Program
and
Abstracts

The 9th Annual Meeting
of the
Association for Chemoreception Science
April 29 - May 3, 1987
Sarasota, Florida

The Association for Chemoreception Sciences acknowledges the generous, continuing financial support provided by the following corporate members:

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PROGRAM FOR THE 9TH ANNUAL MEETING OF THE ASSOCIATION FOR CHEMORECEPTION SCIENCES HYATT SARASOTA - SARASOTA, FLORIDA

SCHEDULE OF EVENTS

WEDNESDAY, AF	RIL 29	4:00 p.m.	SPECIAL INTEREST SCIENCE AND BEER WORKSHOP I - Florida Room
4:00 p.m. to 11:00 p.m.	REGISTRATION - Gallery		"What Can Activity Mapping Methods Tell
7:30 p.m.	OPENING REMARKS AND AWARDS PRESENTATION - Hernando Desoto N & S		Us About Coding of Olfactory Information: "
	Dr. Bruce Oakley, Executive Chairperson, AChemS		Dr. John S. Kauer, Tufts-New England Medical Center, Moderator Open discussion format, with
8:00 p.m.	GIVAUDAN LECTURE - Hernando Desoto N & S		established investigators in the field included in the audience.
	"Overview of Ion Channels"	6:00 p.m.	DINNER BREAK
	Dr. Charles F. Stevens, MD, Ph.D. Yale University	8:00 p.m. to 11:00 p.m.	POSTER SESSION I- Sara Desoto N & S
	Introduction by Dr. Vincent Dionne, Univ. California, San Diego	8:00 p.m. to 8:30 p.m.	"The World of Umami"
9:00 p.m. to 11:00 p.m.	WELCOME RECEPTION - Gallery Cash bar		Introduction by Dr. Kenzo Kurihara, Hokkaido University
THURSDAY, APR	IL 30	FRIDAY, MAY 1	
	REGISTRATION - Gallery	7:30 a.m.	CONTINENTAL BREAKFAST - Gallery
1:00 p.m. 7:30 a.m.	CONTINENTAL BREAKFAST - Gallery	8:00 a.m.	SLIDE SESSION IIA - Hernando Desoto N SLIDE SESSION IIB - Hernando Desoto S
	•	10:00 a.m.	COFFEE BREAK - Gallery
8:00 a.m. 9:30 a.m.	SLIDE SESSION I - Hernando Desoto N & S COFFEE BREAK - Gailery	10:30 a.m.	SLIDE SESSION IIIA - Hernando Desoto N
10:00 a.m.	SYMPOSIUM I - Hernando Desoto N & S	10.00	SLIDE SESSION IIIB - Hernando Desoto S
10.00 2.00	"Olfactory Function in Alzheimer's	12:30 p.m.	Beach shuttle runs 1 p.m 6 p.m., departing from upper lobby, entrance
	Disease and Related Dementias"	1:00 p.m.	CHEMICAL SENSES EDITORIAL BOARD,
•	Chaired by Dr. Richard L. Doty, Univ. of Pennsylvania		LUNCHEON MEETING - State Room
	OLFACTORY SENSORY CHANGES IN ALZHEIMER'S AND RELATED DISEASES	4:00 p.m.	SPECIAL INTEREST SCIENCE AND BEER WORKSHOP II - Florida Room
	Dr. Richard L. Doty Univ. of Pennsylvania		"Early Events in Chemosensory Transduction"
	NEUROPATHOLOGICAL ALTERATIONS IN OLFACTORY PATHWAYS OF ALZHEIMER'S DISEASED PATIENTS	•	Dr. Vincent E. Dionne, U.C. San Diego, Moderator Open discussion format, with
	Dr. Patricio Reyes Jefferson Medical College	-	established investigators in the field included in the audience.
	TRANSNASAL, TRANSNEURONAL TRANSPORT OF MACROMOLECULES: IMPLICATIONS FOR	6:00 p.m.	DINNER BREAK
	ALZHEIMER'S AND RELATED DISEASES Dr. Michael P. Shipley Univ. of Cincinnati	8:00 p.m. to 11:00 p.m.	POSTER SESSION II - Sara Desoto N & S
	MOLECULAR APPROACH TO MARKERS FOR ALZHEIMER'S AND RELATED DISEASES Dr. Kenneth Kosik Harvard Medical School	8:00 p.m. to 8:30 p.m.	MOVIE - Four Flags Room "The World of Umami" Introduction by Dr. Kenzo Kurihara, Hokkaido University
	OLFACTORY IMPAIRMENT IN FOCAL BRAIN DISEASE	SATURDAY, May	2
	Dr. Robert Mair Univ. of New Hampshire	7:30 a.m.	CONTINENTAL BREAKFAST - Gailery
1:00 p.m.	LUNCH BREAK	8:00 a.m.	SLIDE SESSION IV - Hernando Desoto N & S

9:30 a.m.

