for HCl and NaCl. Responses tended to return toward normal values within an hour. The large reduction in the sucrose response parallels earlier results obtained with acid-sucrose mixtures, which are less stimulatory than sucrose alone (Hyman, A.M., and Frank, M.E., 1980. J. Gen. Physiol. 76:143-173). In the present case, the acid is seen to be acting on the sucrose receptor cells rather than by "cross-talk" between nerve fibers, since washing the acid from the tongue would preclude its acting as a specific stimulus for acid-sensitive cells. It is hypothesized that acid treatment may result in alteration in the membrane structure or ion balance at the sucrose receptor sites and that these sites can be largely reconstituted to their original integrity within an hour after the acid is removed. Recovery of function seems to be too rapid to be explained by receptor cell turnover. The potentiation of the KCl response suggests that receptor cells sensitive to KCl may be activated by polarization in the opposite direction to that of sucrose-sensitive cells.

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Genetic differences among mice in taste psychophysics of sucrose octaacetate. PATRICK FRYE, DAVID B. HARDER, MICHAEL E. RASHOTTE, JAMES C. SMITH, GLAYDE WHITNEY (Florida State University)*

SWR inbred mice consistently avoid a 10^{-4} M sucrose octaacetate (SOA) solution in unconditioned two-bottle preference tests whereas mice of all other inbred strains yet tested do not. This finding confirms a previous report (Lush, Genet. Res. Camb., 1981, 38, 93-95) that SWR mice avoid SOA at this concentration while other strains are indifferent. The major strain difference between SWR and other inbred mice is robust when tested with other psychophysical procedures although indicated taste thresholds vary with procedure. In a conditioned taste aversion procedure SWR mice avoided SOA at concentrations from 10^{-3} M to 10^{-7} M but not at 10^{-8} M. Various other inbred strains first failed to avoid SOA at concentrations from 10^{-3} M to 10^{-5} M when tested in a descending concentration series with a conditioned taste aversion procedure. In a single-bottle free-licking procedure SWR mice differ from mice of other inbred strains in response to SOA following extremely brief exposure to the SOA. The genetic uniqueness of SWR mice revealed by taste testing with SOA does not generalize to some other (P.T.C., cyclohexamide) bitter tastants.

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Use of Grays as a reference to scale odor, taste and flavor. MARIA ROSA GARCIA-MEDINA and AMALIA MIRTHA CALVINO. (Laboratorio de Investigaciones Sensoriales, CONICET, Facultad de Medicina, UBA, C.C.53, 1453-Buenos Aires, Argentina)

In this work we explored the possibility to use an scale of grays as comparison stimuli against odor, taste and flavor.

The first part explored the function of grayness by three different techniques: magnitude estimation (with two different ranges of reflectance), magnitude production and magnitude matching.

In the second part taste functions of sucrose, NaCl, and acetic acid and odor and flavor functions of acetic acid, were obtained by means of magnitude estimation.

Finally, matching functions between the three mentioned continua and grays were obtained.

Results obtained by numerical estimation and matching with grays show good agreement. The use of grayness as a reference scale may be of interest to measure suprathreshold chemosensory stimuli.

Ionic Correlates of Chemosensory Behavior of the Ciliate Protozoan, Tetrahymena. LAWRENCE GARDNER and M. LEVANDOWSKY. (Haskins Laboratories of Pace University, New York, NY 10038)

Starved *Tetrahymena thermophila* accumulate preferentially in capillaries containing amino acids in the micro- to millimolar range. The response is abolished by non-toxic levels of calcium blockers (EGTA; oxalic acid; barium salts, ruthenium red). Addition of suitable levels of calcium or strontium restores the response. Washing the cells thoroughly in calcium-free medium, however, does not abolish the response. Thus, an intracellular calcium pool may be involved, rather than a calcium-dependent membrane potential. Changes in the external potassium/calcium ratio had no effect on the response. However, it is reversibly abolished by tetraethylammonium (TEA) salts, but not by amantadine, another reputed potassium blocker.

Spatial Properties of Taste Perception. JANNEANE F. GENT (University of Connecticut Health Center)

Small pieces of solution soaked filter paper have proven to be effective means of stimulation for the study of spatial properties of taste perception. Using this technique, Collings (1974) was able to establish thresholds for representative tastants on various locations of the human tongue and the soft palate. She demonstrated that contrary to the frequently referenced data of Hanig (1901), the threshold for bitter is lowest on the front of the tongue and lower still on the soft palate. In addition, she was able to measure the growth of taste sensation with concentration at discrete locations on the tongue and soft palate.

Similarly, the filter paper method of stimulation has been used to study the decay of taste sensation over time, (adaptation) in humans and in the hamster chorda tympani.

More recently we combined the psychophysical method of direct magnitude estimation of intensity which is much easier than the matching technique used by Collings (1974), with the filter paper stimulator to establish intensity, concentration relationships for various compounds on different areas of the tongue. After trying various shapes and sizes of filter paper stimulators, we found that an annular shape about 235 square mm in area gave the best results.

For the first study, psychophysical functions (intensity versus concentration) were found for the front and side edges (left and right) of the tongue for five different compounds (N=20). Not surprisingly, no differences were found between the left and right halves of the tongue (averaged across subjects). Perhaps surprisingly, there were also no differences found between the mean intensity (over all subjects) on the front (averaged over left and right halves) and the side of the tongue for all concentrations of NaCl, sucrose, citric acid, and QHCL. There did appear to be a difference between perceived intensity on the front versus side for the fifth compound tested, the intensely sweet artificial sweetener, neohesperidin dihydrochalcone.

Olfactory Receptor Field Potential Variations During Regrowth. ROBERT C. GESTELAND, DENNIS L. DIRECTOR & GLORIA D. ADAMEK (Northwestern University, Evanston, IL 60201)

During the regrowth period which follows ablation of the olfactory epithelium induced by zinc sulfate, the thickness of the epithelium and the dendritic extension of the receptor neurons progressively increase. During the early period the neurons have short dendrites like the oldest of the continually-turning-over neurons in the normal epithelium. However the boundary conditions imposed on the field potentials due to the differences in thickness are different. We compare here the waveforms of the field potentials of single receptor action potentials recorded at different depths in the normal epithelium, in the epithelium during regrowth following deciliation and in the epithelium during successive stages of regeneration following ablation. The gross differences can be accounted for by changes in epithelial geometry. We expect that different neuron developmental states will have characteristic waveform signatures. The study relies upon digital recording of each action potential and subsequent selection of those waveforms which are not seriously corrupted by noise. A bucket-brigade signal delay device is described which permits the real-time signal to be used as a data-grab flag. The processor digitizes the delayed analog signal. An analog-to-digital converter feeding a dual-ported memory unit permits high sampling rates and therefore faithful waveform recording. The data acquisition program flow chart and the use of a down-line-loaded dedicated microprocessor as a port on a multi-user, multi-tasking laboratory computer are described.

This work was supported by NIH Grants NS-14663 and NS-18490 and NSF Grant BNS-8117075.

Histological and Histochemical Investigations of Secretory Elements in the Olfactory Mucosa of the Adult Salamander. M.L. GETCHELL and T.V. GETCHELL (Department of Anatomy, Wayne State University School of Medicine, Detroit, MI 48201).

The two major secretory elements in the olfactory mucosa (OM) are sustentacular cells (SC) in the olfactory epithelium and multicellular olfactory glands (OG) in the subjacent lamina propria. In contrast to the dorsal OM which contains a single superficial layer of OG, the ventral OM generally contains three stratified layers of OG: superficial (sBG), middle (mBG) and deep (dBG). Based on location, presence of secretory ducts and characteristics of the acinar cells, we have provisionally identified the glands in the dorsal OM and the superficial layer of glands in the ventral OM as the traditionally defined "Bowman's glands." The middle layer of glands is usually separated from the sBG by olfactory nerve bundles. They resemble the sBG in that their cells have similar staining properties and are frequently found in association with blood vessels and melanocytes. They differ in that the secretory granules are more widely distributed within the acinar cells. We have not observed ducts projecting to the epithelial surface. The dBG lay at the base of the lamina propria. They resemble the sBG and the mBG in that the acini are associated with blood vessels but differ in that they are rarely associated with melanocytes. We have not observed ducts associated with the dBG. The acinar cells have somewhat different staining properties with larger secretory granules than the sBG and mBG. Histochemical techniques show that the acinar cells of all OG contain neutral MPS. In contrast, glands in the adjacent respiratory mucosa contain acidic and sulfated MPS. Both major secretory elements contain neutral MPS; in addition, the SC contain acidic and sulfated MPS. Supported by NIH-NS-16340.

Number of neurons in the rat N.T.S. Gill, J.M.II and Erickson, R.P. Department of Psychology, Duke University.

We have previously suggested that neural gustatory information may exist in the absolute differences between the amounts of evoked activity summated across the participating neurons (Gill *et al.*, A.C.S., 1982). This requires the determination of the numbers of neurons involved, and the differences in each produced by the various stimuli.

Deeply-anesthetized female Sprague-Dawley rats were used with chemical stimulation of the whole oral cavity.

As the first step in this process, we have used several techniques to determine the numbers of neurons in the solitary nucleus of the rat responding to taste stimuli: a) counts of the numbers of neurons encountered in passes through the nucleus with a recording electrode; b) counts of the numbers of neurons retrogradely labeled by injections of HRP in the pontine taste area, and; c) the numbers of neurons in NTS labeled by a neuron-specific enzyme NSE (i.e., glia are not labeled).

In the first method, electrode penetrations are made throughout all portions of the nucleus. The seeing-distance of each electrode (the distance over which a neurons could be detected) was determined from the recordings, and the number of neurons in each penetration noted. The limits of the active volume of the medulla were also determined with the recording electrode. Considering together the seeing distance, the number of neurons within each penetration, and the total volume of the nucleus, the total number of responding neurons could be approximated. Our estimates here are on the order of 500 neurons/NTS.

With the HRP method, it is clear that the neurons are not densely packed in the nucleus; this was to be expected from the results of the neural recordings. Our estimates from this method are on the order of 800 neurons/NTS. The results with NSE were slightly higher, suggesting that not all these neurons project to the pons.

The further required data for this approach to measurement of neural information is measurement of the responses of these neurons to various stimuli, sampled uniformly throughout the nucleus. A basic problem is whether more than one taste function is being sampled.

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Mixture Suppression of Primary Chemoreceptor Responses: Neurophysiological Evidence in Taurine Sensitive Cells. R. A. GLEESON (Monell Chemical Senses Center and C. V. Whitney Laboratory, University of Florida) and B. W. ACHE (C. V. Whitney Laboratory).

Under natural conditions, both the olfactory and gustatory systems are exposed to complex mixtures of chemical stimuli. Although a number of psychophysical studies have begun to define the relationships between sensations evoked by pure compounds and mixtures, only a few workers have addressed this question neurophysiologically. In the present study, single unit recordings from chemoreceptors on the antennule of the spiny lobster reveal a subpopulation of taurine sensitive cells whose response to taurine is inhibited by certain amino acids. A synthetic mixture of amino acids, which mimics the composition of a natural food stimulus (crab muscle tissue) and itself contains taurine, totally and reversibly blocks the taurine response of this group of receptor cells. Analysis of the contribution of individual components in the mixture to the inhibition reveals that several compounds exhibit partial or complete inhibitory activity. These results suggest that the processing of chemical information in quality coding of natural stimulus mixtures is tempered by interactions of the components at the receptor level.

A Clinical Trial of Systemic Corticosteroid Therapy in Anosmics. R.B. Goodspeed, J.F. Gent, & F.A. Catalanotto (Connecticut Chemosensory Clinical Research Center, Schools of Medicine & Dental Medicine, University of Connecticut Health Center); W.S. Cain & R.T. Zgraniski (J.B. Pierce Foundation & Yale University)*

Twenty patients with anosmia were studied to document anecdotal reports of return of olfactory function during corticosteroid therapy. Participants demonstrated clinical evidence or a history of nasal and/or paranasal sinus disease (NPSD), or an alleged previous response to corticosteroids. Treatment consisted of 50 mg. of prednisone daily for 7 days. A 50% or greater decrease in the total circulating eosinophil count (TCEC) was considered to be indicative of a systemic corticosteroid effect. Olfactory function was measured prior to steroid therapy, twice on the first day and on two other occasions during therapy using the standard butanol threshold and odor identification tasks from the Taste and Smell Center. A combined score on the olfactory tests of at least 60% was pre-defined as a positive response to therapy; less than 60% was considered a non-response. There were 6 responders and 14 non-responders. All patients displayed evidence of a systemic corticosteroid effect but there was no difference between the two groups in the mean TCEC prior to or during therapy. There was a significant association between the presence of NPSD and a positive response; all 6 of the responders and 4 of the non-responders had NPSD associated with their smell deficits. Of the remaining 10 non-responders, 4 had smell deficits associated with a viral-like illness and 6 had no clear etiology. Although long-term, high dose, systemic corticosteroid therapy is not justified for the treatment of anosmia, studying possible mechanisms of its effectiveness is a step toward identifying safer therapies.

*Funded by NS 16993-02

Taste Responses To Amino Acids And Quinine HCl In The Southern Leopard Frog, *Rana sphenocephala*. KENNETH D. GORDON and JOHN CAPRIO (Louisiana State University).

Until recently (Yoshii *et al.* 1981,1982), no work had been reported on amino acid (a.a.) taste sensitivity in anurans, although amino acids had been shown to be effective taste stimuli in fishes. Thresholds determined in this study indicate a greater similarity in a.a. taste between *Rana sphenocephala* and mammalian taste (Tateda, 1967) than for fish. Integrated (time constant 0.5 sec.) responses of the glossopharyngeal (IX) nerve were recorded to characterize dose-response curves for selected amino acids and quinine HCl. Teased bundles of the IX nerve were tested for taste sensitivity with search solutions of arginine at 5×10^{-2} M, an amino acid mixture of three known stimulatory amino acids each at 1×10^{-2} M, and quinine HCl at 1×10^{-3} M. Nerve bundles responsive to amino acids were further characterized by additional a.a. stimuli. All stimuli were dissolved in Ringer's and adjusted to pH 8.3. The lowest threshold obtained was to quinine (in the micromolar range), while thresholds for amino acids were almost 1000 times higher. Quinine, lysine and arginine produced primarily a phasic taste response while the glutamic acid response was tonic. Considerable variability of amino acid taste responses existed between teased bundles of IX, but in all multiunit preparations quinine elicited a greater response magnitude than any a.a. tested at equimolar concentration.

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Neurophysiological and Morphological Classes of Pheromone Sensitive Sensilla on the Antenna of *Trichoplusia ni* (Lepidoptera: Noctuidae). A. GRANT, R. MANKIN, and M. MAYER (USDA, ARS, Insect Attractant, Behavior and Biology Research Laboratory, P.O. Box 14565, Gainesville, FL 32604), and R. O'CONNELL (Worcester Foundation for Experimental Biology, 222 Maple Ave., Shrewsbury, MA 01545)*

Scanning electron microscopy (SEM) and recordings from single antennal sensilla of male *Trichoplusia ni* reveal the existence of at least two classes of pheromone-sensitive sensilla trichodea. The sensilla in both classes are innervated by at least two neurons. The two classes of sensilla exhibit different amounts of spontaneous impulse activity and can be categorized as LS (low spontaneous) and HS (high spontaneous). These classes also differ in their response to stimulation with the pheromone and related compounds. One neuron in the HS class is reliably excited by low doses of (Z)-7-dodecen-1-ol acetate (Z-7,12:Ac) and the other neuron is excited by comparably low doses of (Z)-7-dodecen-1-ol (Z-7,12:OH). One neuron in the LS sensilla is reliably excited by high doses of Z-7,12:Ac but shows no response to Z-7,12:OH, while the other neuron is unresponsive to either Z-7,12:Ac or Z-7,12:OH even when doses are increased 10,000 fold. Furthermore, the HS sensilla are activated by relatively few of the pheromone analogs tested, whereas the LS sensilla respond to an array of such materials. SEM examination of electrophysiologically classified sensilla reveal several morphological differences including: the average length (35.3±4.2 µm for the LS and 28.7±2.6 µm for the HS) and the density of cuticular pores (the HS sensilla have twice as many as the LS sensilla). Brief exposure of the HS sensilla to doses of pheromone in the dose response range of the LS sensilla results in long-term alteration of the neurons' impulse activity and response properties.

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The Effect of Water Deprivation on the Two Bottle Taste Preference Measure. EARL C. HAGSTROM (University of New Hampshire)

The two bottle taste preference method frequently used to evaluate taste sensitivity in animals, compares the amount of water and a taste solution consumed over a 24 hour period. A problem develops when evaluating the effects of a brain lesion or drugs on taste sensitivity if a water deficit is induced inadvertently. Such a deficit could possibly confound the consumption measures. In turn this possibility has led to the assumption that the presence of such a water deficit makes the preference technique less sensitive, because the animal drinks indiscriminately to correct the fluid deficit. However the effect of water deprivation on taste preference has not been studied systematically and the purpose of this study was to put the assumption of decreased sensitivity to an experimental test. Charles River CD rats were tested in their home cage. Each rat had continuous access to the two bottles on its cage, one of which was water, and the other a given concentration of the taste solution under study. Position of the water and taste solution were counterbalanced within subject. Reagent quality chemicals were used to make solutions of Quinine Hydrochloride, Sodium Chloride and Sodium Saccharine. For each concentration, the animal was tested twice under ad lib conditions, in which food and water were available for 24 hours preceding testing, and tested twice under water deprivation conditions, in which only food was available during preceding 24 hours. The same procedure was repeated for water deprivation of 48 and 72 hours. At the higher levels of water deprivation, the two bottle preference test records a progressive loss of taste reactivity.

Ultrastructural Changes in the External Plexiform Layer Following the Loss of Mitral Cells. CHARLES A. GREER (Yale Univ. Sch. Med.), NORBERT HALASZ (Hungarian Acad. Sci.) and GORDON M. SHEPHERD (Yale Univ. Sch. Med.).

The neurological mutant mouse Purkinje Cell Degeneration (PCD) loses the majority of its olfactory bulb (OB) mitral cells (MCs) by 4 mon. postnatal. We have examined the effect of this selective neuronal loss upon the ultrastructure of the external plexiform layer (EPL) in PCD mice.

Affected, homozygous recessive, PCD mice and their normal heterozygous littermates were studied. The mice were perfused with phosphate buffered 1% paraformaldehyde and 2.5% glutaraldehyde and the OBs processed for conventional electronmicroscopy.

Within the EPL of affected PCD mice there was a significant reduction in the total area occupied by the large pale dendrites characteristic of MCs. The largest reduction, 63%, was found in the deep EPL. A smaller reduction in the total area occupied, 35%, was found in the superficial EPL.

The frequency of mitral/tufted synapses onto granule gemmules and granule synapses onto mitral/tufted dendrites per unit area of tissue did not differ significantly between control and affected PCD mice. However, these synaptic types showed a significant increase in frequency when plotted as a function of unit area of mitral/tufted dendrites.

The decrease in the total area occupied by large pale dendrites is consistent with their identification as MC dendrites and the prior report on MC loss in PCD mice (Greer et al., Brain Res. 235: 145, 1982). The normal distribution of granule gemmules supports the hypothesis that granule cells are not directly affected by this genetic mutation. Finally, the alterations in the frequencies of synaptic types suggests that a reorganization of synaptic contacts occurs within the EPL following the loss of MC dendrites.

Supported in part by NS 16993

Narrowly Tuned Amino Acid Receptors: Their Role in Lobster Feeding Behavior. LINDA HANDRICH, PAOLA FERME, and JELLE ATEMA (Boston University Marine Program).

In the lobster, *Homarus americanus*, the first two pairs of walking legs are essential for feeding behavior. These taste organs contain several different populations of chemoreceptor cells, including at least 6 types of "specialist" cells that respond maximally and often with extreme selectivity to only one compound. Their behavioral function is not known.

To test their possible role in feeding behavior, lobsters were suspended individually in flowing seawater in 10 l. aquaria and presented with stimulus-spiked agar cubes placed between their walking legs. Lobsters showed the same dosage-dependent response to a six log step serial dilution of homogenized mussel (*Mytilus edulis*), to a purified mussel extract, and to "synthetic mussel", (a mixture of 21 compounds corresponding to their natural concentrations in mussel flesh). An equimolar mixture of the 6 compounds for which specialist chemoreceptors have been found elicited less than half the response of synthetic mussel. The synthetic mixture without the 6 special compounds gave about one quarter the response of artificial mussel except at the highest concentration (a 1:10 dilution of stock solution), when the response reached normal levels. Two special compounds tested individually, betaine and glutamate, gave responses only one-fourth as strong as artificial mussel. These results are in marked contrast to physiological responses of lobster chemoreceptors.

The 6 special compounds are found in the lobster's natural food in are used in prey detection. It appears that stimulation of the 6 specialist cell types is necessary, but alone is not sufficient, to elicit a feeding response from the lobster's walking legs.

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Transplantation and Survival of the Olfactory Neuroepithelium in Oculo. HECKROTH, J.A., GRAZIADEI, P.P.C. (Florida State University)

The reactions of the olfactory neurogenetic matrix to an environment removed from the nasal cavity and olfactory bulb have been studied through transplantation of the olfactory epithelium to the anterior chamber of the eye. Donor tissue from neonatal rats (P5-8) was implanted in the eyes of adult rats. Hosts were sacrificed after survival times of 3 days to 3 months and the transplants observed with light and electron microscopy. Epithelial fragments tended to form cystic or vesicular structures lined by sensory and respiratory epithelia. Within the olfactory epithelium the rapid degeneration of mature neurons and the proliferation of basal neuroblasts to form a new receptor population followed a time course comparable to that seen *in vivo* after axotomy. Longer survivals showed the presence of mature neural elements, as defined by morphological, ultrastructural and immunocytochemical criteria, within the epithelium, as well as fascicles of small, unmyelinated axons in the lamina propria. The neuroepithelium was seen to acquire a significant degree of disorganization. Capillary loops and papillae of the lamina propria were seen invaginated into the epithelium and cords of young receptor cells were present below the basal lamina. The olfactory epithelium has been seen to continue to manufacture mature neural elements in the absence of its normal target and environment. Possible connections of the sensory axons with the host tissue and with adjacent brain transplants, the rate of neurogenesis, and the influence of the normal innervation of the iris on the neuroepithelium are presently under study.

(Supported by NIH grant #NS16421)

Muscarinic Cholinergic Receptors are Present on Olfactory Receptor Cells in the Salamander Olfactory Epithelium. BRITTA HEDLUND, LEONA M. MASUKAWA and GORDON M. SHEPHERD (Section of Neuroanatomy, Yale University School of Medicine, New Haven, CT 06510)

Very little is known about primary molecular mechanisms taking place in olfactory receptor cells. The presence of muscarinic cholinergic receptors on olfactory receptor cells in mouse olfactory epithelium was suggested by Hirsch and Margolis (*in Biochemistry of Taste and Olfaction*, Acad. Press, 1981). We therefore wanted to investigate the possible modulatory role of muscarinic receptors in primary olfactory mechanisms as part of a study correlating biochemistry and electrophysiology of salamander olfactory receptor cells. Muscarinic cholinergic receptors are present in the olfactory epithelium of the salamander, *Ambystoma Tigrinum*, to an amount of 0.04 pmoles/mg protein, measured via binding of the muscarinic antagonist ³H-3-quinuclidinyl benzilate (³H-3-QNB). Seven days after transection of the olfactory nerve, muscarinic receptors are no longer present in the epithelium indicating that they are located on the olfactory receptor cells which are known to undergo degeneration after nerve transection. Separation of the cilia represented a fivefold enrichment in muscarinic receptors, to 0.25 pmoles ³H-QNB specifically bound per mg protein. It is suggested that acetylcholine may act as a modulator of the olfactory receptor cell response.

Sweetness-Inducing Effect of Miraculin in the Rhesus Monkey G. Hellekant (Department of Veterinary Science, University of Wisconsin)

The gustatory effects of miraculin, the sweetness-inducing protein from the miracle fruit *Synsepalum dulcificum*, was studied in the rhesus monkey, *Macaca mulatta*. The intake of five acids was recorded in two bottle preference tests, one bottle containing acid and the other tap water, before and after miraculin treatment. All the acids tasted more pleasant after miraculin. The electrical activity of the chorda tympani nerve to stimulation of the tongue with a variety of sweeteners, acids, sodium chloride and quinine hydrochloride was recorded in anaesthetized animals. Pretreatment of the tongue with 0.3-5 mg miraculin doubled the summated nerve response to the acids and diminished the response to sucrose by about 10%. The enhancement lasted for at least an hour and the diminution up to 20 min. A solution of 0.1 mg miraculin per ml elicited a weak nerve response. No preference over water for this concentration of miraculin was recorded in the two bottle tests. The activity of 29 single taste fibres, selected for their responsiveness to sweetness or acids or both, was recorded after miraculin treatment. Effects were obtained in 9 fibres which were similar but more pronounced than those observed in the summated recordings. Before miraculin, these fibres responded better and to a larger variety of sweeteners (81%) than the other fibres (40%). After miraculin, acids elicited on the average 2.3 times more activity than before, while the response to sweeteners was depressed. In 20 fibres no effect of miraculin was observed. These fibres responded to fewer of the sweeteners and were more stimulated by the non-sweet stimuli than the first group. The results suggest that miraculin acts on those structures in the taste cell membrane that are involved in perception of the sweet taste, making them sensitive to acids. The new quality of sweetness after miraculin treatment is signalled by taste fibres which normally respond to sweet substances but which, under the influence of miraculin, are responding to acids. It is likely that the quality of a taste stimulus is conveyed by the identity of the taste fibres.

Hepato-Gustatory Interactions in the Brainstem: Evidence for Overlapping Second-Order Projections in the Parabrachial and Parvocellular Reticular Nuclei. GERLINDA HERMANN, NICHOLAS J. KOHLERMAN and RICHARD C. ROGERS (Northwestern University School of Medicine, Department of Physiology)

Recent behavioral investigations by Grill and Norgren have clearly indicated that the brainstem is competent to regulate ingestion on the basis of taste cues and that this process may be modulated by the nutritive state of the animal. Other investigators have pointed out that visceral afferent input via the hepatic vagus nerve may be a very important signal for the alteration of taste preferences and ingestive behavior. Given these data and those concerning the organization of special and general visceral afferent input to the brainstem, it seems likely that hepatic afferent input upon low-order gustatory afferent neurons may be responsible for these changes in taste preference and ingestive choice behavior.

In order to attack problems of visceral chemoreceptor modulation of taste-directed behaviors, it was first necessary to establish a) the location of hepatic afferent terminations within the brain, b) whether hepatic input affects identified gustatory neurons in the solitary nucleus and c) if the projections from hepatic and gustatory neurons of the solitary nucleus overlap extensively in the brainstem. We have answered these preliminary questions using a combination of electrophysiological and neuroanatomical methods. First, the hepatic afferents travel entirely in the left cervical vagus and terminate within the left medial solitary nucleus. Second, we have found no evidence for hepatic effects upon the activity of identified gustatory neurons in the anterior solitary nucleus. Third, we have conclusive anatomical evidence for extensive interactions between gustatory and hepatic/visceral vagal projections from the solitary nucleus upon the subjacent parvocellular reticular nucleus and the postero-medial division of the parabrachial nucleus.

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Parabrachial Nucleus Taste Responses in Prepubertal and Adult Rats. DAVID L. HILL (Ctr. Human Growth & Dev., and Dept. Oral Bio./Sch. Dent., Univ. Mich., Ann Arbor, MI 48109)

To determine whether there are functional changes in responses from third-order gustatory neurons after weaning in rats, recordings were made from chemosensitive neurons in the pontine parabrachial nucleus (PBN). Twenty-seven single neurons were studied in rats aged 25-35 days (prepubertal) and 33 neurons in adults. Chemical stimuli applied to the anterior tongue were 0.1M and 0.5M NH_4Cl , NaCl, LiCl, and KCl, 0.1M citric acid, 0.01N HCl, 1.0M sucrose, 0.1M Na-saccharin and 0.01M quinine HCl. Neural activity was measured for the first 5 sec after stimulation of the tongue; a comparable period of prestimulus spontaneous activity was subtracted to yield response frequencies.

Changes in PBN taste response frequencies were apparent between prepubertal and adult rats. Response frequencies to NaCl, LiCl, sucrose, and Na-saccharin increased 50% or more during postweaning development ($p < 0.10$). In contrast, PBN responses to all other stimuli were similar between prepubertal and adult rats ($p > 0.20$). Further differences were apparent when neurons were categorized on the basis of responding maximally to 0.1M NH_4Cl , to 0.1M NaCl or LiCl, or equally to all 0.1M salts; one neuron in each age group responded maximally to 0.1M KCl. Most PBN neurons in adult and prepubertal groups (64% and 89%, respectively) responded similarly to NH_4Cl , NaCl and LiCl, relatively more PBN neurons in adult rats responded maximally to NaCl or LiCl (33% vs. 7%), and a similar proportion responded maximally to NH_4Cl (3% vs. 4%).

Similar to the second-order gustatory neurons in the NTS, striking postweaning changes are found in responses to NaCl, LiCl, sucrose and saccharin. Differences between NTS and PBN neuron responses include frequency changes between age groups to KCl in the NTS and a larger proportion of NTS neurons maximally responsive to NH_4Cl for both age groups. Since responses from NTS and PBN neurons alter during development, adult rats may also differ from younger rats in behavioral responses to some taste stimuli.

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Clinical Evaluation of Taste and Olfaction in Depressed Patients. ANDREA L. JACOBS (University of Michigan), NAOMI E. LOHR, PH.D. (University of Michigan), DOUG KING, M.D. (University of Michigan).

The questionnaire finding that reported taste loss is a highly sensitive and specific clinical marker for melancholia in being further evaluated by means of a battery of psychophysical tests.

Preliminary data and case illustration from the first four months of laboratory evaluation of taste and olfaction function in depressed patients are presented. Even severely ill depressives have been able to co-operate with the tasks. Olfaction is assessed both by means of the University of Pennsylvania Smell Identification Test and an odor identification test developed at the John B. Pierce Foundation Laboratory. Results from the two methods appear comparable. Taste function is assessed both in terms of threshold sensitivity and suprathreshold discriminations. Thresholds are established for NaCl by means of the Staircase-Forced Choice Method (Bartoshuk, 1978) which requires patients to make a series of judgments between pairs of stimuli, each pair consisting of a cup of NaCl concentration and a cup of deionized water. Suprathreshold function is assessed via a Pierce Foundation developed "Magnitude Matching" in which intensities of the four taste qualities are given nonmodulus ratings and, in addition, are matched to an auditory stimulus of varying decibels. Case illustrations are chosen to contrast melancholic with nonmelancholic patients.

PTC Taste Blindness and Caffeine: Further Considerations. JANET E. HOOPER and LINDA M. BARTOSHUK (John B. Pierce Foundation Laboratory).

Taste thresholds are bimodally distributed for PTC, PROP, and chemically related compounds containing the HNCS group. This bimodal distribution has been used to classify people into "tasters" and "nontasters", i.e., those sensitive or insensitive to PTC and/or PROP. A study in our laboratory (Hall, et al. 1975) found an association between PTC and caffeine, a compound which does not contain the HNCS grouping. A later study in our laboratory (reported at AchemS, 1982) failed to confirm the original result. In that experiment we tested a random sample of 50 subjects for caffeine sensitivity. In the present study, we measured caffeine thresholds in subjects sensitive (10) and insensitive (10) to PTC and PROP. In addition, subjects matched intensities of these tastes (as well as NaCl) with intensities of tones, i.e., magnitude matching.

Analyses of the data yielded two major points: 1. Caffeine thresholds were correlated with PROP ($r = .471$, $p < .05$) and PTC ($r = .465$, $p < .05$) thresholds, a finding in accord with our earlier caffeine study (Hall, et al. 1975). PROP and PTC thresholds were highly correlated with each other ($r = .81$, $p < .00001$). One crucial factor in the association of caffeine and PTC seems to be the subject population; a group of subjects selected for taster status is more likely to exhibit a caffeine-PTC or PROP correlation than is a group of randomly selected subjects. Note that correlations found for caffeine and PTC or caffeine and PROP were lower than those found for PTC and PROP, compounds which are clearly structurally related. 2. Tasters matched the lower caffeine concentrations to higher sound intensities than nontasters; the groups did not differ for the higher caffeine concentrations. Thus, dilute caffeine is more bitter to tasters than nontasters. We conclude that knowledge of PTC status in part predicts caffeine's threshold and its suprathreshold perceived intensity. However, the association between caffeine and either PTC or PROP is different from that between PTC and PROP.

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Electron Microscopy Of Olfactory Epithelium From Dysosmic Patients: A Pilot Study. BRUCE W. JAFEK, DAVID T. MORAN, and J. CARTER ROWLEY III (Departments of Anatomy and Otolaryngology, University of Colorado School of Medicine, Denver, CO 80262)

The probable association of ultrastructural changes in the olfactory mucosae of dysosmic patients has been postulated. Confirmation of this hypothesis has awaited the development of a suitable safe biopsy technique and a detailed description of the fine structure of the human olfactory mucosa. We have developed a safe procedure for obtaining small, 1 mm² biopsies from consenting patients under local anesthesia (Lovell, Jafek, Moran and Rowley, 1982, *Arch. Otolaryngol.* 108:247-249). Having completed a detailed study of the normal human olfactory epithelium (Moran, Rowley, Jafek and Lovell, 1982, *J. Neurocytol.* 11:721-746), we set out to investigate the ultrastructure of the olfactory mucosa from patients afflicted with olfactory dysfunction.

In a number of cases, patients with olfactory dysfunction possessed olfactory epithelia that displayed marked ultrastructural alterations. One patient, for example, had suffered post-traumatic anosmia of two years' duration. Electron-microscopic examination of her olfactory epithelium revealed her olfactory receptors were fewer in number and shorter than normal; their olfactory vesicles possessed basal bodies, yet lacked olfactory cilia. The olfactory epithelium was disorganized, many supporting cells displayed extensive vesiculation, and microvillar cells exhibited extensive morphological alteration. In another case, a 30-year-old man who had suffered head trauma and taken an overdose of immunosuppressive drugs presented at the UCHSC otolaryngology clinic for olfactory testing. Diagnosed as anosmic, electron microscopy indicated his olfactory epithelium displayed a number of distinct morphological abnormalities. The epithelium was disorganized, with an increased population-density of superficially-located nuclei. The olfactory receptors themselves, where present, had short, fat dendrites, and were aciliate. In another case, a patient with psychogenic dysosmia, the ultrastructure appeared normal. Since this is a pilot study, detailed investigations of additional cases is essential before definitive statements can be made. If it clear, however, that in several cases of dysosmia, distinct ultrastructural changes are present in the olfactory epithelium. Supported by NIH (IVS 15203).

Response Spectra of the Antennular Chemoreceptors in the American Lobster, *Homarus americanus*. BRUCE R. JOHNSON and JELLE ATEMA (Boston University Marine Program).

We determined the response specificity of the antennular chemoreceptors in the American lobster to a select group of chemical stimuli. Single chemoreceptor units were located electrophysiologically from the lateral flagellum with a search stimulus made up of the following compounds at 10^{-4} M: taurine, OH-proline, glutamate, ammonium chloride, arginine, sucrose, ethanol, alanine, lysine, betaine, aspartate, glycine, leucine, glutamine and proline. Each component of the search stimulus was then tested individually at 10^{-4} M to find which were stimulatory to the chemoreceptor unit. Applied concentrations were diluted by a factor of approximately 0.025 after introduction to the test chamber.

To date, all single units tested were restricted in the number of components of the search stimulus that elicited a response. Most single units responded exclusively to one compound, OH-proline, fewer responded to taurine, and units specific to glutamate and betaine were found infrequently. Some units responded to 2 or 3 compounds but still maximally to one compound such as taurine, OH-proline, ammonium chloride, or proline. Individual compounds were typically more stimulatory to a single unit than the whole search stimulus, suggesting an inhibitory effect of compounds present in the mixture. We are continuing to catalog the specificity of the antennular chemoreceptor units, to obtain population estimates of the different types of units present, and to characterize the inhibitory effect of non-stimulatory compounds. This effect is in marked contrast to behavioral dose-response curves to such compounds and their mixtures.

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Sensory Attributes of Some Stevia Rebaudiana Sweet Principles. S. K. KAMATH, M. L. ANDERSON, N. P. D. NANAYAKKARA, D. D. SOEJARTO, A. D. KINGHORN (College of Associated Health Professions and College of Pharmacy, University of Illinois Health Sciences Center, Chicago) & R. C. GESTELAND (Northwestern University, Evanston, Illinois)

Five Stevia Rebaudiana glycoside constituents: stevioside (STV), steviolbioside (STB), rebaudioside A (RA), rebaudioside B (RB), and rebaudioside C (RC), were evaluated for various sensory attributes on an open-ended magnitude ratio scale by a panel of selected judges. Approximate concentrations of each of the test products equal in sweetness intensity to 0.25M sucrose are: RA, 8.5×10^{-5} M; RB, 1.3×10^{-4} M, STB, 1.6×10^{-4} M; STV, 1.8×10^{-4} M and RC, 5.6×10^{-4} M. At these concentrations RA was noted to be the sweetest and to have low off- and bitter tastes. On increasing the concentration of RA two-fold, the sweetness intensity was more than doubled without significantly decreasing pleasantness or increasing off-, after-, or bitter tastes. RC is the least sweet, bitterest and most unpleasant and has unpleasant off- and after tastes. STV, STB and RB ranked intermediate in their traits. The detection threshold concentration for STV from a triangle forced-choice was found to be about 15 micromoles which matched well with that observed from the concentration-intensity curve.

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Separate Gustatory and General Visceral Sensory pathways in the brains of Ictalurid catfish. JAGMEET S. KANWAL (Dept. Zoology & Physiology, Louisiana St Univ) and THOMAS E. FINGER (Dept. Anatomy, Univ Colorado Med. Ctr.).

Special (gustatory) and general visceral afferents terminate in separate areas of the visceral sensory column in the medulla of goldfish (Morita et al., 1980) and catfish (Finger, unpub. obs.). However, possible differences in the higher order projections of these two zones have not been investigated. In order to trace afferent and efferent connections of these visceral medullary regions, we used horseradish peroxidase (HRP) as a retrograde and anterograde tracer in two species of catfish. Injections were placed in the primary general visceral nucleus (PGVn), the gustatory region of the vagal and facial lobes and into the isthmus and posterior diencephalic visceral centers determined by previous experiments. The results indicate the presence of distinct gustatory and general visceral sensory nuclei in the isthmus region. The facial and vagal (gustatory) lobes project to the superior secondary gustatory nucleus (SSGn). In contrast, the PGVn projects anteriorly to a distinct area lying immediately rostro-lateral to the SSGn. Diencephalic injections of HRP demonstrate that the SSGn and sec. visceral nucleus in turn project to separate regions. Injections placed posteriorly in the ventral diencephalon retrogradely label only the large neurons of the SSGn. More anterolateral diencephalic injections label only small neurons of the sec. visceral nucleus and also anterogradely label fibers in the lateral portion of the inferior lobe and around the lateral recess of the third ventricle. Thus, parallel yet distinct channels exist for processing gustatory and general visceral information within the CNS. Supported by NIH grants NS14819 (to J. Caprio) and NS15258 (to T. Finger).

The Effect of Chronic Formaldehyde Exposure On 2-Deoxyglucose Uptake in the Olfactory Bulbs of Mice. JOHN S. KAUFER, CHARLES A. GREER (Sects. Of Neurosurgery and Neuroanatomy, Yale Univ. School of Medicine).