COFFEE BREAK - Gallery

Beach shuttle runs 1 p.m. - 6 p.m., departing from upper lobby, entrance

10:00 a.m. SYMPOSIUM II - Hernando Desoto N & S

"Perception of Complex Mixtures in Odor and Taste"

Chaired by Dr. David G. Laing, CSIRO, Sydney

NATURE OF COMPLEX STIMULI Dr. David G. Laing CSIRO, Sydney

MIXTURES AND OLFACTORY PHYSIOLOGY Dr. Charles D. Derby Georgia State Univ.

MIXTURES AND TASTE PHYSIOLOGY Dr.David V. Smith Univ. of Cincinnati

MIXTURES AND TASTE PSYCHOPHYSICS Dr. Robert L. McBride CSIRO, Syndey

MIXTURES AND OLFACTORY PSYCHOPHYSICS Dr. William S. Cain Yale University

1:00 p.m. LUNCH BREAK
Beach shuttle r

Beach shuttle runs 1 p.m. - 6 p.m., departing from upper lobby, entrance

BUS TRIP TO MYAKKA RIVER STATE PARK Box lunch and beverage provided Open to those holding tickets purchased at Registration

1:00 p.m. ACHEMS EXECUTIVE COMMITTEE, LUNCHEON MEETING - State Room

5:00 p.m. GENERAL BUSINESS MEETING - Hernando
Desoto S
All AChemS members encouraged to
attend

6:00 p.m. DINNER BREAK

8:00 p.m. to POSTER SESSION III - Sara Desoto N & S 11:00 p.m.

SUNDAY, MAY 3

7:30 a.m. CONTINENTAL BREAKFAST - Gallery

8:00 a.m. SLIDE SESSION VA - Hernando Desoto N SLIDE SESSION VB - Hernando Desoto S

10:00 a.m. COFFEE BREAK - Gallery

10:30 a.m. SLIDE SESSION VIA - Hernando Desoto N SLIDE SESSION VIB - Hernando Desoto S