A variety of data suggests that long-term exposure to either toxic or non-toxic odorants alters the anatomical organization of the olfactory bulb and may impair functional capabilities. We have investigated these possibilities by chronically exposing mice to the odor of formaldehyde and subsequently examining the functional activity in the olfactory bulbs with the 2-deoxyglucose (2DG) technique. C57 BL/6j mice were exposed for 2-4 weeks to the odor of 0.37% formaldehyde. At the end of the chronic exposure period the mice were injected with 2DG and acutely exposed to either formaldehyde, Amylacetate or purified O-air for 1 hr. Conventional processing of the olfactory bulbs for 2DG autoradiography followed. Control mice were treated similarly during chronic exposure to either purified O-air or regular animal colony air. The 2DG autoradiographs from the mice chronically exposed to the odor of formaldehyde showed little evidence of the distinctive foci of high 2DG uptake typically associated with odor exposure. In effect, these autoradiographs appeared homogeneous across the olfactory bulb laminae. The data thus far suggests that the formaldehyde exposure decreases the signal to noise ratio of the afferent information arriving at the glomeruli of the olfactory bulb during acute testing. This observation implies that the coding of olfactory information may be impaired by the chronic exposure to formaldehyde.

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Effects of Stimulus Duration and Concentration on Taste Perception of $MgSO_4$ and HCl. Steven T. Kelling and Bruce P. Halpern, Dept. of Psychology and Section of Neurobiology and Behavior, Cornell Univ., Ithaca, NY 14853.

Perceived intensity and/or quality of taste stimuli, as well as reaction time, can change as a function of stimulus duration, with chemical content or concentration as factors. We studied this with 3.2 mM and 10 mM HCl; 250 mM and 500 mM $MgSO_4$, at durations of 2000, 1000, 100, 80 and 50 msec. Each $MgSO_4$ or HCl solution (Experimental Stimuli) pulse was preceded and followed by H_2O flow. Control stimuli were H_2O pulses at the five durations, preceded and followed by H_2O . For simple reaction time or intensity, equal numbers of one experimental stimulus concentration and the control stimuli were randomly presented. For quality judgments, two experimental stimuli and the control stimuli were given an equal number of times in random order (Kelling & Halpern, *Science*, 1983).

Perceived intensity of HCl at all durations and $MgSO_4 < 1000$ msec varied directly with duration, $p \leq .05$. Intensity of 1000 msec pulses was $\geq 90\%$ of 2000 msec pulse intensity (the modulus duration), but intensity was 40% to 50% for 100 msec pulses; 30% to 50% for 80 msec; 20% to 35%, for 50 msec.

Effects of duration on quality were less general. For $MgSO_4$, modal quality at ≥ 80 msec was bitter, with sour next, but at 50 msec, modal quality for 500 mM $MgSO_4$ was sweet; for 250 mM, sour. With HCl, taste quality changed little across durations. Salty was modal quality for 10 mM, with sour infrequent except at 50 msec. For 3.2 mM, sour (e.g., sour/sweet, salty/sour/sweet) was modal thru 100 msec; at ≤ 80 msec, salty.

Reaction time (RT) did not vary with duration for $MgSO_4$, while 10 mM HCl had only a marginal increase at 50 msec. However, 3.2 mM HCl had minimum RT, 543 msec, at 100 msec; maximum, 673 msec, at 50 msec.

The HCl concentrations had no differential effect on RT. RT for 250 mM $MgSO_4$ was always > 500 mM; range was 685 vs. 595 at 2000 msec to 717 vs. 556 at 50 msec.

These data confirm the general dependence of intensity upon duration, the relative independence of quality at > 100 msec, and the dependence of RT changes upon the chemical (NSF BNS 8014148).

Reported Taste Loss: A Diagnostic Marker For Melancholia. DOUG KING, M.D. (University of Michigan), ANDREA L. JACOBS (University of Michigan), NAOMI E. LOHR, PH.D. (University of Michigan)

Seventy-six psychiatric patients completed our chemosensory-appetite questionnaire. Patients were diagnosed according to Research Diagnostic Criteria (RDC) and for data analysis were grouped as follows: endogenous depression ($n=44$): major depression (did not meet endogenous criteria) ($n=16$): other ($n=16$). Consistent with our pilot results, reported taste loss occurred primarily in patients with endogenous depression. Reported taste loss appears to be a highly sensitive and specific clinical marker for endogenous depression.

Four items of the (yes/no) portion of the questionnaire dealt with taste: 1) Food tastes as good as usual; 2) Food tastes as good as usual when I eat away from home; 3) I have no trouble distinguishing foods; 4) Lately food tastes like cardboard. "No" responses to both 1 and 2 (i.e. reported taste loss) were obtained in 65.9% of patients with endogenous depression and in 18.8% of all other patients. Item 3 did not distinguish endogenous depressives from nonendogenous depressives and other patients. 38.6% of endogenous depressives reported that food tasted like cardboard whereas only 9% of all other patients (3/32) reported this complaint.

Pheromone Plume Structure and Orientation Response. H. C. von KEYSERLINGK (University of California, Riverside)

The distribution of volatile molecules ($TiCl_4$ smoke) which are carried away from an emission point by moving air were recorded on video tape both in the field and in a wind tunnel. The filamentous fine structure of the smoke plume has been recorded with a photocell. The pheromone of *Grapholita molesta* has been added to the smoke and the flight behavior of the responding males in the pheromone bearing smoke plume has been recorded on videotape. The "searching" behavior of flying moths after removal of the odor is compared with that of walking bark beetles and termites. In the light of these results the current concepts of the orientation of insects to an odor source will be discussed.

Ultrastructure of Taste Cells and Synapses in Mouse Vallate Taste Buds. J. C. KINNAMON, R. DELAY and S. ROPER (University of Colorado Medical Center)

Using transmission electron microscopy (TEM) and high voltage electron microscopy (HVEM) of serial thin and thick sections, we have examined ultrastructural features found in mouse taste buds (TBs). We have also generated 3-D models of synapses and taste bud cells with the aid of a PDP-11 computer.

With TEM and HVEM we have found examples of Type I (dark), Type II (light) and Type IV (basal) cells. Cilia were found associated with Type I cells. The cilia had no preferred orientation, were very short, and did not extend into the taste pore. Hence we doubt that they play a primary role in gustation.

Synaptic contacts were found throughout the bottom two-thirds of the TBs. In all cases the taste cell has been presynaptic. Both light and dark cells (including ciliated dark cells) have been observed to form synapses. The most common type of synapse was characterized by a thickened presynaptic region containing periodic dense projections to which clear vesicles were closely apposed. These "active zones" were often flat in profile. Some, however, were characterized by gaps or breaks and curved profiles. Using computerized 3-D reconstructions we found that a single taste cell may synapse repeatedly onto a single neuronal process and/or other neuronal processes. A second type of synapse has been observed much less frequently, characterized by a bleb-like semicircular profile in which vesicles were clustered at the presynaptic density in higher numbers than were found in the first type of synapse. More significantly, the cytoplasm of the postsynaptic region of the neuronal process was very electron dense.

The biological significance of the existence of two types of synapses in TBs is unknown, but we speculate that differing morphological synaptic types may reflect functional differences. This study was supported in part by grants from NIH and Procter and Gamble.

Further Characterization of Response-Eliciting Components of Earthworm Extract. D.M. KIRSCHENBAUM, L.S. REFORMATO and M. HALPERN. (Downstate Medical Center, 450 Clarkson Avenue, Brooklyn, N.Y. 11203).

We previously reported (Halpern et al., *ACHEMS*, 1982) that garter snakes will respond differentially to earthworm extract (EW) placed in a dish and that this differential response is dependent on a functional vomeronasal system. The active components of EW are water soluble, retain their biological activity following lyophilization, following treatment with strong acid or strong base and following heating for 15 minutes at boiling temperature. Bradford and Lowry protein tests indicate that biological activity and protein content covary. Recent experiments indicate that the response-eliciting components of EW gradually lose activity when subjected to 100°C for 30, 60, 120 minutes and after 240 minutes lose all activity. Ammonium sulfate precipitation studies indicate that the active constituents of EW are precipitated by 90 and 100% ammonium sulfate. Using dialysis membranes with pore sizes (cut-offs) of 12-14,000, 15,000, 25,000 and 50,000 MW samples of EW were dialyzed overnight against dH₂O and tested for activity. Samples dialyzed with 50,000 MW cut-off membrane lost virtually all their activity. Gel filtration, using Sephadex G75 and physiological saline as the eluent, results in two peaks: one in the region of 60,000 MW±10,000 and one in the region of 17,000. These studies suggest that the active ingredient in EW may be a glycoprotein or a small molecule associated with a large protein.

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Maintenance of Chorda Tympani Salt Taste Responses After Nerve Transection in Rats. Y. KITADA* (Dept. Oral Biol., Sch. Dent.), R.M. BRADLEY (Dept. Oral Biol., Sch. Dent., & Dept. Physiol., Sch. Med.), C.M. MISTRETTA (Dept. Oral Biol., Sch. Dent., Ctr. for Human Growth and Dev., & Sch. Nursing, Univ. Michigan, Ann Arbor MI 48109).

To determine whether neurophysiological responses to four salts change differentially or in parallel during long recording sessions after nerve transection, we recorded from the cut chorda tympani nerve in 13 rats while stimulating the anterior tongue with 0.1M NaCl, LiCl, NH₄Cl and KCl. When the nerve was left undisturbed, the taste response remained at or above initial magnitudes for an average of 10 hours, and declined to 50% of initial level by 15 hours. If attempts were made to reposition or redissect the nerve, the responses were maintained for a much longer time. Response magnitudes were at or above initial levels for 13 hours, on average, and the 50% level was not reached until 20 hours. Taste responses were still obtained in some rats at 21 to 27 hours after nerve cut. Responses to the four salts altered in parallel for up to 17 hours after nerve cut. However, in two rats responses to NaCl and LiCl declined disproportionately, relative to NH₄Cl and KCl, after 20 hours. Thus the relative chorda tympani responses to salts are stable over a long period, exceeding the 8-10 hour length of a usual recording session. Since salt taste responses were maintained over longer periods after redissecting the nerve, the dissected portion of the chorda tympani apparently had deteriorated with time, but the more distal portion did not.

*Present address: Dept. Physiol., Okayama Univ., Dent. Sch., Japan. Supported by N.I.H. Grant DE 05728 and N.S.F. Grant BNS 80-15737 to RMB and CMM, and N.I.H., N.I.D.R. Research Career Dev. Award DE 00066 to CMM.

Dissociation of Olfactory Epithelium of Rana pipiens. STEVEN J. KLEENE, ROBERT C. GESTELAND (Northwestern University, Evanston, IL 60201).

We report a method for preparing suspensions of frog olfactory epithelial cells without the use of proteolytic enzymes. The epithelia are first immersed in isotonic Ringer to dissolve mucus. They are then incubated at room temperature and without agitation in a medium which breaks down the epithelial structure. The medium is a Ringer modified by the addition of buffers to maintain alkaline pH and low calcium concentration. Since DNA released from broken cells sometimes makes suspension difficult, the epithelia are given a final incubation in a DNase solution. The tissues are placed in Ringer or amphibian cell culture medium. As the epithelia are peeled away from the cartilage, the epithelial cells begin to go into suspension. Final dispersion is achieved by any of several mechanical means. Olfactory neurons and respiratory epithelial cells in the suspension are easily identified by their vigorously motile cilia; the cell bodies are rounded. Up to 130,000 neurons (either as single cells or in small multiples) are obtainable from one frog. The cells of the suspension are viable as judged by exclusion of trypan blue and ability to synthesize RNA. They can be maintained in culture; the ability to synthesize RNA declines with a half-time of between 1 and 2 days.

This work was supported by NIH Grants NS-06600, NS-18490, and NS-14663 and NSF Grant BNS-8117075.

Na⁺-K⁺ ATPase ACTIVITY RESPONSES OF INSECT ANTENNAL AND COW OLFACTORY TISSUE PREPARATIONS TO INSECT PHEROMONE COMPONENTS. Robert B. Koch, Stanley Myers*, Shahla Shahsavari* and Jeanne Smith*. Dept. of Biochem., MSU, Mississippi State, MS 39762.

Antennae from the Cabbage Looper, *Trichoplusia ni* and pheromone components (Z)-7-dodecen-1-ol acetate (Z7:12AC) and dodecan-1-ol acetate (12 AC) were obtained from Dr. Steven M. Perkovich, ARS-USDA Lab., Gainesville, Fla. Nerve ending particle (NEP) and microsomal fractions were prepared from homogenates for separate male and female antennae samples. Enzyme response studies to the pheromone components were conducted on the four fractions. Comparative experiments using the insect pheromone components were also conducted using cow olfactory NEP and cow brain and mouse brain microsomal fractions. Responses of Na⁺-K⁺ ATPase activity to the pheromone were compared to preparations from higher animals. At 10⁻⁴M pheromones, Na⁺-K⁺ ATPase activity from antennal preparations were inhibited by greater than 50%. At decreasing concentrations of pheromones, the Na⁺-K⁺ ATPase activity from the male moth NEP fraction showed an apparent alternating stimulation-inhibition response with the highest stimulation observed at 10⁻⁹M (equivalent to 10⁻¹² mole per ml). Na⁺-K⁺ ATPase activity from male and female microsomal and female NEP fractions, although inhibited at 10⁻⁴M pheromones, showed no stimulation at lower concentrations. Previous unpublished results using the NEP fractions from *H. virescens* antennae homogenates show similar Na⁺-K⁺ ATPase activity responses to specific chemical attractants. In contrast to the strong inhibition of Na⁺-K⁺ ATPase activity in antennae NEP preparations at 10⁻⁴M pheromone, cow olfactory tissue NEP preparations showed greatest stimulation of the enzyme activity at 10⁻³ and 2.5 x 10⁻³M (equivalent to 10⁻⁶ and 2.5 x 10⁻⁶ mole per ml) concentrations for different odorant chemicals. Preliminary studies on the effects of partial replacement of the phospholipids in SDS solubilized NEP preparations indicate that both type of fatty acid and phosphate binding group (e.g. choline etc.) are important for odorant perturbation of ATPase activity. Brain Na⁺-K⁺ ATPase activity showed no response to the above concentrations of pheromone components.

A Parametric Study of the Stimulation Variables Affecting the Magnitude of the Olfactory Response II. Testing Conclusions Drawn from Reductions of the 3-variable Model. D. B. KURTZ*, M. M. MOZELL, S. W. SWIECK (SUNY, Upstate Medical Center, Syracuse, N. Y. 13210) and D. E. HORNUNG (St Lawrence University, Canton, N. Y. 13617)

In their parametric study, Mozell, Sheehee, Swieck, Kurtz and Hornung (manuscript in preparation) have shown the effects of volume (V) and time (T) to be equal and opposite and the effects of number of molecules (N) and T to be equal and opposite. Because of these relationships, two models involving flow rate (F) and concentration (C), have been found to be excellent predictors of the response (i.e., the [F,N] and [C,T] models, respectively). In addition, since the effects of N and T were not found to be equal and opposite, the [D,V] model (involving delivery rate) predicted the response poorly. To test these results, 3 sets of 4 sniffs were presented to each of 6 frogs. In set 1, F and N remained constant while V and T increased in tandem. In set 2, C and T remained constant while N and V increased together. In set 3, D and V were held constant while N and T increased in tandem. The results show that, as expected, set 1 sniffs produced the same size responses despite an 8-fold increase in V and T, and set 3 sniffs produced successively larger responses as N and T increase together despite a constant D and V. However, set 2 sniffs also produced successively larger responses (to a much smaller degree than the [D,V] sniffs) as N and V increased together despite a constant C and T. These results support the previous findings that the effects of V and T are equal and opposite, and the conclusion that the effect of F adequately incorporates the combined effects of V and T. In addition, the conclusion that D does not adequately incorporate the combined effects of N and T is also supported. However, because the size of the response changes with a constant C and T, C can not be considered to adequately incorporate the combined effects of N and V.

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Perceived Intensity and Quality of Binary Odor Mixtures. DAVID G. LAING, HELMUT PANKHUBER, MARY E. WILLCOX* and ELIZABETH A. PITTMAN (CSIRO Division of Food Research and *Division of Mathematics and Statistics, P.O. Box 52, North Ryde, Sydney, NSW, Australia 2113)

As yet no method has been developed for predicting the odor quality of a mixture. Accordingly the main aim of this study was to investigate how the two principal factors, intensity and quality, affect perception of components in binary mixtures. The stimuli were 4 dissimilar odorants, each of which was readily identifiable and described by a single commonly used word; benzaldehyde (almond), eugenol (cloves), propionic acid (vinegar), and α -carvone (spearmint). For each of the 6 possible odor pairs there were 36 stimuli; 5 concentrations of each odorant, 25 mixtures and a blank (deodorized air). The 36 stimuli of each odor pair were presented via an air dilution olfactometer in a randomised sequence and each stimulus was evaluated 3^x over 6 sessions using an interval scaling procedure. These results indicated that: (i) the odorant with the highest (unmixed) intensity always predominated or was the only component perceived. (ii) both odorants were perceived when the (unmixed) intensities of the components were approximately equal. (iii) Only one odorant was perceived when the intensity differences between components exceeded a critical value. This was frequently demonstrated as a relatively sharp change in quality. (iv) Maximum interaction (suppression) occurred between components of high intensity whilst weak odorants had little or no effect on others of weak or strong intensity. (v) There was no evidence for synergism. (vi) An excellent correlation of predicted and experimental values was obtained with the vector model of intensity summation (Berglund et al. 1973), but this model cannot account for the sharp transitions in quality which occurred. Overall it is the intensity not the specific quality of a component which determines whether it will be perceived in a binary mixture. This result indicates that advocates of analytic or synthetic functioning for olfaction will need to think again if they are to account for the principles of interaction found here.

Sniffing Naturally Is the Best Way to Perceive Odors. DAVID G. LAING (CSIRO Division of Food Research, P.O. Box 52, North Ryde, Sydney, NSW, Australia 2113)*

Recently Laing (1982) described the characteristics of human sniffing episodes during odor perception and suggested that the techniques used by individuals may be close to those providing optimum perception. This suggestion is investigated in the present study with twenty one humans during threshold and intensity tests with butanol, cyclohexanone and pentyl acetate. Olfactory responses obtained using natural sniffing techniques were compared with those where the number of sniffs, interval between sniffs and size of sniffs were varied. The results indicate that it is very difficult to improve on the efficiency of sniffing techniques of individuals and that a single natural sniff provides as much information about the presence and intensity of an odor as do seven or more sniffs. A single natural sniff and the first sniff of a natural sniffing episode were shown to have similar characteristics and most significantly both were unaffected by changes in the concentration and type of odor. Overall, the results indicate that humans achieve optimum odor perception during threshold and intensity measures using their natural multi-sniff technique, or with a single sniff. For the 'average' human this occurs with a sniff of inhalation rate, volume and duration of 30 L min⁻¹, 200 cm³ and 0.4 s respectively. The use of several sniffs in a sniffing episode appears to be a confirmatory action rather than a necessary one, except for the perception of odor mixtures where several sniffs are likely to be needed to aid discrimination of the components. Data from the present and earlier study (Laing 1982) provide the information necessary for the development of a standard olfactometer and standard procedures for measuring the olfactory responses of humans.

LAING, D.G. Perception 11, 221-230, 1982

The Effect of the Withdrawal of Diuretics on the Preference for Salt, the Perception of Salt Intensity and the Sodium Intake of Well-controlled, Free-living Hypertensive Individuals. BARBARA LEVINE (New York University, NY, NY) MABEL M. CHAN (New York University, NY, NY)

The purpose of the study was to determine whether the withdrawal of diuretic antihypertensive therapy had an effect on the preference for salt, the perception of salt intensity and the sodium intake of hypertensive individuals and, concomitantly, to determine whether the dietary sodium restriction in a subgroup deprived of diuretics produced a different effect. Data were collected from 82 hypertensive patients who had been on diuretic antihypertensive medication for five years. The control group consisted of those patients whose use of diuretics was continued. The two treatment groups consisted of those patients whose use of diuretics was discontinued with one group receiving dietary intervention for a low sodium, high potassium diet. The patients were tested at baseline (while on diuretics) and four months after treatment was initiated for preference for salt, perception of salt intensity, sodium intake (as determined by urinalysis), and degree of sodium depletion (as determined by plasma renin concentration). Factorial analysis of variance with repeated measures on the data indicates that the three treatment modalities were not found to have different effects on changes in preference for salt or perception of salt intensity between baseline and four months later. Differences among treatment modalities also were not found for sodium intake as reflected by urinary sodium. However, differences were found among treatment modalities for changes in sodium depletion (plasma renin concentration). Patients who were withdrawn from diuretics on the average experienced a greater decrease in renin than patients who were not withdrawn from diuretics implying that there was a change in sodium balance produced by the withdrawal of diuretics. Since urinary sodium did not change, there is no evidence that sodium ingested was increased by the modest negative balance during chronic treatment with diuretics. Salt appetite, as reflected by salt intake and salt taste perceptions, appears to remain stable despite shifts in sodium balance.

Prolonged, Exaggerated Intake of Saccharin Induces Morphine-Tolerance in Rats. ISRAEL LIEBLICH, EDNA COHEN, JUDITH GANCHROW, ELLIOTT M. BLASS, and FELIX BERGMAN (The Hebrew University, Jerusalem, Israel).

Gustatory afferent pathways offer an opportunity for interaction with opiate and catecholamine systems thought to be important in pleasure and reward. One such relation has been reported: rats genetically selected for high rates of self-stimulation (infer: pleasure, reward) exhibited increased consumption of saccharin (1,2). We hypothesized that, if increased release and utilization of endogenous opioid peptides contributed to these behaviors, they might be reflected in cross-tolerance to morphine. We examined the effect of prolonged exposure to 3 mM sodium saccharin on latency of pain response (hotplate test) following morphine and saline injections. Excessive saccharin intake for 28 days cancelled the strong pain-relieving effect of morphine injection (2.5 or 5.0 mg/kg) easily observed in the control group receiving only water to drink for 28 days. We conclude that chronically elevated saccharin intake can interact with the endogenous opiate system as reflected by morphine tolerance.

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Anterior to Posterior Variation in Neurogenesis, Epithelial Thickness and Receptor Cell Density in the Olfactory Epithelium of the Tiger Salamander. ALAN MACKAY-SIM (University of Wyoming), URMÍ PATEL (NIMH).

Recent electrophysiological studies of the salamander olfactory epithelium have revealed two types of odorant-induced patterns of responses across different epithelial regions: first, regional patterns of responsivity which were characteristic of each odorant; second, an anterior to posterior gradient in responsivity that is independent of odorant and concentration. This last finding led us to hypothesize anterior to posterior gradients in epithelial thickness, cell density or rates of neurogenesis. The present study investigates this hypothesis. Epithelial thickness was measured at standardized distances along the anterior to posterior axis of epithelia from 13 animals whose heads were processed for paraffin histology and cut at 10 μ m. Neurogenesis and cell density were measured in six animals injected with 3H-thymidine (5 μ Ci/g b.w.) and processed after 5 or 20 d survival for Epon embedding and autoradiography. Longitudinal sections of 1 μ m were examined using an Apple II computer with a digitizing pad viewed through a camera-lucida attachment to a microscope. All receptor, supporting and basal cell nuclei were counted, labelled nuclei being categorized separately. Electron microscopic analysis allowed us to better estimate the proportions of receptor cells in different epithelial regions. These experiments indicate clear anterior to posterior gradients in receptor cell density, rates of neurogenesis and epithelial thickness consistent across animals. The anterior epithelium is thicker, contains more cells and undergoes neurogenesis more slowly than the posterior. Such anterior to posterior differences could possibly provide a chemical polarity within the epithelium, upon which could rest the mechanism for the correct maintenance of epithelium-to-bulb connectivity.

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Dexamethasone Suppression Test Results Do Not Predict Reported Taste Loss in Psychiatric Patients. NAOMI E. LOHR, PH.D. (University of Michigan), DOUG KING, M.D. (University of Michigan), ANDREA L. JACOBS (University of Michigan)

The dexamethasone suppression test (DST) has been shown to be useful in distinguishing depressed patients with an endogenous profile (i.e. melancholia) from depressed patients without this clinical profile. About 50% of endogenous depressives fail to adequately suppress cortisol secretion ($< 5 \mu\text{g/ml}$) after oral administration of dexamethasone whereas only 5-10% of non-endogenous depressives and other psychiatric patients show inadequate suppression. Failure of dexamethasone to suppress cortisol secretion is thought to indicate limbic-hypothalamic dysinhibition.

We studied the relationship of DST results and reported taste loss in 20 endogenously depressed patients. As suggested by our pilot results, reported taste loss was not significantly different in DST suppressors and nonsuppressors suggesting that different mechanisms underly reported taste loss and failure of dexamethasone suppression in endogenous depressives.

Gustatory responsiveness of neurons in the hamster thalamus. ALAN MACKAY-SIM (Dept. Zool. and Physiol.), DAVID V. SMITH (Dept. Psychol., Univ. Wyoming)

There is now a substantial volume of data on the gustatory responsiveness of peripheral and brainstem neurons in the hamster. The present study reports preliminary data from single cells in the ventromedial portion of the ventrobasal thalamus, recorded from 9 male hamsters, anesthetized with urethane. Gustatory thalamus was located using anodal electrical stimulation of the anterior tongue. Chemical stimuli were: 0.1 M sucrose, 0.03 M NaCl, 0.003 M HCl and 0.001 M QHCl. Twenty-six neurons were isolated from within the region of the electrically evoked response. Of these, 13 responded to chemical stimulation of the anterior tongue. The responsive cells had a mean spontaneous rate of 2.3 spikes/sec. Excitatory responses were predominantly phasic and inhibitory responses were mainly tonic. Consequently, a cell was considered to be excited by a stimulus if the mean firing rate over the first 3 sec of stimulation differed from the mean spontaneous rate by more than 1.64 times its standard deviation (z-score, $p < .05$). A cell was considered inhibited if its firing rate over the first 10 sec of stimulation differed from the immediately preceding spontaneous rate according to a t-test ($p < .05$). By these criteria, of 52 stimulus presentations to these cells, 29% were excitatory and 17% were inhibitory. Of the 13 responsive cells, 7 were only excited by these stimuli, 3 were only inhibited and 3 showed both excitatory and inhibitory responses. Median excitatory responses were 2.8, 4.2, 3.5 and 1.6 spikes/sec to S, N, H and Q, respectively. Of particular interest with regard to quality coding is the increased inhibition observed in the thalamus compared to brainstem levels and the increased responsiveness to QHCl (6 out of 13 cells were excited by QHCl). These results are comparable to those in the rat (Scott & Valowitz, 1978) and the dog (Ninomiya & Funakoshi, 1982) in terms of spontaneous rates, proportions of excitatory and inhibitory responses, and firing rates to excitatory stimuli.

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Chemical signals of female gender to naive male mice are of rostral origin and have low volatility. JOHN C. MAGGIO, GLAYDE WHITNEY (Florida State University)

In a series of five experiments, we examined the ability of socially naive male mice to detect the presence of a female (as measured by the males' emission of ultrasonic vocalizations). Exp. 1 demonstrated that males, when restricted from extensive female contact via a single wire-mesh partition, showed a lower incidence of ultrasound emission and emitted less ultrasound with longer latencies than they and other males did when allowed to freely interact with a female. Exp. 2 showed that a double wire-mesh partition, prohibiting any male-female tactual contact, completely eliminated naive male ultrasound production. This was true even in the presence of heavily soiled female cage shavings (Exp. 3). Exp. 4 showed that the double partition was less effective in inhibiting male ultrasound emission if the males had previously had one 3-min. unrestricted exposure to a female 48 hrs. earlier. The above results suggest that chemosensory recognition of the presence of a female by socially naive male mice depends on close social interaction. Further, if there is a female-secreted chemosignal of gender to naive males, it is (a) of extremely low volatility, and (b) not present in female-soiled cage shavings. Thus, Exp. 5 examined what female body areas naive males investigated and ultrasounded to during their first post-weaning heterosexual social interaction. Results indicated that naive males pay particular attention to the rostral portion of the female, specifically the chin and ventral neck region. Ultrasound emission was found to highly correlate with male contacts of these areas. These findings (1) support recent contentions (Wysocki, Nyby, Whitney, Beauchamp, & Katz, 1982) that the chemosubstance(s) responsible for male ultrasound emission is of low volatility and (2) suggest that the anterior portion of the female, and not urinary or vaginal secretions, may be the source of the ultrasound eliciting signaller of "female" to naive males.

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Pheromone Detection and Discrimination by *Trichoplusia ni* at Behaviorally Relevant Concentrations. R. W. MANKIN, A. J. GRANT, and M. S. MAYER (USDA, ARS, Insect Attractants, Behavior and Basic Biology Research Laboratory, P.O. Box 14565, Gainesville, FL 32604)

Typically male moths discriminate and respond to low concentrations of their pheromone blends with various sexual behaviors. Because the discriminative process at the periphery and the central nervous system is dependent upon concentration and because it can be manipulated for insect control purposes, it is important to study the peripheral neural responses at behaviorally relevant concentrations. We compare the response functions of various pheromonally excited behaviors (orthokinesis and upwind anemotaxis) of male *Trichoplusia ni* (Hübner) to the peripheral neural responses at these relevant concentrations. The necessity for psychophysical comparisons is discussed and illustrated.

Olfactory Perception in Korsakoff's Psychosis: Correlation with Brain Noradrenergic Activity. ROBERT G. MAIR (VA Medical Center, Davis Park, Providence, RI), RICHARD L. DOTY (Smell and Taste Center, University of Pennsylvania, Philadelphia), and WILLIAM J. MCENTEE (VA Medical Center, Davis Park, Providence, RI)*

In patients with Korsakoff's psychosis, the ability to discriminate odors is impaired without apparently affecting absolute olfactory sensitivity or odor memory (Mair et al., J. Exper. Psychol. 6: 445, 1980). Korsakoff's psychosis has also been associated with reduced central noradrenergic (NE) activity (McEntee & Mair, Science 202: 905, 1978). We have studied odor discrimination with a recognition memory paradigm and odor identification using the University of Pennsylvania Smell Identification Test (UPSIT), a forty item test in which microencapsulated odorants are matched to one of four verbal descriptors. Groups of Korsakoff patients were impaired in their ability to perform both the odor discrimination (n=16) and identification (n=23) tasks. There were significant correlations between a measure of brain NE activity (the concentration of 3-methoxy-4-hydroxy-phenylglycol in lumbar CSF) and performance on both the discrimination ($r = 0.60$, $n = 16$, $p < .01$) and identification ($r = 0.91$, $n = 7$, $p < .01$) tasks. The relationship between brain NE activity and olfactory perception is consistent with anatomical evidence of an extensive NE innervation of the olfactory bulb and all of its primary projection areas. The results also demonstrate that the UPSIT, an easily administered and standardized test of olfactory function, provides a reliable measure of olfactory perception comparable to that provided by more elaborate psychophysical methods.

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Bursting Characteristics of Neurons in the Nucleus of the Horizontal Limb of the Diagonal Band. J. E. MARCHAND and F. MACRIDES (Worcester Foundation for Experimental Biology, Shrewsbury, MA 01545)

The nucleus of the horizontal limb of the diagonal band (HDB) is the principal source of cholinergic projections to the olfactory bulbs and to the cortical regions which receive inputs from the olfactory bulbs (Macrides et al., JCN, 203: 495-514, 1981). The present study examined the possibility that the HDB drives θ activity in the olfactory system, and compared the bursting characteristics of single units in the HDB with the hippocampal θ rhythm that is driven by cholinergic neurons in the medial septum. Recordings were made in paralyzed hamsters which had their normally pulsatile nasal airflow interrupted; units which support centrifugal projections to the main olfactory bulb (MOB) were identified using antidromic stimulation (constant latency, high frequency following, and collision tests) and unit recording sites were verified using iontophoretic injections of pontamine sky blue. The antidromic stimulation electrodes in the MOB also served as recording electrodes. The antidromically driven units in the HDB exhibited periods of bursting in the θ range. The slow waves recorded in the MOB also exhibited periods of θ activity, as did HDB units which could not be driven antidromically. The latter units may drive θ activity in other projection targets of the HDB. Similar units were recorded in the adjacent region thought to correspond to the nucleus basalis (NB) of primates and to support cholinergic projections to medial and frontal neocortex. The θ activity in the HDB, NB, and MOB was not invariably correlated with that in the hippocampus, indicating that θ pacemaker activity in the medial septum can be decoupled from that in other components of the medial septum-diagonal band complex and thus that the θ activity of neurons in the HDB and NB is not driven by the medial septum. These various findings support the hypothesis that neural mechanisms previously thought to be characteristic of the septo-hippocampal system also operate in the olfactory system.

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Magnitude Matching of Taste Intensity: Which Modalities Work? L.E. Marks (J.B. Pierce Fndn., Yale U.), J.C. STEVENS (J.B. Pierce Fndn., Yale U.), J.F. GENT (J.B. Pierce Fndn., U. Conn. Health Center), and L.M. BARTOSHUK (J.B. Pierce Fndn., Yale U.).

Not all perceptual modalities appear equally capable of providing absolute cross-modality matches. In particular, the present studies indicate that lightness of grays, though a convenient continuum to use, may be too malleable to provide absolute cross-modality matches. An earlier report at these meetings (Marks, Rifkin, Bartoshuk, & Stevens, 1980) showed that the method of magnitude matching (Stevens & Marks, 1980) can be used effectively to distinguish tasters from non-tasters of the bitter substance 6-n-propylthiouracil (PROP). When asked to give magnitude estimates of both the taste intensity of PROP and NaCl and the loudness of a 1000-Hz tone, the derived cross-modality matching functions readily distinguished PROP non-tasters from tasters (the classification based on an independent, threshold operation). In the present study, we asked each of 13 non-tasters and 28 tasters to judge within a single session the taste intensity of PROP, sucrose, and NaCl, the loudness of 1000-Hz tones, and the lightness of gray surfaces. Cross-modality matches of taste intensity to loudness and to grays alike appeared to distinguish non-tasters from tasters. However, a final experiment questioned that conclusion, suggesting that it was the context of the loudness judgments that made absolute matching of grays to taste possible. This experiment asked two groups of subjects to judge the taste intensity of NaCl and the lightness of grays; each group of 13 Ss received a different range of NaCl concentrations. The results indicated that the subjects made relative but not absolute comparisons of taste to lightness. Hence it appears that subjects were able to use loudness but not lightness as a yardstick to assess taste intensity.

Physiological and Morphological Changes in the Salamander Olfactory Epithelium After Nerve Transection. LEONA M. MASUKAWA, BRITTA HEDLUND, THANE BENSON and GORDON M. SHEPHERD (Section of Neuroanatomy, Yale University School of Medicine, New Haven, CT 06510)

The morphology and physiological properties of cells in the olfactory epithelium of the tiger salamander, *Ambystoma*, have been examined in normal animals and at 1, 2 and 4 weeks after olfactory nerve transection. Intracellular recordings were obtained and cells were classified into 3 main groups. Type I, presumed supporting cells, had properties similar to glial cells, i.e., high resting membrane potentials, low input resistances and no spike activity. Type II were receptor cells because of their intermediate position within the epithelium, their spiking activity, and high input resistance. Type III, presumed immature receptor cells, were located deep in the epithelium and had the characteristics of receptor cells except that they lacked the ability to generate spikes. One week post transection, the ratio of Type II to Type I cells encountered was 0.5:1.0, and increased to 0.83:1.0 after 2 weeks and 1.5:1.0 after 4 weeks. By approximately 4 weeks after transection, the receptor cell population had increased to control levels. The ratio of Type III to Type I cells was unchanged during these time periods. The decrease and recovery in the receptor cell population was paralleled by changes in the receptor cell layer as shown in lum plastic sections. During regrowth, the spikes recorded from presumed receptor cells characteristically showed multiple components. Unexpectedly, a population of elements similar to Type I cells was observed that had higher input resistances indicating a possible loss of electrical coupling. This study thus provides evidence for electrophysiological changes in both receptor and sustentacular cells after nerve transection.

Behavioral and Physiological Effects of Capsaicin in Red-Winged Blackbirds. J. RUSSELL MASON (Monell Chemical Senses Center), JOEL A. MARUNIAK (Dept. of Physiology and Clinical Smell and Taste Research Center, University of Pennsylvania).

We injected red-winged blackbirds (*Agelaius phoeniceus*) subcutaneously with capsaicin and assessed (a) changes in basal body temperature, (b) ability to discriminate warm from cool drinking water, and (c) sensitivity to oral and topical applications of capsaicin.

As predicted from studies with mammals, the injections disrupted thermoregulation when the ambient temperature increased, and eliminated discrimination between warm and cool drinking water. In contrast to effects on mammals, however, the injections also disrupted thermoregulation when the ambient temperature decreased. In further contrast, injections failed to observably diminish oral or topical sensitivity to capsaicin and apparently induced a capsaicin preference in two-bottle drinking tests.

To our knowledge, the present work is the first to report physiological and behavioral effects of capsaicin on birds, and the first evidence that such exposure can induce preference behavior, at least under some circumstances, in an infrahuman species.

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Chorda Tympani Nerve Responses to Taste Stimuli in Young and Aged Rats. M. MCBRIDE (School of Nursing) and C.M. MISTRETTA (Schools of Dentistry & Nursing and the Center for Human Growth & Development, Univ. of Michigan, Ann Arbor MI 48109)

To determine whether taste responses differ as a function of age, neurophysiological recordings were made from the chorda tympani nerve in male and female Fischer 344 rats. Three age groups were studied: 5-7 mos (N=18); 24-25 mos (N=20); 29-32 mos (N=21). Chemical stimuli were 5 salts (0.1M NaCl, LiCl, KCl, NH₄Cl, MgCl₂), 2 acids (0.025M citric acid, 0.01N HCl), 1.00M sucrose, 0.04M quinine HCl, and concentration series (0.025 - 0.75M) of the 5 salts. A significance level of $p < 0.05$ was used for data analysis. All chemicals elicited taste responses at all ages. However, integrated responses, relative to NaCl, increased with age for NH₄Cl and decreased for MgCl₂. Relative to NH₄Cl, KCl and MgCl₂ responses decreased. When shapes of the response/concentration functions were compared across age groups, the curve for NaCl in the youngest age group was significantly different than that for the two older groups. No other differences in salt concentration functions were observed. Response ratios for sucrose increased and those for citric acid decreased, relative to NaCl. All age-related trends were in similar directions for males and females, except citric acid ratios which differed as a function of age for males only. Although significant age-related differences in some response ratios were found, they were relatively small. The most substantial changes were obtained for MgCl₂ and sucrose response ratios, and the differences were greatest between youngest and oldest groups. In general, there are not substantial deficits in response magnitudes from aged taste buds to a wide range of stimuli and intensities.

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Further Evidence of Taste Enhancement with Simulated Human Drinking. HERBERT L. MEISELMAN (U.S. Army Natick R & D Labs, Natick, MA 01760), BRUCE P. HALPERN and STEVEN KEL-LING (Cornell University, Ithaca, NY 14853).

Meiselman and Halpern (1973) demonstrated significant enhancement of perceived taste intensity using an alternating stimulus of NaCl or sucrose and water. Halpern & Meiselman (1980) also failed to observe any taste sensory adaptation with variations in pulse duration, stimulus compound, stimulus concentration, or individual subjects. The present studies used improved apparatus, improved procedure, different subjects, and a better temporal stimulation model of human drinking. Six studies were done using alternating taste stimuli. Tastants were 50, 250, or 500 mM NaCl. Alternating liquids were distilled water, 36 mM Na⁺ artificial saliva, the three NaCl tastant solutions, or no liquid (air). Significant enhancement of taste intensity was observed with all three tastant intensities, and with either water or artificial saliva as the alternating stimulus. The adaptation function obtained by alternating NaCl with itself was not different than the function obtained by alternating NaCl with air. Hence, simple cessation of flow is not sufficient to produce enhancement. The condition modelling human sipping (1 sec tastant and 3 sec alternating liquid) produced enhancement. Enhancement was obtained when the 50 mM tastant was alternated with the 36 mM Na⁺ artificial saliva. This provides further evidence of the sensitivity of the taste system to small changes, in agreement with McBurney's (1976) demonstration of gustatory sensitivity for slightly different alternating stimuli. Alternating taste stimulation deserves more widespread attention as a method and a research topic.