12:30 p.m. MEETING ADJOURNS

VOLUNTEER PAPER SESSIONS

THURSDAY, AP	RIL 30 - Slide Presentations	P6 - 12	B DYATE OVERHALE THE TAXABLE TO THE
	"CLINICAL ASPECTS OF SMELL AND TASTE"	P6 - 12	R. BLAIR SKINNER, FRED E. ROSE, MAGDALENA M. JENSEN, CLAIRE MURPHY (San Diego State University) AND WILLIAM S. CAIN (Pierce Foundation, Yale
Slide Session Chaired by Dr Center	n I - Hernando Desoto N & S r. D. V. Smith, Univ. Cincinnati Medical		University) Age-related Differences in Short Term Memory for Olfactory and Visual Stimuli.
8:00 - 1*	DAVID V. SMITH, ROBERT A. FRANK. MYLES L. PENSAK AND ALLEN M. SEIDEN (Taste and Smell Center, University of Cincinnati Medical Center) Characteristics of Chemosensory Patients and a Comparison of Olfactory Assessment Procedures.	P7 - 13	MAGDALENA M. JENSEN (San Diego State University), AND CLAIRE MURPHY (San Diego State University) Suprathreshold Evidence for Quality-Specific Aging in the Taste System.
8:15 - 2	DANIEL A. DEEMS AND RICHARD L. DOTY (Smell and Taste Center, University of Pennsylvania) Odor Detection Ability Decreases With Age.	. P8 - 14	BRUCE W. JAFEK, M.D., PAMELA M. ELLER, BARBARA A. ESSES, M.D., AND DAVID T. MORAN, PH.D. (University of Colorado School of Medicine) Post-Traumatic Anosmia: Ultrastructural Correlates.
8:30 - 3	J. WEIFFENBACH (National Institute of Dental Research) AND E. KOSS (National Institute on Aging. Isolated Deficit for Recognition of Airborne Chemosensory Stimuli in Early Alzheimer's Disease.	P9 - 15	BARBARA A. ESSES, M.D., BRUCE W. JAFEK, M.D., DANIEL J. HOMMEL, M.D., AND PAMELA M ELLER (University of Colorado Schooi of Medicine) Histological and Ultrastructural Changes of the Taste Bud
8: 45 - 4	RICHARD L. DOTY AND MARK FERGUSON-SEGALL (Smell and Taste Center, University of Pennsylvania) Odor Detection Performance of Rats Following d- Amphetamine Treatment: A Signal Detection Analysis.	P10 - 16	Following Radiation: A Pilot Study. WILLIAM S. CAIN, LAI-CHUE SEE, AND TARIK TOSUN (John B. Pierce Foundation Laboratory and Yale University) Charting Odor and Irritation of Formaldehyde at Environmentally
9:00 - 5	L. BARTOSHUK (J. B. Pierce Foundation Laboratory), M. FRANK, T. HETTINGER (University of Connecticut), C. PETERSON, V. RAMIREZ AND J. RODIN (Yale University) Effects of HCL on Taste: Taste and Bulimia.	P11 - 17	Realistic Concentrations. FEDERICO GONZALES AND ALBERT I. FARBMAN (Dept. of Neurobioloty, Northwestern Univ., Evanston, IL 60201) The Effect of an Antidepressant Drug, Amitriptyline, on Growth of Axons from
9:15 - 6	R. G. SETTLE, J. D. AMSTERDAM AND E. ABELMAN (Smell and Taste Center and Depression Research Unit, University of Pennsylvania) Sucrose Taste Perception in Depressed Patients.	P12 - 18	Olfactory and CNS Neurons in Explant and Cell Cultures. CECILE S. ROSE, PETER G. HEYWOOD, AND RICHARD M. COSTANZO (Medical College of Virginia, Richmond, VA 23298) Impairment of Olfactory Function in
	abstract number		Workers Chronically Exposed to Cadmium Fumes.
	IL 30 - Poster Presentations		NEADIV EVENES. PROGUENTAROV AND PROGUENT
Poster Session	n I - Sara Desoto N & S	P13 - 19	"EARLY EVENTS: BIOCHEMISTRY AND BIOPHYSICS"
P1 - 7 P2 - 8	"CLINICAL AND INDUSTRIAL" LISA M. PIKE, MELVIN P. ENNS AND DAVID E. HORNUNG (St. Lawrence University, Canton, NY) Intensity Effects on the Odor of the Enantiomers of Carvone. H. N. WRIGHT (Clinical Olfactory		ALBERT I. FARBMAN, LYNN M. COLLETTI (Dept. of Neurobiology, Northwestern Univ., Evanston, IL 60201) GORAN HELLEKANT, R. ALBRECHT AND H. VAN DER WEL (Dept. of Veterinary Sciences, Univ. of Wisconsin, Madison, WI 53706) A Possible Role for Call in turnover of taste cell membrane binding sites for
	Research Center and Department of Otolaryngology, State University of New York Health Science Center at Syracuse) Odorant Identification as a Function of Concentration.	P14 - 20	the sweet protein, thaumatin. R. DINO RULLI AND RICHARD C. BRUCH (Monell Chemical Senses Center) Ligand Binding Specificity of the Olfactory L-Alanine Receptor of Channel Catfish.
•	ANNETTE Z. DREITH (Dept. of Nutrition, Univ. of California, Davis, CA 95616) Calcium Appetite in Dahl Rat Strains Differing in Susceptibility to Low Renin Hypertension.	P15 - 21	M. LEVANDOWSKY AND DELMA BRATWOLD (Haskins Labs of Pace University, NY., NY 10038) Calmodulin Blockers and Chemosensory Responses in the Cilate, Tetrahymena.
	CLAIRE MURPHY AND REBECCA M. NERISON (San Diego State University) Odor Memory Decrements in Alzheimer ¹ s Disease.	P16 - 22	KUMIKO SUGIMOTO (Monell Chemical Senses Center) AND JOHN TEETER (Monell Chemical Senses Center and University of Pennsylvania) Voltage-Dependent Ionic
	ROBERT J. CONTRERAS (University of Alabama at Birmingham, Department of Psychology, Birmingham 35294) Increased	D17 00	Currents in Isolated Taste Receptor Cells of the Tiger Salamander.
	Cardiovasuclar Reactivity in Adult Sprague-Dawley Rats Exposed Perinatally to a High Salt Diet.	P17 - 23	A. C. KOEHNE (University of Wurzburg, FRG) J. DUBNOW (SUNY, Stony Brook) AND G. D. PRESTWICH, (SUNY, Stony Brook) The Molecular Ontogeny of Pheromone Reception in the Gypsy Moth, Lymantria dispar.