Vomerolnasal Lesions Before Sexual Experience Impair Male Mating Behavior in Hamsters. MICHAEL MEREDITH (Dept. Biol., Florida State University, Tallahassee, FL 32306).

Sexual experienced male hamsters continue to mate after vomeronasal or olfactory lesions but not after both systems are damaged (Meredith et al, 1980, Science 207, 1224). The effect of experience was investigated in five min. mating tests with either behaviorally receptive intact females (female test) or anesthetized males whose anogenital area was smeared with vaginal fluid from a receptive female (scented male test). All animals were tested twice preoperatively and twice postoperatively with each test in counterbalanced order. Compared with sham operated males, the performance of animals whose vomeronasal organs (VNOs) were removed before their first post-weaning contact with females (VNXi group) was severely reduced (e.g. 72% fewer intromissions per minute (I/M) in the female test ($p < 0.002$) and 53% fewer mounts with pelvic thrusts (MT) in the scented male test ($p < 0.02$). In contrast, VNXe-group animals whose VNOs were removed after some sexual experience (3 ejaculations) showed no reduction in performance following surgery and an increase in performance with repeated testing. Even after equivalent opportunities to interact with females (3 one hour sessions) the mean performance of VNXi animals was still significantly lower than that of VNXe animals ($p < 0.002$, female test I/M; $p < 0.05$, scented male test MT). The results suggest that the difference between groups may be due to the failure of the first mating interaction. 40% of VNXi animals showed no mating behavior whatever in any postoperative test. The other VNXi animals did not differ from VNXe animals. These results support the hypothesis that vomeronasal sensory input is most critical for inducing appropriate response in naive animals and that experienced animals can use alternative, probably olfactory, cues (Meredith, 1983, In Pheromones and Mammalian Reproduction. J. Vandenberg, ed. In Press). After VNX all animals could discriminate food odors. Preliminary Histology indicates no damage to the olfactory system and near complete removal of vomeronasal organs. Neither the presence of residual vomeronasal tissue nor testosterone levels at sacrifice were postoperative behavior.

Ultra-Rapid Freezing Studies on Rat Olfactory and Nasal Respiratory Epithelia. B. Ph. M. MENDO (Neurobiology, Northwestern University, and Psychological Laboratory, Utrecht, The Netherlands).

Freeze-substitution of neither chemically-fixed, nor cryo-protected olfactory epithelium of the Sprague-Dawley rat indicates the absence of arm-like structures on the ciliary axonemal doublets. Tapering elements of those cilia usually have rather electron-dense matrices. Membranes of olfactory cilia and supporting cell microvilli can be distinguished on the base of their electron-density. Freeze-etching, using deep-etching, indicates the presence of different mucous textures in olfactory and respiratory epithelium surfaces. Intramembranous particle densities of most structures are lower than in chemically fixed, cryo-protected tissue samples. In addition to particles, the membranous fracture faces display holes upon etching. Pooling densities of holes and particles gives values matching those found previously in fixed cryo-protected tissue, indicating that such holes are most likely etching artifacts formed at the site of particles. Particle densities at outer membrane surfaces are higher than intramembranous particle densities; highest densities are found at inner membrane surfaces. In contrast to outer membrane surfaces, the relative distribution of the latter densities over nine different regions of various cells does not match that of the fracture faces, indicating that outer membrane surface particles are presumably more closely related to fracture face particles than inner membrane surface particles. Densities of intramembranous particles, intramembranous particles + holes and outer surface particles are highest in supporting cell microvilli, followed by the olfactory cilia and respiratory cell microvilli and lowest in respiratory cilia. This distribution pattern resembles that found for fracture face particles in fixed cryo-protected tissue. Preliminary experiments did not reveal altered particle distributions upon odor stimulation.

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Taste Bud Distribution in The Hamster. INGLIS MILLER, JR. (Bowman Gray School of Medicine, Wake Forest University) and DAVID V. SMITH (University of Wyoming)

Current experiments indicate a convergence on neurons of the nucleus of the tractus solitarius by nerve impulses generated from separate taste receptor populations in the hamster. Definition of the receptor distribution in this species is required to interpret these results. Observations were obtained from microscopic examination of serial sections of entire heads from young hamsters 19-26 days of age and from excised tissue of animals 120 - 150 days old. Adult hamsters contained a mean, bilateral total of 723 taste buds. Fungiform papillae contained one taste bud each for a mean of 65 (s.d.= 10, N=6) per unilateral half of the tongue or 18% of the total with no differences between young and adult animals. Foliate papillae in adults contained 115 (s.d.=13, N=6) taste buds per side for 32% of the total, while young hamsters had 71 (s.d.=12, N=3). A single vallate papilla possessed 168 (s.d.=27, N=4) taste buds in adults (23% of total) and 81 in the young (s.d.=22, N=4). The region of the nasoincisor ducts contained a mean of 6 (s.d.=1.4, N=10) taste buds per side for 1.7% of the total. There was a mean of 88 (N = 2) taste buds on the soft palates of adults (12%) and a mean of 63 (s.d.=7, N=4) on young hamsters. The epiglottis and contiguous regions of the esophagus contained a mean of 70 (s.d. = 9, N=5) taste buds (10% of total) with no difference between young and adult hamsters. Other regions with taste buds included the sublingual organ containing a total of 5 (0.7%), the buccal wall with a bilateral total of 10 (1.4%), and the nasopharynx with a total of 10 (1.4%). Apparent differences between young and adult hamsters require additional study. Inference about quality coding in neurons of the central gustatory pathway requires appropriate consideration of the convergence from receptors of diverse populations.

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Most mammals demonstrate a preference for the taste of sodium when it is offered in low concentrations but they show an aversion to higher concentrations. To determine the ontogeny of the preference-aversion function for sodium in rats, rat pups of several ages were offered e-ionized water or one of a wide range of NaCl concentrations. The solutions were presented by infusion through an anterior mouth catheter while the pups were in a warm chamber.

Five-day-old pups showed no evidence of a preference or aversion for sodium with the concentrations tested, in comparison to water intake. However, 12-day-old pups consumed significantly more saline than water when the saline concentration offered was in the 1.5% - 3% (wt/vol) range.

These results suggest a shift in sodium preference with age. Numerous investigators have found that adult rats prefer relatively weak concentrations of saline (approx. 1.5% or less) and find stronger concentrations aversive. But 12-day-old pups, as reported here, are indifferent to saline concentrations of less than 1.5% and actually prefer concentrations that adult rats reject. These data are consistent with data reported for humans; new born infants are indifferent to a range of NaCl concentrations which are preferred by 6-month-old children (Mallor & Desor, 1974; Desor, 1975) and rejected by 1½ - 3 yr-old children (Beauchamp & Maller, 1977).

The difference between the neonatal and adult preference-aversion functions for sodium may reflect the development of the rat's gustatory system. Many of the rat's gustatory structures mature largely after birth (Mistretta, 1972; Hill & Almlí, 1980). Thus, neonatal rats may be less sensitive to NaCl than adults and the appearance of the adult-like preference-aversion function for sodium might be tied to the maturation of the gustatory system.

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What Is The Evolutionary And Functional Significance Of Cilia And Microvilli Located At The Site Of Stimulus Reception On Chemoreceptors? DAVID T. MORAN, J. CARTER ROWLEY III, and BRUCE W. JAFEK, Departments of Anatomy and Otolaryngology, University of Colorado School of Medicine, Denver, CO 80262.

A survey of the fine structure of vertebrate and invertebrate sense organs reveals that a wide variety of photoreceptors, mechanoreceptors, and chemoreceptors employ cilia and/or microvilli at the site of stimulus reception. Photoreceptors: The rods and cones of the vertebrate retina, for example, have a connecting cilium that supports the rhodopsin-bearing outer segment. Invertebrate eyes, on the other hand, are often rhabdomeric; the visual pigment lies atop an ordered set of microvilli. Mechanoreceptors: Many invertebrate mechanoreceptors, such as chordotonal sensilla found in Arthropod proprioceptors, have cilia at the site of stimulus reception. Vertebrate mechanoreceptors, such as the hair cells of the Organ of Corti in the cochlea, have an ordered array of stereocilia--which are actually microvilli--at the site of mechanical stimulation. Chemoreceptors: Taste receptors from different animals can either be ciliated or microvillar. The contact chemoreceptors of insects, for example, center their function around bipolar neurons that bear cilia. Cells from mammalian taste buds, on the other hand, have microvilli upon their sensory surface. Likewise, olfactory receptors from different animals have been shown to be ciliated, microvillar or, as in the case of the duck, both. Mammals typically have ciliated olfactory receptors in their olfactory epithelium, and microvillar olfactory receptors in their vomeronasal epithelium. Aquatic vertebrates such as the goldfish typically have ciliated and microvillar receptors commingled throughout the olfactory epithelium. Humans have ciliated olfactory receptors; their olfactory epithelium contains microvillar cells whose ultrastructure suggests they may be sensory cells.

What is the significance of the frequent use of cilia and microvilli by chemoreceptor cells? It seems likely that sensory cells have been evolutionarily conservative in utilizing two readily available, biologically ancient classes of proteins, tubulin and actin, to construct two classes of cell surface extensions--cilia and microvilli--that present specialized transducer regions of the cell membrane for optimal exposure to chemical stimuli. Supported by NSF (BNS 8210327) and NIH (NS 15203 and RRQ0592).

Specificity of Glucose Satiety in Rat.
DOUGLAS G. MOOK (University of Virginia)

Hungry but non-thirsty rats drink concentrated glucose solutions with avidity, but the postingestive effects of such solutions produce rapid and prolonged satiety: Rats drink small quantities, and show little further drinking if access is prolonged. Severity of deprivation does not affect amount drunk. However, intake of the same commodity (glucose) in powdered form has quite different properties: Amount ingested rises with deprivation to values significantly higher than those associated with solutions. This happens even if the powder is presented only after solution intake has come to an end, and at a time when solution intake would remain inhibited. This is not a matter of palatability, novelty, or rejection of liquidity per se. And since the postingestive consequences of solution- and powder-intake rapidly converge, we suggest that the stimulus properties of the diet must feed forward to set the "criterion" of satiety, determining the value of the postingestive load that the controlling system will accept.

Olfactory mucosa transplanted into the fourth ventricle.
E. E. Morrison and P. P. C. Graziadei. Florida State University, Tallahassee, Florida 32306

The olfactory neuron of the adult animal can be replaced from a neurogenic matrix, however it is not known what factor(s) govern the neuronal replacement. To better understand the neurogenesis and maturation of this neuron we have utilized the method of brain transplantation. We have carefully transplanted neonatal mucosa (p4-p10) into the fourth ventricle of 20 adult rats. The post operative examination period was 10-90 days. The transplant was attached to the cerebellum; medulla or area postrema; was promptly vascularized, and formed a series of inter-connecting vesicles lined by respiratory and olfactory epithelium. At early survival times the transplanted neurons degenerate and are replaced by new neurons from the neurogenic matrix. At later survival times (> 20 days) the maturing neurons and their fibers are olfactory marker protein positive and are seen penetrating the brain tissue. Although the olfactory fibers penetrate the host brain no apparent glomerulization is evident. When the transplant attached to the medulla or area postrema the host brain reacted by forming a tissue bridge through which myelinated fibers enter the transplant. The transplanted neuroepithelium displayed peculiar characteristics. There appeared to be partial disorganization of the sensory epithelium while the respiratory remained intact. The olfactory neurons formed cord-like structures below the basal lamina, apparently streaming away from the epithelium and penetrating the host brain. We conclude that the olfactory neuroepithelium undergoes rapid neuronal replacement even when removed from its normal locus. The neurons detach from the epithelium and migrate to the host brain; although their axons penetrate the brain they do not form glomeruli. The long term survival and cell interaction are currently being investigated. (Supported by NIH grant NS16421).

Does Preferred Concentration of NaCl, Sucrose or Citric Acid Shift Over the Life Span? CLAIRE MURPHY (San Diego State University)

Modest decrements in both taste threshold sensitivity and, more recently, suprathreshold sensitivity have been associated with the aging process. The present study was designed to investigate the existence of changes in preference for various concentrations of single tastes and of the same single tastes in more complex chemosensory mixtures. One hundred subjects aged 18-26 years and 100 subjects 65 or more years participated. Testing is still in progress on 100 subjects aged 32-45 years. The stimuli were NaCl (.05, .10, .20, .40 M); sucrose (.05, .10, .20, .40 M); citric acid (.0006, .0012, .0024, .0048 M); and the same four concentrations of NaCl in low-sodium vegetable juice, the same four concentrations of sucrose in a lemon-flavored beverage base, and the same four concentrations of citric acid in the same lemon-flavored beverage base. The four stimuli within a stimulus category were randomized, and order of presentation of the six categories was also irregular. Subjects rinsed with deionized water before each stimulus. Subjects judged pleasantness or unpleasantness on a bipolar line scale where distance from zero indicated magnitude of pleasantness or unpleasantness. Marks to the right of zero indicated pleasantness, marks to the left of zero indicated unpleasantness. The peak preferred concentration for each category of stimuli, for each age group was analyzed for age-associated shifts in preference. The experimental design also permitted comparisons among the relative preferences for stimulus categories for the different age groups. Results will be discussed in the light of recent studies on age-associated changes in chemosensory function.

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Behavioral Responses and Chorda Tympani Responses to l- and d- Amino Acids in the Hamster. GEOFFREY H. NOWLIS (University of Connecticut Health Center).

Human beings report that many of the amino acids are either bitter, sweet, or bitter and sweet; the d-isomer is likely to be called sweet, while the l-isomer is more likely to be called bitter. The behavioral responses reported here indicate that hamsters apparently classify these stimuli in a similar fashion, for example, hamsters made ill after tasting d-phenylalanine display an acquired aversion for sucrose, while hamsters made ill after tasting l-phenylalanine display an acquired aversion for quinine. Responses of the whole chorda tympani and of chorda tympani single units help to elucidate such a classification system. The whole nerve is shown to be responsive to a wide range of concentrations of l- and d-alanine, l- and d-phenylalanine, and glycine, as well as sucrose, glucose, fructose, NaCl, and quinine hydrochloride. Units (seven to date of this abstract) are first categorized as most responsive to mid-range concentrations of sucrose (n=3), NaCl (n=2), HCl (n=1), or quinine (n=1), then tested for their responsiveness to the other stimuli mentioned above. Sucrose-best units are nicely responsive to d-phenylalanine, glycine, d-alanine, less responsive to l-alanine; their spontaneous activity tends to be inhibited by both l-phenylalanine and quinine hydrochloride. NaCl-best and HCl-best fibers are not well stimulated by lingual stimulation with these amino acids; the one quinine-best fiber sampled to date was strongly stimulated by l-isomers, less by d-isomers.

Simultaneous Neural Recordings from Glossopharyngeal and Chorda Tympani Nerves of the Mouse. Nejad, M.S., Beidler, L.M. (The Florida State University)

The whole nerve as well as single fiber activities of chorda tympani and glossopharyngeal nerves of several mammals in response to diverse taste stimuli have been studied. However, simultaneous recording from the chorda tympani and glossopharyngeal nerves of a single animal has not been investigated. In present described experiments the tongues of mice (C57BL/6J, DBA/2J, SWR/J) were chemically stimulated and neural responses were simultaneously recorded from chorda tympani (cn) and glossopharyngeal (gn). In order for the chemical stimuli to reach the taste buds in the papillary furrows of the foliate and circumvallate papillae: either, a) the tongue momentarily was pulled out, or, b) the hypoglossal nerve was electrically stimulated to elicit tongue movements. Both methods proved to be effective and seemed to enhance the transport of chemical solutions to taste cells of the foliate and circumvallate papillae. Although both methods were extremely effective in producing gustatory gn response, they had no or little effect on the cn response. Integrated taste responses of gn appeared to be more phasic than those of cn. The relative magnitude of the integrated taste responses of gn to 0.5 M NaCl decreased to about 25% of the initial phasic response, whereas the cn response stayed constant. Decrease of the phasic response to 3mM quinine and 1mM sucroseoctacetate (soa) was about 50%. Both cn and gn of the three strains of mice responded to: NaCl, NH₄Cl, sucrose, citric acid, HCl, quinine, l-arginine, d-tryptophan. SWR/J also responded to soa. The gn response to soa was much higher than the cn response. It was concluded that simultaneous recordings from mouse gn and cn is feasible and their relative contributions to the total taste response can be evaluated for many taste stimuli.

Preferences and Aversions for Sucrose, NaCl, HCl, and Quinine in Rats Under Varying Deprivation Conditions. GEOFFREY H. NOWLIS (University of Connecticut Health Center).

If hunger and satiety can be understood in terms of an organism's ability to regulate its tendency to ingest or reject tasty substances on the basis of its recent history of food intake, one ought to be able to devise a simple test of this model using stimuli representative of the mammal's four taste categories. Booth (1972) devised a simple schedule in which rats' taste preferences were tested either having been just fed their first meal of the day, being ready to eat their second meal two hours later, or being 16 hours food deprived, water being available ad lib. Results only suggestive of an influence of deprivation on preference were found, perhaps because of the complexity of the taste test: two-bottle preference between a saccharin solution and the same saccharin adulterated with (various concentrations of) quinine. The current study used the same schedule conditions, but a potentially simpler taste test: one-bottle one minute consumption of one of three concentrations of sucrose, NaCl, HCl, quinine hydrochloride, or distilled water. Despite some constraints on the dependent variable (a ceiling effect reflecting the amount of fluid an animal can consume in one minute), some clearcut differences in preferences emerged among the three conditions, including preferences for weak, and even medium concentrations of quinine and HCl in those animals ready to eat their second meal. A simple interpretation of the observations of this study would hold that any of the four taste categories may serve as a food cue under the appropriate conditions.

Booth, David. Taste reactivity in starved, ready to eat and recently fed rats. *Physiol. & Behav.*, 8:1069-76 (1972).

A CRITICAL PERIOD EXISTS FOR THE NEURAL INDUCTION OF CIRCUMVALLATE TASTE BUDS. B. Oakley and M.A. Hosley, Department of Zoology, Neuroscience Lab. Bldg., Ann Arbor, MI 48109.

The rat possesses a single midline papilla which contains taste buds that are bilaterally innervated and trophically maintained by the glossopharyngeal nerves (C.N. IX). In our experiments, we investigated the inductive role of the nerve supply upon the formation of the taste buds. The right IXth nerve was removed at 3 days post-partum and circumvallate taste buds counted in operated and control rats at ages 5 to 90 days. By age 90d, normal rats have 625 ± 83 SD circumvallate taste buds with previous work having shown that 88-90% of these taste buds remain following unilateral IXth nerve denervation in adult rats.

In contrast, rats unilaterally denervated at 3d have only 37% of the normal number of taste buds at 90d (230 ± 36). Those taste buds which form are distributed throughout the papilla indicating that the remaining nerve is able to innervate the entire papilla. The deficiency in taste bud numbers was further increased if, in addition to removal of the right IXth nerve at 3d, the left (contralateral) IXth nerve was crushed at the same time; 25 ± 27 circumvallate taste buds form by 90d. When unilateral denervation was carried out on progressively older animals (0d-20d), more taste buds were formed e.g., 476 ± 94 taste buds formed in animals operated on at 20d and sacrificed at 90d. We propose an early sensitive period in which the nerve fibers and taste bud precursors must interact to ensure the formation of normal numbers of taste buds.

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Should All Tastes be Described in Terms of Primaries? M. O'MAHONY, L. ROTHMAN, T. ELLISON, L. BUTEAU. (Dept. Food Science & Technology, University of California, Davis, CA 95616)

Many taste descriptive techniques limit the number of available descriptions to possible combinations of only four adjectives: 'sweet', 'sour', 'salty' and 'bitter'. While doing this, some techniques do not establish a common language between subject and experimenter, defining these adjectives by associating them with suitable standard stimuli. Such lack of definition makes subjects' reports difficult to interpret. Having defined the four descriptive adjectives in terms of standards, the question becomes one of whether the four adjectives are appropriate for describing the tastes of stimuli not generally associated with any one of these individual adjectives (e.g. MSG, sodium benzoate). The answer depends simply on how the four terms are defined. However, it might be asked whether the four adjectives are appropriate, should they be defined in terms of typical stimuli used by psychophysicists. Can the taste of MSG or sodium benzoate be described in terms of the tastes of, say, fructose, citric acid, NaCl and quinine? Definition of the four adjectives and their 11 possible combinations was achieved by requiring subjects to learn appropriate adjectival responses, by identifying the solutes in solutions of NaCl, fructose, QHCl and citric acid and all their 11 possible mixture combinations. The application of these adjectives was broadened by varying the concentrations of the solutes, giving a wide variety of sensations; a total of 59 standard solutions was employed. Once subjects had been trained to respond to all standard solutions with the appropriate adjectives, they then tasted MSG, sodium benzoate, NaHCO_3 , sodium citrate and KCl, believing them to be further mixtures. This was to ascertain whether they would now apply the four adjectives to the new stimuli in any consistent way. They did not. This suggests, after several control experiments, that such adjectives might be inappropriate for describing the taste of these stimuli. This has implications for several taste descriptive techniques.

Detection Thresholds in Mice. R. J. O'CONNELL, J. C. WALKER, S. PARKER, K. HEALY (The Worcester Foundation for Experimental Biology, 222 Maple Ave., Shrewsbury, MA 01545)*

At the last AChES meeting we reported on a computerized apparatus which made it possible to train both hamsters and mice to respond differentially in the presence or absence of odor stimuli. We have now concentrated our efforts on procedures for evaluating detection thresholds in individuals during a single block of trials. To this end the odor delivery port has been equipped with a set of computer-controlled valves and a multi-stage olfactometer has been constructed so that the animals can be presented with one of six different odor streams each of which may contain either an odor stimulus or a blank. The olfactometer can dilute a saturated odor stream by a factor of 1×10^{10} and individual output streams may span any contiguous 2 log unit range of dilution. In the course of measuring detection thresholds we devised a variety of control experiments to verify that discriminative responding was under the control of odor stimuli alone. We have evaluated the ability of mice to exploit, in the absence of odor stimuli, auditory cues associated with operation of the odor valves and differences in flow rate and temperature of the stimulus stream on odor and no-odor trials. We have seen no evidence that any of our subjects can use either the sound or location of the valves as a cue for discriminative responding. However, small differences in either the volume flow rate or temperature of the stimulus stream allowed animals to respond correctly on up to 80% of the trials. When these parameters are held constant, the animals perform at chance levels. We have now determined the concentration-response functions for amyl acetate in the mouse. With each animal, odor concentrations were presented in ascending, descending and random orders. The schedule of stimulus presentation had little effect on the detectability of odor stimuli, although there was a tendency for animals to shift their response times during the course of sessions in which an ascending or descending schedule was employed. Threshold estimates for amyl acetate were approximately 10^{-8} of vapor saturation ($10^{-11.7}$ M).

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Dendritic Ramification and Axonal Branching of Mitral and Tufted Cells in Rat Olfactory Bulb. EDWARD ORONA, JOHN W. SCOTT, AND ELIZABETH C. RAINER, Department of Anatomy, Emory University, Atlanta, Georgia 30322.

The morphology of the output cells of the main olfactory bulb, the mitral and tufted cells, was studied in adult rats. Extracellular iontophoretic injections of horseradish peroxidase were made in the external plexiform layer (EPL). Tissue was reacted by either the Hanks-Yates or diaminobenzidine procedure. Injections labeled small numbers (10-30) of neurons permitting reconstructions of individual cells. Since all labeled cells had dendrites which entered the injection site, analysis was restricted to cells with long dendrites. The cell type labeled was determined by injection depth in the EPL. Secondary dendrites of each cell type were confined to one of three partially overlapping zones in the EPL. Secondary dendrites of most mitral cells (Type I) were confined to the deepest zone of the EPL. An intermediate zone of the EPL contained the secondary dendrites of middle tufted and a second class of mitral cells (Type II). The third (superficial EPL) zone was close to the edge of the glomerular layer and contained the secondary dendrites of external tufted cells. The few labeled internal tufted cells had secondary dendrites in either intermediate or deep zones. The extent of axonal branching was related to cell type. External tufted cells had extensive axon collaterals in the internal plexiform layer (IPL) and granule cell layer. The axonal branching of middle tufted and Type II mitral cells was less extensive. Type I mitral cells had little or no branching in the IPL. No cell types had axons re-entering the EPL. Our observations on the secondary dendrites are consistent with recent descriptions of the morphology of the output cells (Macrides & Schneider, JCN, 208: 419, '82; Kishi et al., *Neurosci. Lett.*, 28: 127, '82). These data coupled with the finding that separate granule cell populations innervate different zones of the EPL (Orona & Scott, *Neurosci. Abstr.*, #7.1, '82), indicate the need for a sublaminal analysis of the functional properties of the output cells.

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The Effect of Sucrose, Aspartame, and Vegetable Oil Rinses on the Recovery of Sweet Taste after Gymnemic Acid Treatment. JOSEPH PAGNOZZI and WILLIAM JAKINOVICH, JR. (Dept. of Biological Sciences, Lehman College, CUNY, Bronx, N.Y. 10468)

Gymnemic acid prevents the taste response to sweet stimuli. The purpose of this experiment was to investigate the mechanism by which gymnemic acid suppresses sweet taste in humans.

Four types of behavior experiments on human subjects were carried out. First, a two minute gymnemic acid solution (leaf extract) mouth wash was followed by one minute water rinses, and magnitude estimations of sucrose were made at 5 minute intervals. In three other experiments, aspartame, sucrose and vegetable oil solutions were used as rinses instead of water.

The recovery time for sweet taste was found to vary depending upon the rinse that was used. For example, the recovery time for sweet taste after water, aspartame and sucrose rinses were found to be the same. However, the recovery time was faster after vegetable oil rinses. This later finding supports the proposal that gymnemic acid is being removed from the lipids of the cell membrane by the vegetable oil.

However, the failure of the sweetener rinses, sucrose and aspartame, to increase the recovery time, seems to us to be non-supportive of the competitive inhibition model for gymnemic acid inhibition.

This work was supported by a grant from the National Institutes of Health.

The Size and Distribution Characteristics of Mitral Cells in the Olfactory Bulbs of Rats Exposed to Different Olfactory Environments. H. PANHUBER, D.G. LAING, E.A. PITTMAN and M.E. WILLCOX* (CSIRO Division of Food Research, and *CSIRO Division of Mathematics and Statistics, P.O. Box 52, North Ryde, Sydney, NSW, Australia 2113)

There is now a considerable amount of evidence that perikaryon size of some second and higher order sensory neurons is influenced by sensory input during development. While most of this evidence comes from experiments with the visual and auditory systems early work on olfaction has shown that odor specific changes occur in the mitral cells of the olfactory bulb following exposure of young rats to single odorants.

Using cell size as a criterion we have systematically mapped the size and distribution characteristics of mitral cells in the olfactory bulbs of rats reared in either a laboratory ('normal'), purified air or cyclohexanone environment. On analysis; the distribution of cell size for some 4% of mitral cells in the bulbs of 'normal' rats was found to be non-uniform. The pattern of the distribution of cell size was consistent from rat to rat and bulb to bulb. Comparison of the size distribution of 'normal' with deodorized air or cyclohexanone exposed animals shows that while this underlying 'normal' type pattern remains in the deodorized air group the cells are smaller overall. In the case of the cyclohexanone exposed group the underlying 'normal' pattern appears to be altered.

Ad Libitum Mixing to Preference for Salts in Broths and Sucrose in Lemonade, Compared to Hedonic Scaling. ROSE MARIE PANGBORN, KAREN S. BRADDOCK, and LEE J. STONE, (University of California, Davis).

EXP. I. Preferences were obtained from 20 students, and from 20 middle-aged adults to NaCl, MSG, glycylamide HCl (GLY), and KCl, singly and in combination, using ad libitum salting of chicken broth with (a) crystals in opaque salt shakers and with (b) high-concentration solutions. Responses were compared to graphic hedonic scaling of concentration series of each salt and salt mixture. Results indicated that:

- (1) Except for KCl, more salt was added using solutions than shakers, e.g., for NaCl, 3150 ± 2394 vs. 2546 ± 1862 ppm Na^+ , respectively.
- (2) The students demonstrated a good correlation between hedonic and ad lib. procedures except for KCl, which was described as bitter-salty, and GLY, which was considered metallic-sour. Older subjects showed a high correlation between the two procedures only for NaCl.
- (3) Older subjects added more salts in ad lib. mixing than did younger subjects, e.g., for NaCl, 3715 ± 2362 vs. 2351 ± 1619 ppm Na^+ , respectively.

EXP II. Preferences were obtained from 100 University students for NaCl in beef broth and sucrose in lemonade, using ad lib. mixing with high-concentration solutions, and scaling of hedonic responses to a concentration series on a 20-point, structured scale. The two methods were highly related, with those who added higher ad lib. amounts ascribing their maximum hedonic value to a higher concentration of NaCl or sucrose. Hedonic scaling resulted in three types of response distributions with concentration, ascending, descending, and inverted-U. Most subjects demonstrated similar hedonic distributions for the two media, that is, they added greater amounts of both NaCl and sucrose, or ascribed greater hedonic values to higher concentrations of the two.

Chemosensory Research Support: National Institute of Neurological and Communicative Disorders and Stroke. JACK PEARL (National Institute of Neurological and Communicative Disorders and Stroke).

Virtually all of the Institutes of the NIH support extramural research projects in the chemosensory area. Within the NIH the NINCDS is the major source of this support. Investigators in the chemosenses compete well for research support provided by the NINCDS. Over the past five years the rate of increase for the NINCDS funding of chemosensory research has been more than 3 times greater than the rate of increase for the NINCDS funding as a whole. The increase in chemosensory research support has been for both regular individual research and program projects. However, there has been little growth in the chemosenses for some of the areas of training. In the chemosensory area there is currently one Research Career Development Award and no Teacher Investigator Development Awards. The description of the chemosenses was based largely on the information provided by the NIH computer retrieval of information for scientific projects and abstracts from the Smithsonian Science Information Exchange. The computer retrieval system includes a classification of the research based on primary, secondary, and tertiary emphasis of descriptors of research, e.g., taste, taste threshold, taste disorders.

Functional Activity in the Rat Olfactory System In Utero. P.E. PEDERSEN, W.B. STEWART, C.A. GREER, and G.M. SHEPHERD (Secs. of Neuroanatomy, Neurosurgery, and Gross Anatomy, Yale University School of Medicine).

The vomeronasal system and the main olfactory system process odors concerned with sexual/social behaviors in adult rodent species. While the main olfactory system has been clearly demonstrated to be important throughout development, little is known about the functional importance of the vomeronasal system in neonates. This is despite the morphological maturity of receptors at birth and the early embryonic development of the accessory olfactory bulb, the first relay of the vomeronasal system. In the course of a developmental study of the rat olfactory system, we have used the 2-deoxyglucose (2DG) method to assess functional activity in main and accessory olfactory bulbs of near term fetal rats.

Sprague-Dawley pregnant rats were injected with 200 μ Ci/kg 14 C-2DG I.V. on Day 22 of gestation. One hour later, the dam was killed by decapitation. Each fetus was immediately delivered by Caesarean section, its brain was removed and was prepared for autoradiographic analysis.

Enhanced 2DG uptake appeared in the accessory bulb of 18 of 20 experimental animals from 4 different litters. Correlations of autoradiographs with histological sections indicated that maximal 2DG uptake occurred throughout the entire glomerular sheet of the accessory bulb. Foci of 2DG uptake were evident in the main bulb of only 4 animals. These were localized in the dorsocaudal portion of the main bulb.

These findings demonstrate that the 2DG technique can be used to investigate cerebral glucose metabolism in utero. Moreover, the high 2DG uptake in the accessory bulb suggests fetuses may use the vomeronasal system to detect their chemical milieu prenatally.

Research is supported by NIH Grants #NS16993 and #NS06978.

Ultraviolet Effects on Olfactory Receptor Response. JUDITH O. PRETELL (Dept. of Dermatology, Harvard Med. Sch.) JOHN S. KAUER (Secs. Neurosurgery & Neuroanat. Yale Med. Sch.)

Generation of an appropriate immune response requires both the recognition of antigen and the proper communication between cells of the immune system via specialized cell surface receptors. We have hypothesized that the surface membranes which confer chemical sensitivity on olfactory receptor neurons may be analogous to receptors on cells in the immune system. Since ultraviolet radiation (UV) has been shown to alter the turnover of several receptors on immune cells and thereby alters the immune response¹, we tested to see if UV might cause changes in the olfactory receptor response as well. The electro-olfactogram (EOG) from the nasal epithelium of the salamander (*Ambystoma tigrinum*) was used to examine changes in response after UV irradiation of the exposed mucosa. Experiments were carried out on 9 animals in which the ventral nasal mucosa was opened and EOGs were recorded in response to controlled odor stimulation with amyl acetate. The mucosa was UV irradiated with a source having a major peak at 254nm (16000 μ W/cm² at 8cm). EOG records were taken before, during, and after several short (1-2 min) pulses of UV separated by 7-15 min of no exposure. Peak EOG amplitudes were plotted as percent of control obtained before UV pulsing. The results are as follows: EOGs from a single site on the mucosa after UV showed a decrement of response to 10-20% of control with 2-5 min total UV exposure. EOGs from two sites on the same mucosa, where one site was shielded from UV, showed a decrement only at the unshielded site, but not at the shielded position. EOGs monitored for up to 36 hrs after UV showed a return to 60% or more of the control amplitude within 10-15 hrs. In summary, UV exposure of the salamander nasal mucosa causes a reversible decrement in the EOG. Toxic photoproducts, if formed, appear not to spread to unirradiated sites since EOGs from shielded portions of the same mucosa were unaffected. These experiments suggest that UV may reversibly change structures in the olfactory mucosa related to the generation of the EOG and therefore may be potentially useful in dissecting components of the initial response of receptors cells to odor.

¹Bach, F.H. et al. 1976. *Nature* 239:273. Supported by USPHS grant #NS-17275 to JSK.

Central Organization and Asymmetry of the Olfactory System in the Winter Flounder, *Pseudopleuronectes americanus*. P. D. PRASADA RAO and THOMAS E. FINGER (University of Colorado School of Medicine and Marine Biological Laboratory).

Flatfishes begin life as bilaterally symmetric animals but undergo a subsequent period of metamorphosis during which one eye migrates across the midline. Adult flatfishes also show a marked asymmetry in the peripheral olfactory apparatus (Yamamoto, 1982) with the upward facing organ being larger. Accordingly we undertook this study of winter flounder, *Pseudopleuronectes americanus*, to examine whether the central olfactory system maintains the peripheral asymmetry. In the adult winter flounder the right organ, being located on the upper side, is large (10 or 12 lamellae) while the left one is small (5 or 6 lamellae). The right olfactory bulb is approximately 3 times larger than the left, and the right telencephalon is 8% larger than the left. Horseradish peroxidase (HRP) and degeneration techniques were used to trace the central connections of the olfactory bulbs. Neurons afferent to the olfactory bulb occurred in the basal part of the preoptic region and bilaterally in 2 zones of the telencephalon and in the mesencephalic tegmentum. Each olfactory bulb projects bilaterally to restricted areas of the telencephalon, preoptic and tuberal areas with ipsilateral connections being heavier. Corresponding to the differences in the peripheral olfactory system, the central olfactory projections are also asymmetrical. The total volume of the terminal field formed by the right bulb (0.21 cm³) is approximately twice the volume of the terminal field formed by the left bulb (0.1 cm³). The right bulb projections constitute 2.6% of the volume of the ipsilateral telencephalon, whereas those of the left bulb occupy only 1.8% of the ipsilateral telencephalon. The contralateral projections of the right bulb occupy 1.9% of the telencephalic hemisphere, while those of the left occupy only 0.6% of the contralateral telencephalic hemisphere. The asymmetry in the projections of the right and left bulbs may be due to postmetamorphic differential growth of the olfactory system on the two sides. However, the difference in size of the telencephalic hemispheres cannot be attributed solely to the asymmetry in the primary olfactory targets, but may be due to differences in higher order olfactory projections or connections with other parts of the central nervous system.

Increased Taste Preferences and Decreased Neural Responses in Spontaneously Hypertensive (SHR) Rats. TIMOTHY W. PRIEHS, RUDY A. BERNARD AND DANIEL A. MAAS (Michigan State University, East Lansing, Michigan 48824-1101)

Contreras has shown that the enhanced NaCl preference of sodium deprived and adrenalectomized rats is accompanied by a specific decrease in gustatory neural activity. We, along with others, have previously reported that adult SHR rats show increased preference for NaCl and KCl when compared to Wistar Kyoto (WKY) controls. We did these experiments to examine the preferences of SHR rats for other taste stimuli and to determine whether changes in taste preferences are accompanied by changes in gustatory neural activity. Preferences were measured by the two-bottle choice method, in which each stimulus was presented for a four-day period and its position changed on a restricted random schedule. Ascending concentration series of NaCl, CaCl₂, HCl, QHCl and sucrose were presented to 12 SHR and 13 WKY rats. The SHR consistently displayed higher preferences than the WKY at almost every stimulus concentration except quinine. At the conclusion of the preference tests gustatory neural activity was measured from the same animals using whole nerve chorda tympani preparations. NaCl and KCl solutions were tested in half-log steps from .001 M to 1.0 M, and CaCl₂ from .001 M to .3 M. Additionally, .001 M and .003 M HCl, .001 M QHCl and .01 M and .1 M NH₄Cl were tested. The reference stimulus of 0.1 M NaCl was chosen because both groups showed almost equal preference for this, but not other, NaCl concentrations. The value of the initial peak discharge was used in all calculations. We found that the SHR had lower neural activity than the WKY for each stimulus tested. The decreased neural responsiveness of the SHR to NaCl is similar to that of Na deprived or adrenalectomized rats. In view of these and our other results, modification of gustatory nerve activity may be a general mechanism for modifying taste preferences.

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Retention of Odorants by the Human Nose, MICHAEL D. RABIN, J. ENRIQUE COMMETTO-MUNIZ* and WILLIAM S. CAIN (John B. Pierce Foundation Laboratory and Yale University, New Haven, CT).

We present a method to measure the retention of volatile substances by the human nose. In this technique, a vacuum source is used to draw odorized air into one nostril and out the other. The air passes from one nostril to the other via the choana and, upon exiting the nostril, passes into a gas sampling valve of a gas chromatograph. The technique permits quantitative analysis of deposition in the nose. Stimuli were five n-aliphatic alcohols ranging from ethyl alcohol to hexyl alcohol. Three subjects participated. The nose proved an extremely efficient filter, retaining an average of 98% of incoming mass. As could have been expected, higher alcohols were retained somewhat less efficiently than the lower alcohols. Percent retention ranged from 98.5% for ethyl alcohol to 95.2% for hexyl alcohol. Degree of retention seemed relatively independent of absolute concentration. The range of retention was surprisingly narrow in view of the infinite water, and hence mucus, solubility of ethyl alcohol and low water solubility of hexyl alcohol. One of the three subjects contributed most strongly to the decreased retention of the higher alcohols. His data departed from those of the other two subjects in a highly reliable fashion and indicated the existence of sizable individual differences. Such differences may represent differences in the internal geometry of the nose. Our method is now being used to measure the relation between rate of adaptation and adsorption by the nose.

*Fellow of the Consejo Nacional de Investigaciones Científicas y Técnicas, República Argentina, on leave from the Laboratorio de Investigaciones Sensoriales (CONICET-Fac. de Medicina, UBA), Buenos Aires, Argentina.

A Chemical Cue With Multiple Functions. D. RITTSCHOF, E.S. BRANSCOMB and J.D. COSTLOW (Duke University Marine Laboratory, Beaufort, NC 28516)

Balanoid barnacles are sessile, hermaphroditic, and non-self fertilizing. They are endowed with impressive sexual members enabling them to "reach out and touch someone" if that someone is relatively close. Barnacle cyprids, the larval settling stage, are known to use chemical cues in settling gregariously. Gregarious settlement improves reproductive success. Since the early 1960s it has been known that glycoproteins extracted from homogenates of adult barnacles (settlement factor) promote cyprid attachment and metamorphosis.