P18 - 24	J. MICHAEL SASNER AND JUDITH VAN HOUTEN (Univ. of Vermont) Use of Covalently Crosslinked Ligand and Anti-ligand Antibody to Identify Chemoreceptors in Paramecium.	P31 - 37	ALAN GRANT, ROBERT O'CONNELL (The Worcester Found. for Exp. Biology, Shrewsbury, MA 01545) AND THOMAS EISNER (Sec Neurobiology and Behavior, Corneil University, Ithaca, NY 14853)
P19 - 25	MICHAEL GAGNON AND MARK WRIGHT (Univ. of Vermont) Cyclic AMP and Chemorecaption in Paramecium.		Neurophysiological Responses in Female Utetheisa ornatrix (Lepidoptera: Arctiidae) to a Male Produced Pheromone.
P20 - 26	MICHAEL S. LIDOW, ALBERT I. FARBMAN (Dept. of Neurobiology & Physiology, Northwestern University, Evanston, IL 60201) JAMES I. MORGAN AND JAMES HEMPSTEAD (Roche Institute of Molecular	P32 - 38	DALTON WANG, PING CHEN AND MIMI HALPERN (SUNY Health Science Center at Brooklyn) Further Isolation and Purification of the Garter Snake Chemoattractant in Earthworm Wash.
	Biology, Nutley, NJ 07110) In vivo and In Vitro Studies of the Localization of Antigens Recognized by Monoclonal Antibodies Neu-4, Neu-5 and Neu-9 in Rat Olfactory Epithelium.	P33 - 39	XIAN-CHENG JIANG, DALTON WANG AND MIMI HALPERN (SUNY Health Science Center at Brooklyn) Isolation and Purification of Earthworm Alarm Pheromone and Carter Snake Chemoattractant from Electric Shock - Induced Earthworm Secretions.
P21 - 27	BRUCE P. BRYANT (Monell Chemical Senses Center) AND JOSEPH G. BRAND (Monell Chemical Senses Center and Veterans Administration) Structure-Activity Relationships in the Arginine Receptive Taste Pathways of the Channel Catfish, Ictalurus punctatus	P34 - 40	MICHAEL A. ADAMS AND PETER B. JOHNSEN (Monell Chemical Senses Center, University of Pennsylvania, 3500 Market Street, Philadelphia, PA 19104) Feeding Enhancement in the Herbivorous Fish Tilapia zillii by Amino Acids.
P22 - 28	JOAN YONCHEK AND THOMAS E. FINGER (Univ. Colorado Medical School, Denver Co). Monoclonal Antibodies Directed Against Catfish Taste Receptors: Immunocytochemistry of Brain, Olfactory Epithelium and Oral Taste Buds.	P35 - 41	KAY L. GREENFIELD (Monell Chemical Senses Center, Philadelphia) GISELA EPPLE, ANNE M. BELCHER (Monell Chemical Senses Center; German Primate Center, Gottingen) JOSEF SCHNEIDER (German Primate Center) AND AMOS B. SMITH III (Monell Chemical Senses Center;
P23 - 29	D. E. KOLODNY AND L. M. KENNEDY. (Dept. of Biology, Clark Univ., Worcester MA 01610) Hodulcin Suppresses Behavioral and Receptor Cell Responses to Sucrose in the Blowfly.		Chemistry Department, University of Pennsylvania) Chemical Communication in the Saddle Back Tamarin: Do Non-Volatiles Play a Role?
P24 - 30	"ABSTRACT WITHDRAWN"	P36 -42	W. MICHEL, J. J. ROBINSON II AND J. CAPRIO (Department of Zoology and