Barnacle drills are predatory snails that use chemical cues to locate barnacles in order to eat them. The molecules used by drills to locate barnacles are sticky peptides (3000 to 6000 Daltons). These peptides have been concentrated, purified and partially characterized from seawater bathing living intact barnacles (snail attractant). Snails detect attractant in solution.

Snail attractant peptides and settlement factor were compared in cyprid behavior and settlement assays. Responses of *Balanus amphitrite* cyprids to both preparations were similar. Settlement factor and snail attractant both evoked investigative behavior and facilitated attachment and metamorphosis. On a concentration basis snail attractant was approximately 1000-fold more effective than settlement factor in both assays. Metamorphosis was facilitated by snail attractant at concentrations approximately 1000-fold more dilute than literature reports of effective levels of purified settlement factor. In contrast to use of the cue by snails, modification of cyprid behavior and facilitation of metamorphosis appear to be mediated by contact phenomena. Cyprid behavioral and metamorphic responses are temporally separated and thus not rigidly linked. Both effects on cyprids however could be operating through a common sensory pathway. For barnacles it appears that the benefits of gregarious settling outweigh the risks.

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Olfactory Bulb Lesions and Weight Gain in Neonatal Rats. JUDITH M. RISSER and B.M. SLOTNICK (The American University)

Preliminary data on the effects of olfactory bulb lesions on growth of neonatal rats has been obtained. Six-day-old pups were individually anesthetized in an ice bath and subjected to complete, partial, or no aspiration of the olfactory bulbs. Twenty-four hours later pups were tested for nipple attachment behavior (Teicher & Blass, 1976) and then replaced with their dam. Pups were weighed daily and those that lost more than four grams within the first four postoperative days were sacrificed by perfusion with formalin. Remaining pups were sacrificed six days after surgery. Heads were decalcified, embedded in 10% gelatin, and cut in the horizontal plane on a freezing microtome. Sections were stained with thionine.

To date, results are available for three pups with removal of the olfactory bulbs and for eight pups with partial olfactory bulb lesions. Twelve sham operated pups served as controls.

Controls showed normal nipple attachment and gained weight on each day of the postoperative period. The olfactory bulbectomized pups failed to show nipple attachment and lost weight on each day after being returned to the dam. Most pups with partial lesions (lesions confined largely to the medial or lateral half of the olfactory bulbs) failed to show nipple attachment and lost weight for the first 2-3 postoperative days but then began to gain weight. Pups with medial lesions recovered more slowly than those with lateral lesions. Preliminary histological analysis indicates that recovery occurs independently of damage to the accessory olfactory bulb. Studies in progress include animals with discrete electrolytic lesions of the bulb.

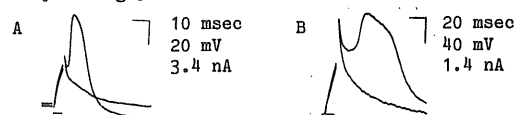
Taste Cells Generate Action Potentials. S. ROPER (University of Colorado Medical Center)

Until now, it has been held that the membrane properties of taste cells are passive; membrane resistance is ohmic and only graded receptor potentials are generated. I have reinvestigated the membrane properties of taste cells in the mudpuppy, *Necturus maculosus*, and have evidence that these earlier concepts may need revision.

Lingual epithelium was removed and stretched out in a chamber containing Ringer. Taste cells and their apical processes could be seen when viewed at 400X with Nomarski optics. Micropipettes were inserted into cells under direct visual control. [Ca] was elevated to 5 mM to improve the stability of recordings.

Resting potentials up to -90 mV were recorded from taste cells. Input resistances were obtained by injecting 20-100 msec current pulses through the recording electrode. The membrane properties from 11 taste cells in which resting potentials and input resistances were measured were -43±6 mV and 200±30 MOhms, respectively (means±s.e.m.).

More importantly, when taste cells were depolarized by injecting brief pulses of current through the recording electrode, impulses were evoked. Impulses were abolished by 1-5 µM tetrodotoxin (TTX), but were restored in the presence of TTX by adding 5 mM TEA:



Intracellular records from two taste cells. Superimposed traces show responses to threshold stimulation (below). A, in Ringer. B, Ringer + 5 µM TTX + 5 mM TEA.

These data suggest that impulses in taste cells from the mudpuppy have Na and Ca components. The presence of Na/Ca spikes may play a pivotal role in taste transduction and synaptic excitation of gustatory afferent terminals; influx of Ca in other cells is known to release transmitters and hormones.

Comparative Ultrastructure Of Olfactory Epithelia From Three Species Of Marine Fish. J. CARTER ROWLEY III and DAVID T. MORAN, Department of Anatomy, University of Colorado School of Medicine, Denver, CO 80262.

We have investigated the ultrastructure of olfactory epithelia of three species of marine fish by scanning and transmission electron microscopy. The Nassau grouper (*Epinephalus striatus*), the mangrove snapper (*Lutjanus griseus*), and the blue-striped grunt (*Haemulon sciurus*) are all equipped with olfactory rosettes--organs of olfaction located beneath each nostril. Each rosette is subdivided into lamellae, and each lamella is partially covered by olfactory epithelium. Whereas the olfactory rosette of the grouper is rather small, those of grunt and snapper are relatively large. Examination of the olfactory epithelium of each species by electron microscopy shows the sensory surfaces display significant differences in cellular composition and distribution, summarized below.

The Nassau Grouper: The olfactory epithelium of the Nassau grouper contains five morphologically distinct cell types: ciliated olfactory receptors, microvillar cells, ciliated epithelial cells, supporting cells, and basal cells. The ciliated olfactory receptor and the microvillar cells appear to be uniformly distributed throughout the sensory epithelium.

The Mangrove Snapper: As in the grouper, the olfactory epithelium of the snapper is composed of ciliated olfactory receptors, microvillar cells, ciliated epithelial cells, supporting cells, and basal cells. Although the ciliated receptors are uniformly distributed throughout the sensory epithelium, the microvillar cells are not. Microvillar cells are present at the tip of the lamella, but are absent from its base.

The Blue-Striped Grunt: The olfactory epithelium of the blue-striped grunt differs markedly in organization from that of the grouper and the snapper. The grunt has ciliated olfactory receptors, but appears to lack microvillar cells. In addition to ciliated epithelial cells, supporting cells, and basal cells, the olfactory epithelium of the grunt contains a large number of well-developed, uniformly distributed mucus-secreting goblet cells. Supported by NSF Research Grant No. BNS 82-10327.

Topographic Organization of Connections between the Main Olfactory Bulb and Pars Externa of the Anterior Olfactory Nucleus in the Hamster. T. A. SCHOENFELD AND F. MACRIDES (Worcester Foundation for Experimental Biology, Shrewsbury, MA 01545)

The centrifugal projections of pars externa (pE) terminate almost exclusively in the contralateral main olfactory bulb (MOB) and appear to be topographically organized (Davis & Macrides, JCN, 203:475-493, 1981). The present study characterized the bulbar connections of pE, using wheat germ agglutinin-HRP and TMB to visualize both retrograde and anterograde transport. Injections in restricted sectors of the MOB produced retrograde labeling of cells in restricted sectors of the contralateral pE. Likewise, injections in restricted sectors of pE produced restricted anterograde labeling in the contralateral MOB. While overlap existed, the lateral sector of pE was found to project preferentially to the rostral MOB; the more medially-situated dorsal and ventral sectors projected to dorsocaudal and ventrocaudal sectors of the MOB, respectively. Punctate injections in pE also produced retrograde labeling of mitral and tufted cells (including external tufted cells) in highly circumscribed sectors of the ipsilateral MOB that were roughly homotopic to the anterograde terminal labeling in the contralateral MOB. Because the great majority of external tufted cells do not project caudal to the retrobulbar area, this result represents a topographic organization of inputs to pE from these neurons. For mitral, internal tufted and middle tufted cells, the topographic retrograde labeling might have been accentuated by uptake in fibers of passage; however, this restricted labeling is inconsistent with a diffuse projection to pE from these MOB output neurons. Although it is unclear whether pE receives projections from MOB output neurons in addition to external tufted cells, we can conclude that both the afferent and efferent connections of pE with the MOB are topographically organized. This organization of rostral pathways linking the two main olfactory bulbs markedly contrasts with the lack of clear, sector-to-sector topography in the efferent and centrifugal afferent connections of the MOB with caudal cortical and subcortical areas.

This research was supported by NSF grant BNS 81-18767.

Evidence for a CO₂ Receptor in the Gill Spines

of the Horseshoe Crab, *Limulus polyphemus*. Jack Schlein (York College) and Stephen Lakatos (Hillcrest High School)

A small spine located on the inner lobe of each half gill of the horseshoe crab, *Limulus Polyphemus*, has previously been indicated as a chemoreceptor through a structural study (Schlein and Leonardi, 1981). The specific chemical modality to which the spines were sensitive remained unknown until the current study. A Y-shaped maze was constructed out of plexiglass and fitted with a controlled seawater pumping system. Crabs moving "upstream" from the base of the Y were faced with a clear choice at the confluence of the flow from the two arms. Initial trials with no stimuli present indicated no preference for right or left. The following stimuli were then presented: A) fresh water as opposed to seawater; B) O₂ pumped into one arm via an airstone as opposed to N₂ in the other. N₂ per se does not affect *Limulus* respiration (Waterman and Travis, 1953). C) CO₂ pumped into one arm as opposed to normal CO₂ in the other. In each case statistically significant preferences were seen. These trials were then repeated after removal of the gill spines. Preferences remained significant in the cases of the O₂ gradient and the salinity gradient but disappeared in the case of CO₂. A second group of crabs was run with and without spines and utilizing CO₂ as the stimulus choice. In this second group crabs were run alternately so as to avoid the possibility of a "patterning" preference developing. The results of this series of runs was similar to the first, a significant preference for the arm away from CO₂ with the spines intact and no significant preference with the spine removed. The results indicate that the gill spines provide an external chemoreceptor for CO₂ in the horseshoe crab as was hypothesized to exist by Hyde in 1898.

A Mutant Defective in Chemoresponse to and Binding of Folic Acid. STEPHANIE SCHULZ, MARIA DENARO, JUDITH VAN HOUTEN (Department of Zoology, University of Vermont, Burlington, VT 05405)*

Wild type *Paramecium tetraurelia* detect the soluble, essential nutrient folic acid, and swim to accumulate in it. Measures of instantaneous binding and initial rates of folate uptake suggest that there are saturable binding (Kd 65 uM) and uptake systems for this compound. We have previously established that measurable folate uptake is not necessary for chemoreception. We are now asking whether specific binding sites may function in *Paramecium* chemoreception of folate. In the present study, we use mutant *Paramecium* and inhibitors to perturb chemoresponse and binding. In contrast to normal cells, mutant d4-534 that cannot detect folate shows reduced, non-saturable binding of folate, perhaps representing the non-specific component of the total binding of folate to normal cells. To characterize the specificity of folate binding to normal cells, we identified compounds which compete with folate for chemoattraction and have determined whether they inhibit binding. Cyclic AMP, a strong, specific inhibitor of attraction to folate, appears to be a competitive inhibitor of binding. CAMP reduces binding of folate to the low, nonsaturable binding characteristic of mutant d4-534. When wild type binding data are corrected for non-specific binding using either the d4-534 values or values in the presence of CAMP, Scatchard and other analyses give an apparent Kd of 50 uM and a binding site concentration of 1.5 pmol/mg protein. Preliminary studies indicate that another mutant, mt9, has a decreased number of binding sites with a Kd similar to wild type. These studies support a role for specific folate binding in the chemosensory pathway in *Paramecium*.

*Supported by NSF # BNS 12176

Discrimination Inferences from Pleasantness Ratings of Taste Stimuli. R. GREGG SETTLE (Smell and Taste Center, University of Pennsylvania, Philadelphia, and VA Medical Center, Philadelphia)*

A major interest of our laboratory has been the development of clinical tests of taste dysfunction. A segment of our program has examined the potential utility of hedonic responses to make inferences about taste discrimination. Hedonic responses as a function of concentration can be classified into three types: monotonic increasing, monotonic decreasing, and concave functions. Using an Analysis of Variance model, variability in the data derived from pleasantness scaling tasks was partitioned into appropriate sources (concentration, presentation, and concentration by presentation effects). The concentration effect was decomposed into its orthogonal trend components (linear, quadratic, cubic, etc.). The linear component permits an estimate of the variability accounted for in ascending and descending functions and the quadratic component in concave functions. The proportion of variability accounted for by these two components provides an estimate of discriminability across concentrations. Our findings indicate that, for some stimuli (e.g., NaCl), a relatively large number of subjects (approximately one quarter) show better discrimination based upon pleasantness ratings than upon intensity ratings, indicating that hedonic measures provide useful information about discriminative capacity. In our clinic, plots of pleasantness functions have proved useful in determining the classification of borderline hypogeusics. The results of this work indicate pleasantness ratings can be used to evaluate the ability of subjects to discriminate among concentrations of tastants.

*Supported by grant NS 16365 from the National Institute of Neurological and Communicative Disorders and Stroke.

Exuberant Locus Coeruleus Input to Rat Olfactory Bulb. M.T. SHIPLEY (University of Cincinnati College of Medicine), F.R. HALLORAN and J. DE LA TORRE (Northwestern University Medical School)

The locus coeruleus (LC) is currently one of the most fashionable structures in the brain; it is situated in the dorsolateral pons and, in the rat, contains approximately 1600 neurons (Swanson, Br. Res '76). All of these neurons are presumed, on the basis of strongly convergent evidence, to be noradrenergic and numerous neuroanatomical studies have indicated that LC is the major and probably the sole source of noradrenergic innervation of the telencephalon. The LC has been implicated in functions ranging from arousal, motivation, and learning to sleep and possibly a guidance role in the development of connections.

Following injections of WGA-HRP into OB we were surprised to find intense retrograde labelling of sizable proportion of ipsilateral LC neurons and a few contralateral ones as well. Large OB injections labeled 450-500 LC neurons or about 30% of the total population; smaller injections labeled fewer cells. The number of LC neurons labelled was manifestly greater than following comparable injections in neocortical sensory fields (visual, auditory, somatic sensory, gustatory) or in insular, entorhinal or perirhinal cortex.

On the basis of these results it would appear that the OB is one of the major targets of LC neurons. It will be interesting to determine whether LC-OB neurons send collaterals to other cortical areas and to examine the organization of LC terminals and adrenergic receptors in the bulb.

How this exuberant LC-OB pathway fits into current theories of LC function is unclear; in any event, the OB should be an ideal structure for investigating the organization, function, development and plasticity of this fascinating central noradrenergic pathway.

Supported by NIH NS 19730, NINCDS 18490 and USAMRDC DAMD 17-82-C-2272.

Structure-Taste Relationship of Glutamyl Valine, the 'Sweet' Peptide for the Fleshfly. ICHIRO SHIMADA (Dep. Biol. Sci., Tohoku Univ., Kawauchi, Sendai 980, Japan), YUJI MAKI (Dep. Chem., Fac. Sci., Yamagata Univ., Yamagata 990, Japan)

L-Glutamyl-L-valine (Glu-Val) is one of the most stimulative dipeptides for the sugar receptor of the fleshfly. On the other hand, glutaryl-L-valine (Glt-Val) was almost ineffective. While N-acetylation of the α -amino group of the glutamyl (Glu) moiety in Glu-Val almost abolished responses, N-formylation of the group decreased the response appreciably but its effectiveness was clearly maintained. L- γ -O-Methyl-glutamyl-L-valine [Glu(γ -O Me)-Val] and L-glutamyl-L-valine (Gln-Val) still gave moderate excitation, while L-glutamyl-L-valine methyl ester resulted in extremely decreased responses. Valeryl-L-valine, having neither the α -amino nor the γ -carboxyl group of the Glu moiety, was naturally ineffective in stimulating the sugar receptor. These results suggested the presence of a specific accessory site for the Glu moiety in Glu-Val located close to the aliphatic carboxylate (T) site in the sugar receptor. Furthermore, the stimulating effect of optical isomers of glutamyl valine and its analogues was also examined and their structure-taste relationships will be discussed.

THE EFFECT OF CONCANAVALIN ON A RAT OLFACTORY MUCOSA: PREFERENTIAL INHIBITION OF THE RECEPTOR FOR SWEATY-SMELLING FATTY ACIDS.

Stephen Shirley, Ernest Polak & George Dodd.
Warwick Olfaction Research Group, University of Warwick UK.

We have previously reported that the lectin concanavalin A irreversibly depresses the EOG to some odorants but not to others. The selectivity of this inhibition has been examined using data from 60 odorants and we report the results here. For each odorant the EOGs have been measured in the presence of both Ringers Solution (u=5) and Ringers Solution containing concanavalin A at 0.5 mg/ml (n=5), using a superfused rat head preparation. This preparation was sufficiently stable for the experiments and superfusion of the olfactory mucosa with the various solutions did not alter the EOGs to 'control' odorants. The results are expressed as, R, the normalized EOG relative to the 'control' odorant, i-amyl acetate.

Odorant Type	N	Range R	N with R < 0.4
Acids	16	0.35 - 0.81	5
Aldehydes	9	0.60 - 0.96	0
Alcohols	13	0.71 - 1.02	0
Misc.	17	0.73 - 1.02	0
Thiols	2	-	0

The odorants in the various classes have comparable molecular properties other than the polar group. There is a preferential but not absolute inhibition of the signal to small, C_4 - C_6 acids. This effect can be inhibited by methyl-mannoside. The results are possible evidence for a distinct receptor system for sweaty-smelling fatty acids. The pattern of inhibitors of the acids resembles the pattern of anosmic thresholds to these odorants for humans.

Searobin Chemoreception: Responses from the Chemoreceptors on the Free Fin Rays. WAYNE L. SILVER (Monell Chem. Senses Ctr.) THOMAS E. FINGER (Univ. of Colorado Med. School)

The free fin rays of the searobin, *Prionotus carolinus* are innervated solely by spinal nerves and are sensitive to chemical stimuli despite the absence of gustatory or olfactory receptors. In this study we examined the sensitivity of these chemoreceptors by recording multi- and single unit responses from the spinal nerves innervating the fin rays. Responses to chemical stimuli were obtained from 26 of 44 animals. Squid extract (SE) elicited a vigorous response and of the 24 compounds in SE, 15 were tested and 10 were found to be stimulatory. Betaine (trimethylglycine), present in high concentrations in SE, was the most stimulatory compound tested and produced the lowest threshold ($10^{-6.2}$ M). Removal of the methyl groups of betaine led to an increase in threshold concentration and a decrease in relative stimulatory effectiveness. The order of relative effectiveness for stimulatory compounds at 0.01M was betaine > dimethylglycine > L-aba > sarcosine > L-ala > gly > L-cysh > trimethylamine > L-pro > L-leu > L-thr > L-ser > L-arg > L-phe > ammonium chloride. Nonstimulatory compounds at 0.01M included L-gln, L-glu, L-met, taurine, trimethylamine oxide, choline chloride, inosine, urea (0.1M), sucrose (1.0M), and acetic acid (0.2M). We conclude that the searobin's spinally innervated fin ray chemosense responds to relatively low concentrations of substances particularly prevalent in the animal's natural diet and thus may play an important role in its feeding behavior.

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Unilateral Odor Deprivation: Time-Course of Neurochemical Changes. L.K. SMITH, P.C. BRUNJES & R. MCCARTY. Dept. of Psychology, Univ. of Virginia, Charlottesville, Va. 22901

Odor deprivation induces significant reductions in olfactory bulb size. To date, neither the time-course of this change nor its developmental underpinnings have been studied. To understand the detailed effects of odor deprivation we are studying neurochemical changes in the olfactory bulb following unilateral nostril occlusion. This study examines deprivation-induced changes in the development of bulbar catecholamines. Littermate pairs of male or female Long-Evans rats had one nostril occluded on postnatal Day 1 (birth = Day 0) and were sacrificed at 4 or 8 days of age. A radioenzymatic thin-layer chromatographic assay was used to determine levels of dopamine (DA) and norepinephrine (NE). Differences in catecholamines between "deprived" and "nondeprived" olfactory bulbs in experimental rats were compared to levels found in control animals. At both ages controls showed identical catecholamine levels in the two bulbs. However, in the experimental animals there was a significant decrease in the DA content of the "deprived" olfactory bulb at Day 8. Preliminary data indicate a similar trend in the 4 day-old rats and additional data will be presented for this group as well as 12 and 30 day-old pups. These results suggest that the intrinsic DA-containing neurons of the olfactory bulb are rapidly affected by odor deprivation in early life. Although we found no difference in NE levels in experimental animals, preliminary studies show decreased AChE-staining in "deprived" olfactory bulbs within one week of occlusion. This suggests that odor deprivation differentially affects normal development of specific centrifugal afferents to the olfactory bulb.