P25 - 31	TIMOTHY S. McCLINTOCK AND BARRY W. ACHE (C. V. Whitney Lab, University of Florida) <u>Ion Channels from Lobster</u> Olfactory Receptor Neuron Somata.		Physiology, Louisiana State Univ., Baton Rouge, LA 70803) Olfactory and Gustatory Responses of the Channel Catfish, Ictalurus punctatus, to Nucleotides.
P26 - 32	I. SCHMIEDEL-JAKOB, P. A. V. ANDERSON AND B. W. ACHE (C. V. Whitney Laboratory, University of Florida) Effects of Different Ionic Environments on the Receptor Potential of Olfactory Receptor Cells in the Spiny Lobster.	P37 - 43	PETER C. DANIEL AND CHARLES D. DERBY (Department of Biology, Georgia State University, Atlanta, Georgia 30303) Mixture Interaction Analysis: A Polynomial Response Summation Model Which Incorporates the Beidler Equation.
	"CHEMICAL ECOLOGY"	FRIDAY, MAY	1 - Slide Presentations
P27 - 33	PAUL A. MOORE AND JELLE ATEMA (Boston University Marine Program) <u>Evaluation</u> of Odor Plume Parameters For Positional	Slide Sessio	"EARLY EVENTS: BIOCHEMISTRY" on II A - Hernando Desoco N
	Information Relative to the Source of the Plume.		r. A. I. Farbman, Northwestern Univ.
P28 - 34	RAINER VOIGT AND JELLE ATEMA (Boston University Marine Program, Marine Biological Laboratory, Woods Hole, MA 02543) Spatial-Temporal Filtering in Olfactory Chemoreceptor Cells.	8:00 - 44	R. G. VOGT, G. D. PRESTWICH (SUNY, Stony Brook) AND L. M. RIDDIFORD (U. of Washington) Visualization of a Putative Pheromone Receptor-protein Using a Tritium-labeled Photoaffinity Analog.
P29 - 35	R. W. MANKIN AND M. S. MAYER (Insect Attraactants, Behavior, and Basic Biology Research Laboratory, Agric. Res. Serv., U. S. Dept. Agric., Gainesville, FL) AND A. J. GRANT (Worcester Foundation for Experimental Biology,	8:15 - 45	HAYDEN G. COON, ROBERTO NININI AND FRANCESCO CURCIO (NIH, National Cancer Institute, Labortory of Genetics, Bethesda, MD) Clonal Cell Strains from Rat Olfactory Epithelium and Monoclonal Antibodies that Help Characterize Them.
	Shrewsbury, MA) Response Characteristics of Pheromone Receptor Neurons on Cabbage Looper Moth Antennae.	8:30 - 46	JONATHAN PEVSNER (Dept. of Pharmacology, Johns Hopkins Sch. Med., Baltimore MD 21205) RANDALL R. REED (Dept. Mol. Biol. & Genetics, Howard Hughes Med. Inst.,
P30 - 36	D. E. WOOD, CHARLES D. DERBY, DONALD H. EDWARDS (Department of Biology, Georgia State University, Atlanta, Georgia 30303) AND RICHARD A. GLEESON (C. V. Whitney Laboratory, University of Florida, Rt. 1, Box 121, St. Augustine, Florida 32086). An Analysis of the Courtship Display Behavior in the Blue Crab (Callinectes sapidus)		Johns Hopkins Sch. Med.) AND SOLOMON H. SNYDER (Dept. Neuroscience, Johns Hopkins Sch. Med.) Cloning and Sequence Analysis of Two cDNAs Encoding Rat Odorant-Binding Protein.

8:45 - 47	BARBARA MANIA-FARNELL, AND ALBERT I. FARBMAN (Dept. of Neurobiology, Northwestern Univ., Evanston, IL 60201) Immunohistochemical Localization of GTP-binding Protein in Rat Olfactory EpitheliumD during Prenatal Development.	9:15 - 57	JACQUELINE B. FINE-LEVY AND CHARLES D. DERBY (Department of Biology, Georgia State University, Atlanta, Georgia 30303) Quality Coding in Olfaction by Spiny Lobsters: Behavioral Discrimination.
9:00 - 48	DAVID T. JONES AND RANDALL R. REED (Howard Hughes Medical Institute, Johns Hopkins University) Isolatin and Characterization of Five G-Protein Encoding cDNA Clones from Rat Olfactory Epithelium.	9:30 - 58	NADIA GIRARDOT AND CHARLES D. DERBY (Department of Biology, Georgia State University, Atlanta, GA 30303) Neural Discrimination of Odorant Quality in the Spiny Lobster: Multivariate Analysis.
9:15 - 49	T. HUQUE*, J. G. BRAND*,+, J. L. RABINOWITZ+ AND D. L. BAYELY* (*Monell Chemical Senses Center, 3500 Market Street, Philadelphia, PA 19104; and + Veterans Administration Medical Center and University of Pennsylvania, philadelphia, PA 19104) Phospholipid Turnover in Catfish Barbel (Taste)	9:45 - 59	J. S. KAUER (Tufts-New England Medical Center, Boston, MA) Real-time Video Recording of Olfactory Bulb Evoked Potentials in the Salamander Using a Voltage-sensitive Dye. "EARLY EVENTS: BIOPHYSICS"
	Epithelium with Special Reference to Phosphatidylinositol-4,5-bisphosphate.	Slide Session Chaired by Da	a III A - Hernando Desoto N . S. D. Roper, Colorado State Univ.
9:30 - 50	RICHARD C. BRUCH, R. DINO RULLI (Monell Chemical Senses Center) AND ARDITHANNE G. BOYLE (Monell Chemical Senses Center; Veterans Administratin Medical Center) Olfactory L-Amino Acid Receptor Specificity and Stimulation of Potential	10:30 - 60	PEDRO LABARCA, SIDNEY A. SIMON AND ROBERT R. H. ANHOLT (Department of Physiology, Box 3709, Duke University Medical Center, Durham, NC 27710) Ion Channels of Olfactory Cilia
9:45 - 51	Second Messengers. HENRY G. TRAPIDO-ROSENTHAL, RICHARD A. GLEESON AND WILLIAM E. S. CARR (C. V. Whitney Laboratory and Dept. of Zoology, University of Florida, St. Augustine, FL	10:45 - 61	TADASHI NAKAMURA AND GEOFFREY H. GOLD (Dept. of Physiology, Yale U. Sch. of Med., New Haven, CT 06510) A Cyclic Nucleotide—gated conductance in Olfactory Receptor Cilia.
	32086) Blochemistry and Physiology of Purinergic and Taurinergic Chemosensory Systems. "OTHER CHEMORECEPTION: QUALITY CODING"	11:00 - 62	GABRIELLE NEVITT (Department of Zoology, University of Washington, Seattle, Washington, 98195) Electrical Properties of Olfactory Receptor Cells Isolated from Coho Salmon (Oncorhynchus kisutch).
Slide Sessio	n II B - Hernando Desoto S r. M. Meredith, Florida State Univ.	11:15 - 63	KEIICHI TONOSAKI AND MASAYA FUNAKOSHI
8:00 - 52	JOEL WHITE AND MICHAEL MEREDITH (Dept. of Biological Science, Florida State University, Tallahassee, FL 32306) Intracellular Recordings from Ganglion		(Dept. of Oral Physiology, Sch. of Dent., Asahi Univ., Hozumi, Hozumi, Motosu, Gifu 501-02, JAPAN) Effects of Injection of Cyclic-GMP into the Mouse Taste Cells.
	Cells of the Elasmobranch Nervus Terminalis.	11:30 - 64	JOHN A. DESIMONE, GERARD L. HECK, AND KRISHNA PERSAUD (Dept. of Physiology,
8:15 - 53	ROBERT E. JOHNSTON AND CHERYL PFEIFFER (Cornell University) Effects of Vomeronasal and Main Olfactory Lesions on Communicative Behaviors and Hormone		Virginia Commonwealth University, Richmond, VA 23298) Transmucosal Inward Currents Coincide With Neural Excitation and Adaptation to NaCl in the Rat.
8: 30 - 54	M. N. LEHMAN (Univ. Cincinnati Coll. Med.) S. W. NEWMAN (Univ. Michigan) AND A. J. SILVERMAN (Columbia Univ. P & S)	11:45 - 65	M. KIM AND C. M. MISTRETTA (Nursing and Dentistry, Univ. Michigan, Ann Arbor, MI 48109) Effects of Potassium Channel Blockers on Rat Chorda Tympani Nerve Responses.
	The Origin of Luteinizing Hormone- Releasing Hormone (LHRH) in the Vomeronasal System of the Hamster.	12:00 - 66	S. A. SIMON, V. F. HOLLAND, R. ROBB AND R. P. ERICKSON (Dept. of Psychology,
8:45 - 55	JOHN J. LEPRI, CHARLES J. WYSOCKI, YAIR KATZ AND LINDA M. WYSOCKI (Monell Chemical Senses Center, 3500 Market Street, Philadelphia, PA 19104) JAY B.		Duke University, Durham, NC 27710) Transection of Chorda Tmpani Nerve Affects Epithelial Transport Across Dog Tongue.
9:00 - 56	LABOV (Department of Biology, Colby College, Waterville, ME 04901) AND N. JAY BEAN (Department of Psychology, Vassar College, Poughkeepsie, NY 12601) Vomeronasal Chemoreception, Sexual Experience and Reproductive Behavior of Male House Mice and Prairie Voles.	12:15 - 67	SUE C. KINNAMON AND STEPHEN D. ROPER (Department of Anatomy, Colorado State University, Ft. Collins, Co 80523 and Rocky Mountain Taste and Smell Center, Denver CO 80262) Identification of the Apical Specialization in Dissociated Taste Cells by a Selective Staining Procedure.
9:00 - 56	B. OAKLEY, D. R. RIDDLE, C. R. BELCYSINZKI, C. L. DeSIBOUR AND S. E. HUGHES (University of Michigan, Ann Arbor, MI 48109 and Washington University, St. Louis, MO 63119) Inhibition of Chorda Tympani Taste		
	Responses in a Model System.		