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Effects of Olfactory Deprivation on Layers and Neural Populations in Mouse Olfactory Bulb. L. C. SKEEN, B. R. DUE, F. E. DOUGLAS (Department of Psychology and Institute for Neuroscience, University of Delaware).

Unilateral anosmia in neonate mice, induced by nasal cautery one day after birth (P1), results in a dramatic retardation in the development of the deprived main olfactory bulb (MOB). Volumetric analyses were conducted on P40 in order to quantify the relative degree of shrinkage in each layer of the deprived MOB. Microprojector drawings (75x) of Nissl-stained sections (24µm thick) through the deprived and experienced MOB's were entered into a microcomputer with a graphics tablet. Laminar volumes were then calculated using a modified version of the algorithm of Stefan *et al.* ('82). Results show a significant ($p < 0.01$), nonuniform reduction of the following layers of the deprived MOB: External plexiform layer (41%), internal plexiform layer (38%), granule cell layer (33%), mitral cell layer (27%), and glomerular layer (21%). Measures of cell density and nuclear diameter were obtained from plastic sections (4µm thick) stained with toluidine blue-O. These measures were then corrected with the methods of Hendry ('76) and Bayer ('82) to obtain population estimates for the deprived and experienced MOB's. Results show that mean tufted cell density is unchanged, while mitral cell density is increased by 37% in the deprived MOB. Further, whereas overall granule cell density is unchanged, analyses of granule cells partitioned into three subtypes--show a decreased density of dark granule cells (18%) and a concomitant increase in the density of neuroblasts (300%) in the deprived bulb. Population estimates from these data reveal a reduction in tufted cells (24%) in the deprived MOB; mitral cells are unaffected. There is a dramatic reduction in the number of dark granule cells in the deprived MOB (43%), while neuroblasts are increased by 194%. Olfactory deprivation therefore results in the loss of granule cells through either accelerated cell death or retarded cell proliferation, and appears to retard the differentiation of dark granule cells.

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Variability in the Attractivity of Different Natural Waters to Migrating Elvers of the American Eel. PETER W. SORESENSEN (Graduate School of Oceanography, University of Rhode Island, Narragansett, R.I. 02882)

Migrating larvae (elvers) of the American eel (*Anguilla rostrata*) are positively rheotactic in stream water. Because this response is not shown by olfactory-ablated animals, or evoked by well water, a chemical attractant(s) is believed responsible. The responses of recently collected elvers to 6 different natural waters and a variety of synthesized odorants were assayed in a Y-maze to determine the nature of scent recognition.

Cross-tests between elvers and their stream waters showed streams to vary in relative attractivity. While elvers never exhibited opposing preferences their behavior was often significantly different, suggesting that elvers acquire a mild preference for their own stream water. Dilution experiments indicated that a water's relative attractivity is determined by attractant concentration. Because water attractivity varied within river systems the attractive fraction is thought to be in a state of constant flux with the ecosystem. There was a negative correlation between water attractivity and salinity in an estuary. Migrating elvers are thought to employ both klinotaxis and rheotaxis to position themselves within attractive waters.

This work was supported by the National Marine Fisheries Service.

Investigations into the Origin of Fresh Water Odors Which Influence the Behavior of Migrating Elvers. PETER W. SORENSEN (Graduate School of Oceanography, University of Rhode Island, Narragansett, R.I. 02882)

Migrating larvae (elvers) of the American eel (*Anguilla rostrata*) possess a strong tendency to swim into flowing stream water. An odorant(s) characteristic of fresh water is held responsible. To determine the source of the attractant(s) a wide variety of items found in streams were collected and washed with well or deionized water. These washings were then assayed in a Y-maze against an appropriate blank.

Elvers exhibited 4 responses: a preference for the washing, a preference for the blank, no preference, and the tendency to avoid making any choice. Washings of 12 items were attractive. Among these were conspecifics, decaying leaf litter, particular plants, and 2 species of mature fish. Nonattractive leaves became attractive when cultured in a sterile nutrient media to which stream water had been added. Three items were repulsive, 9 had no influence, and 9 inhibited a choice. Washing concentration affected the nature of the elvers' response. Elvers were thought to responding to a bouquet of odors with animal, plant, and microbial origins.

This work was supported by the National Marine Fisheries Service.

Food and Fluid Intake Patterns in Rats as a Function of Sucrose Concentration. ALAN C. SPECTOR (Florida State University), JAMES C. SMITH (Florida State University)

It is well established that multiple factors control the ingestion of sugar solutions in rats. Oropharyngeal sensations such as taste, smell, texture, and temperature play an important role along with postingestional mechanisms. Since various concentrations of a sugar solution differ in taste, texture, calories, and rates of absorption, it is no surprise to find that total consumption of sugar solution in 24-hour periods as a function of concentration is not a linear phenomenon. Much of our knowledge on sugar consumption in the rat is derived from total intake measures. However, not much is known about the patterns of consumption which account for differences in total intake. The present study represents an initial attempt to describe these patterns. Our laboratory has developed a computerized system for monitoring the food and fluid intake of rats. This system records the number of licks on each of two bottles (per rat) and time spent eating during consecutive 30 second intervals throughout a 24-hour period. A graphical representation of the 24-hour intake pattern can be obtained. In the present experiment, 5 rats were presented with water, chow, and a sucrose solution during 23-hour periods. All animals received each of four sucrose concentrations: 0.1M, 0.25M, 0.5M, and 1M.

As has been previously shown from total intake measures:

- 1) Calories consumed from the sugar solution increased with increasing concentrations approaching asymptote at 0.5M.
- 2) Food intake decreased as calories consumed from sucrose increased.

From examining the pattern data it was found that:

- 1) The bout frequency for both sucrose and food decreased as the concentration of sucrose solution increased.
- 2) The sucrose bout size increased to a peak at 0.5M.
- 3) The sucrose bout duration increased to a broad peak spanning 0.25M-0.5M.
- 4) When water was the only fluid available drinking occurred primarily during the dark phase of the L/D cycle.

However, as the concentration was increased sucrose consumption became relatively uniform over a 23-hour period.

Comparison of Peripheral and Brainstem Excitatory Responses to Chemical Stimulation of the Cat Epiglottis. H.M. STEEDMAN (University of Michigan, Ann Arbor, MI 48109), R.M. BRADLEY and C.M. MISTRETTA (University of Michigan).

We previously reported responses from the superior laryngeal nerve (SLN) to chemical stimulation of the cat epiglottis. Now we present data on brainstem responses to the same chemical stimuli, to compare the responses from primary and secondary neurons supplying the epiglottis.

Single unit responses were recorded from 30 adult cats anesthetized with sodium pentobarbital. The stimulation area was limited to the epiglottis, larynx and part of the pharynx. Stimuli were: 0.5 M KCl, NH_4Cl , NaCl and LiCl, 0.01 N HCl, 0.005 M citric acid and distilled water. Solvent and rinse for these chemicals was 0.154M NaCl solution.

Mean peak response frequencies (imp/sec) in excess of spontaneous activity and the percent of units responding to each chemical were calculated for 41 medullary and 31 SLN recordings:

	KCl	NH_4Cl	NaCl	LiCl	H_2O	HCl	cit	spont.
Medulla	21±16	21±23	12±8	11±7	20±20	21±21	13±9	3±5
%	98	88	10	5	51	78	32	
SLN	42±24	31±19	8±0	0	21±11	37±25	27±17	1±2
%	100	52	3	0	32	58	35	

Generally, a higher proportion of brainstem neurons responded to each chemical stimulus. As in the periphery, KCl, NH_4Cl and HCl were the most effective stimuli while NaCl and LiCl were least effective. However, spontaneous activity rates were higher in the brainstem, whereas peak response frequencies for some chemicals were lower. This latter observation suggests that there are excitatory and inhibitory influences on the brainstem neurons.

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Saccharin Preference and Aversion in Streptozotocin-Diabetic Rats.

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Saccharin has been characterized as having a dual taste in the rat gustatory system, one component being sweet or glucose-like and the other bitter, or quinine-like. Rats with diabetes mellitus induced by alloxan have been reported by several investigators to reject or show a markedly diminished preference towards saccharin. This aversion has been attributed to both a conditioned taste aversion associated with alloxan-induced illness as well as to a decreased sensitivity to the sweet component of the dual taste.

In the present experiments it is demonstrated that previous exposure of rats to saccharin reduces saccharin aversion in streptozotocin induced diabetes. In addition, with prior exposure to the substance diabetic animals display saccharin preference functions over a wide range (.001 to 5.0%) which are indistinguishable from those of normal animals. While the dual taste quality of saccharin is not questioned, the rejection of the substance by diabetic animals may be as likely due to the novelty of the "bitter" component as to a decreased sensitivity to the "sweet" component.

Specificity of the Gerbil's Sweet Taste Inhibitor, Methyl 4,6-Dichloro-4,6-Dideoxy- α -D-Galactopyranoside. VASILIKI VLAHOPOULOS and WILLIAM JAKINOVICH, JR. (Dept. of Biological Sciences, Lehman College, CUNY, Bronx, N.Y. 10468)

The electrophysiological taste responses to sucrose and saccharin are inhibited by the sugar, methyl 4,6-dichloro-4,6-dideoxy- α -D-galactopyranoside (Dicl-gal). To understand the nature of this inhibition, electrophysiological experiments were conducted, comparing the gerbil's chorda tympani nerve responses to sucrose with responses to mixtures of sucrose and Dicl-gal derivatives.

The results of these experiments suggest a direct correlation between inhibition and the structure of the particular compound. In this vein we have observed:

1. Inhibition requires the presence of a methyl group at the C-1 position.
2. Furthermore, the orientation of that methyl group is important, axial being favored over equatorial.
3. Likewise, the orientation of the chlorine atom at the C-4 position is essential for inhibition, again, axial being favored over equatorial.
4. Our data also indicate that both chlorine atoms, at positions C-4 and C-6 are required for inhibition.
5. In addition, the sequential arrangement of the chlorine atoms was found necessary for inhibition.
6. Substitution of the two chlorine atoms by a large, hydrophobic benzylidene group caused no inhibition at all.

In summary, Dicl-gal, which possesses an axial methoxy group at C-1 position, and two chlorine atoms at axial position C-4, and equatorial C-6, appears to be the most potent inhibitor of the sucrose electrophysiological response in the gerbil. Further research is planned along the lines of structure-functional relationships.

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Morphology of Taste Afferents in the Solitary Nucleus of the Hamster. MARK C. WHITEHEAD (Dept. of Oral Biology, Univ. of Connecticut Health Center, Farmington, CT 06032)*

The solitary nucleus (SN) is the first level of the gustatory system where processing of taste information can occur. A structural basis for that processing was investigated. Labeling of the chorda tympani with horseradish peroxidase resulted in Golgi-like filling of central taste afferents. Individual afferents, their sizes, branching pattern and synaptic endings were studied with light and electron microscopy.

Taste axons in the solitary tract (ST), range greatly in size; from 0.2-1.5 μ m in diameter. Similarly, labeled neuronal somata in the geniculate ganglia range from 10-30 μ m in diameter. Thick fibers are restricted to the rostral half of the SN; fine fibers only are found caudally. The axons, as they descend in the ST, send multiple collaterals, at intervals of about 100 μ m, medially into the nucleus. Thus single axons project to somewhat divergent rostral-caudal levels. The collaterals, in the SN, branch and bear numerous small swellings which are confined to areas of neuropil. The swellings correspond in electron micrographs to labeled synaptic endings located in centers of glomeruli. These endings are ovoid or scalloped, indented by structures which surround them. They contain large, round vesicles and synapse, by means of symmetrical or asymmetrical contacts, on small dendrites and spines. Unlabeled endings, surrounding the labeled ones in the glomeruli, contact the dendrites receiving taste afferent input or the taste endings themselves; some resemble presynaptic dendrites, others are axonal endings packed with small, pleomorphic vesicles.

Chorda tympani afferents are sensitive to particular taste qualities (Frank, '73). Their divergent projections suggest that neurons of the SN responsive to certain tastants may be broadly distributed. The functional characteristics of the different sized primary afferents are not known although thickness, conduction velocity and taste sensitivity may be correlated. The synaptic milieu of the taste endings allows for the possibility of modulation of taste elicited activity in afferent endings or second-order neurons by other inputs.

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Olfactory Deficits in Cystic Fibrosis: Distribution and Severity. JAMES M. WEIFFENBACH (National Institute of Dental Research), VINCENT P. MCCARTHY, (National Institute of Arthritis, Diabetes, and Digestive and Kidney Diseases)

The reported association of cystic fibrosis (CF) with heightened chemoreceptive sensitivity has been challenged by evidence of an olfactory deficit in CF patients. The challenge, however, rests solely on tests with a single odorant. To provide additional definition of olfactory function in CF, thresholds for another odorant were obtained and performance on a multiple choice olfactory recognition test assessed.

The median threshold of twenty CF patients for detection of the floral odorant PEMEC was higher and significantly different from that of fifteen unaffected control subjects. Three patients displayed thresholds markedly higher than those of any other participants. These three patients were also the only participants to perform at chance level on the fifty multiple choice items of the recognition test. Whereas individual scores identified anosmic patients, median scores failed to discriminate the patient from the control group.

The overall deficit in threshold performance of the patients in the present study reinforces an earlier challenge to the reported hypersensitivity of CF patients to chemosensory stimuli. In addition, the present study suggests that the olfactory deficit is specific to a subgroup of patients and that it is general or complete in affected individuals.

Cyclic nucleotides and Sugar Reception in the Fly. HELMUT WIECZOREK (Zoological Institute, University of Regensburg, FRG)

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The involvement of cyclic nucleotides in sensory transduction has been proved so far only for photoreceptors of vertebrates. For taste receptors of insects there exist solely two contradictory electrophysiological studies: Daley and Vande Berg (BBA 437, 211, 1976) reported that the phosphodiesterase (PDE) inhibitor aminophylline as well as cAMP and dibutyryl cAMP inhibit, and that dibutyryl cGMP (but not cGMP) enhances the response of the fly's labellar sugar receptor. I could confirm only the effect of aminophylline. However, I found no influence of cAMP, dibutyryl cAMP, and cGMP; dibutyryl cGMP inhibits the response of the sugar receptor (cited by Hansen and Wieczorek, in: Biochemistry of Taste and Olfaction, ed. by R.H. Cagan, p.139, 1981). In recent electrophysiological experiments I detected that theophylline inhibits the response of the sugar receptor already within the first 10 ms after onset of stimulation. The inhibition increases continuously during the one-second application. Its time course can easily be interpreted if permeation of theophylline across the membrane and intra-dendritic inhibition of cGMP-specific phosphodiesterases is assumed. This hypothesis is under investigation. Biochemical experiments led to the following findings. Specific guanylate cyclase activity as well as the concentration of cGMP are 10-fold higher in sensilla-rich labella than in proximal, sensilla-poor parts of the fly's proboscis (haustella). On the other hand the specific activity of adenylate cyclase is 3-fold higher in the haustella, and cAMP concentrations are equal in labella and haustella.

Mating behavior in male hamsters is dependent on chemosensory input from olfactory and vomeronasal systems. While olfactory bulb (OB) removal eliminates mating behavior (Murphy & Schneider, 1970), vomeronasal nerve (VNN) section eliminates it in 40% of the animals (Winans & Powers, 1977). Both treatments disrupt the Nervus Terminalis (NT) whose function in mating or other behaviors is not known. Early anatomists described the NT in embryo and adult vertebrates from fish to man, but the lack of a selective stain made it difficult to trace its central fibers. LHRH (Schwanzel-Fukuda, 1980) and AChE (author), however, have recently been found in NT cells and fibers; the presence of LHRH along with NT fibers in the medial preoptic nucleus hint to a sex related role. In order to determine the anatomical relationship between the NT and the other two chemosensory systems, I have mapped the distribution of Ganglion terminale (GT) cells from the rostral pole of the OB to the nerve's entrance into the ventral forebrain. Five day old hamsters were perfused with fixative, and heads were stained *en bloc* with the Tsuji method of demonstrating AChE, embedded in paraffin and sectioned (10 μ). Because the *en bloc* procedure did not stain cells at the most caudal end of the nerve, several heads were frozen-sectioned (80 μ) and stained. AChE-containing GT cells were counted and/or mapped in every section. GT cells begin at the ventromedial border of the OB about 0.3 mm caudal to the rostral pole of the OB. Proceeding caudally, the cells cluster around and in the VNN for 0.7 mm. The two nerves then diverge, the VNN travelling dorsally toward the accessory OB and the NT descending to the ventral surface of the forebrain. At the level of the rostral hippocampal rudiment (HR), the GT cells merge with the molecular layer of the HR and disappear shortly thereafter. The histochemical identification of the GT cells combined with the immunocytochemical localization of LHRH should enable the further tracing of the NT's central connections as well as facilitate the study of its function.

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The complaint of olfactory dysfunction has proven to be somewhat vague in its description. One explanation for the apparent lack of an adequate definition of the olfactory complaint, beyond the anecdotal level, seems to be an incomplete consensus on physiological models which can account for the psychophysical perceptual process of odor identification. One difficulty is the multitude of individual odorants which can be discriminated from one another and identified in isolation as being unique. Another difficulty is that odor identification does not appear entirely dependent upon the olfactory nerve alone, but also includes the trigeminal as well as possibly the glossopharyngeal and vagus. At the psychophysical level, we must, at least for the moment assume that the perceptual process of odor identification, and hence the complaint of olfactory dysfunction, depends upon the relative contribution of all these physiological inputs. The consequent psychophysical problem is to infer how disruption of these inputs affect the perception of odorants.

Ten odorants were administered with the sniff-bottle technique in individualized random order to 20 normal subjects (10 male, 10 female) in a closed set and the identification responses subjected to a confusion matrix analysis. The percent correct among the 10 odorants for these normal subjects was greater than 90%. This result was predicted from a previous study with 50 subjects on the presumed identifiability of these 10 odorants from a group of 107. Subjects who had a history of exposure to toxic substances were then evaluated and found to have decreased performances which were dose related and further influenced by individual differences. A patient with Kallmann's Syndrome was also examined. The results suggested the type of confusions to be expected in the identification of odors when input from the olfactory nerve is not present. Such results illustrate the feasibility and practicality of confusion matrix analyses in further refining and defining the complaint of olfactory dysfunction.

It is known that mice can distinguish the H-2 haplotypes of other mice by scent, and that urine is a potent source of the characteristic odors governed by H-2. It is now shown that genetic disparity confined to the Qa:Tla region of chromosome 17 adjacent to H-2 is accompanied by distinctive urinary scents which can be distinguished by trained mice in a Y maze. This was confirmed by the transfer of training procedure in which mice trained to distinguish the urine of B6 mice from the urine of recombinant congenic B6-Tla^a mice successfully distinguished the urines of Tla^a and Tla^b homozygous F₂ segregants of the cross B6 x B6-Tla^a in the absence of reward, including blind trials with coded urine samples. It is also shown that genetic disparity confined to the K region of H-2 is accompanied by distinctive urinary scents recognized in the Y maze. Thus mice trained to distinguish the urines of B10 from B10.A congenic mice successfully distinguished various combinations of B10, B10.A and B10.A(2R, 3R, 5R and 18R) recombinant congenic mice representing genetic differences limited to the H-2K region. It is suggested that the individual scents of mice comprise several odorous components and may be incidental to quantitative or qualitative metabolic variations arising from polymorphism of genes such as H-2.