	"CHEMICAL ECOLOGY" n III A - Hernando Desoto N r. R. A. Gleeson, Univ. Florida	P5 - 79	SUAT GURKAN AND ROBERT M. BRADLEY (Dept. Oral Biol., School Dent., Univ. Michegan, Ann Arbor, MI 48109) Electrical Stimulation of the Autonomic
10:30 - 68	RICHARD K. ZIMMER-FAUST (Marine Science Institute, University of California, Santa Barbara, CA 93106) RICHARD A. GLEESON AND WILLIAM E. S. CARR (C. V. Whitney Marine Laboratory, University of Florida, St. Augustine, FL 32086) Behavioral and Physiological Responses	P6 - 80	Supply of von Ebner's Glands in the Rat. THERESA A. HARRISON AND ROBERT M. BRADLEY (Dept. Oral Biol., School Dent., Univ. Michigan, Ann Arbor, MI 48109) Electrical Stimulation of Rat Medullary Neurons Controlling von Ebner's Glands
10:45 - 69	Chemoreceptors. DAN RITTSCHOF, RICHARD B. FORWARD, JR. AND MONA C. DeVRIES (Duke University	P7 - 81	BARBARA K. GIZA AND THOMAS R. SCOTT (Department of Psychology and Institute for Neuroscience, University of Delaware, Newark, DE 19716) Cholecystokinin Administration Does Not
	Marine Laboratory, Beaufort, NC 28516) Peptide Pheromones Synchronize Crustacean Egg Hatching and Larval Release.	P8 - 82	Influence Taste-evoked Activity in Rat NTS. LESLIE L. WIGGINS (Oxford University),
11:00 - 70	PETER W. SORENSEN, (Department of Zoology, University of Alberta, Edmonton, Alberta, Canada) TOSHIAKI J. HARA (Department of Fisheries and Oceans, Freshwater Institute, Winnipeg,		DAVID V. SMITH AND ROBERT A. FRANK (University of Cincinnati) Taste Processing in the Rabbit: Generalization of Learned Taste Aversions.
	Manitoba, Canada) AND NORMAN E. STACEY (Department of Zoology, University of Alberta, Edmonton, Alberta, Canada) <u>High</u>		"Taste: Anatomy"
11:15 - 71	Olfactory Sensitivity of Mature Male Goldfish to Prostaglandins, Presumed Spawning Pheromones. C. E. LINN JR., AND W. L. ROELOFS (Dept.	P9 - 83	T. A. CUMMINGS AND S. D. ROPER (Dept. of Anatomy, Colorado State University, Fort Collins, CO 80523) A Freeze Fracture Study on the Apical Specializations of Taste Cells in the Mudpuppy, Necturus
	Entomology, NYS Ag Expt Station, Cornell Univ., Geneva, NY 14456) Pheromone Components and Active Spaces: What do Male Moths Smell and When Do They Smell It?	P10 - 84	WAR L. ST. JEOR AND JOHN C. KINNAMON (Dept. MCD Biology, Univ. of Colorado, "Boulder) Ultrastructure of Gerbil Fungiform Taste Buds
11:30 - 72	JELLE ATEMA (Boston Univ. Marine Program, Woods Hole, MA) <u>Tuning and</u> Noise: The Acoustic Analogy	P11 - 85	PHILLIP S. LASITER (Dept. Psychol., Florida Atlantic Univ.), DAVID L. HILL
11:45 - 73	RICHARD K. ZIMMER-FAUST AND GORDON SMYTH (University of California at Santa Barbara) <u>Difference Thresholds for Odor</u> <u>Detection</u> by an Aquatic Animal. M. LEVANDOWSKY (Haskins Labs of Pace		(Dept. Psychol., Univ. of Virginia, Charlottesville), AND DIANE A. MOORMAN (Dept. Psychol., Florida Atlantic University) Relations Between Planar Dendritic Growth and Cytochrome Oxidase Activity in the Pontine Taste Area of Developing Rats.
	University, N.Y., NY 10038) J. KLAFTER AND B. S. WHITE (Exxon Research and Engineering Co., Annandale, NJ 08801) When is Chemoreception Useful in the Nutrition of Grazing Microzooplankton?	P12 - 86	T. HAYAMA AND J. CAPRIO (Department of Zoology and Physiology, Louisiana State University, Baton Rouge, LA 70803) Organization and Responses of Facial Lobe Subnuclei in the Channel Catfish, Ictalurus punctatus.
PRIDAY, MAY	1 - Poster Presentations	P13 - 87	INGLIS MILLER, JR. (Bowman Gray Sch.
Poster Sessi	on II - Sara Desoto N & S "TASTE: PHYSIOLOGY"		Med. Wake Forest Univ.) Fungiform Taste Pore Quantification in Living Rabbits.
Pl - 75	MOHSSEN S. NEJAD AND LLOYD M. BEIDLER (Department of Biological Science, The		"MIXTURE INTERACTIONS"
P2 - 76	Florida State University, Tallahassee, FL 32306) Palato-Lingual Taste Responses of the Mongolian Gerbil. THOMAS P. HETTINGER AND MARION E. FRANK	P14 - 88	PETER C. DANIEL AND CHARLES D. DERBY (Department of Biology, Georgia State University, Atlanta, Georgia 30303) An Assessment of Behavioral Olfactory Discrimination in The Sping Lobster
	(University of Connecticut Health Center, Farmington, CT 06032) Specificity of Amiloride Inhibition of Taste Responses in Single Fibers of the Hamster Chorda Tympani Nerve.	P15 - 89	Using an Habituation Paradigm. SIZUKO YAMAGUCHI, MIKA KIMURA AND YASUSHI KOMATA (Central Research Laboratories, A jinomoto Co., Inc.)
P3 - 77	HARRY WMS. HARPER (Stauffer Chemical Co.) AND B. W. KNIGHT (Rockefeller University). The Correct Method of Signal Processing for Whole-nerve Recordings.	P16 - 90	Contribution of Salivary Level of Glutamate to the Cause of Umami Taste of Ribonucleotides SUSAN HUBAY AND ROBERT A. FRANK (Dept.
P4 - 78	TAKAMITSU HANAMORI AND DAVID V. SMITH (University of Cincinnati College of Medicine) Concentration-Response		of Psychology, Univ. of Cincinnati) The Sweetness of Binary Mixtures of Sucrose and Stevioside: An Information Integration Analysis.
	Functions of Single Chemosensory Fibers in the Hamster Superior Laryngeal Nerve.	P17 - 91	T. E. ACREE, A. B. MARIN AND J. BARNARD (Cornell University, Geneva) An Introduction to Charm Analysis.

P18 - 92	J. E. COMETTO-MUNIZ, M. R. GARCIA- MEDINA, A. M. CALVINO AND S. M. HERNANDEZ (Laboratorio de Investigaciones Sensoriales, Conicet - Esc. Sal. Publ., Fac. Medicina, UBA,	P30 - 104	K. A. HAMILTON, R. M. KREAM AND J. S. KAUER (Tufts-New England Medical Center, Boston, MA) Neurochemical Analysis of the Salamander Olfactory Bulb.
	C.C.53, 1453 Buenos Aires, Argentina) Perceptual Properties of Pungent Odorants.	P31 - 105	ULRIKE GRUNERT AND BARRY W. ACHE (C. V. Whitney Laboratory, University of Florida, St. Augustine, FL 32086) Fine Structure of the Olfactory (Aesthetasc) Sensilla of the Spiny Lobster.
	"OLFACTION: PHYSIOLOGY"	P32 - 106	M. M. DASTON AND G. D. ADAMEK (Dept.
P19 - 93	T. A. HARRISON, J. W. SCOTT, R. K. CONINE AND D. P. WELLIS (Department of Anatomy & Cell Biology, Emory University, Atlanta, Georgia 30322) Stimulus Response Functions of Rat	132 - 100	Anat. & Cell Biol., Univ. of Cincinnati) Neighborliness of Cobalt-Filled Frog Olfactory Axons from Epithelium to Olfactory Bulb.
	Olfactory Bulb Neurons	P33 - 107	ANDREW N. CLANCY, THOMAS A. SCHOENFELD
P20 - 94	B. R. JOHNSON (Cornell University, Ithaca, NY) R. VOIGT, C. L. MERRILL AND J. ATEMA (Boston University Marine Program, Marine Biological Lab., Woods Hole, MA) Stimulus Intensity Discrimination by Lobster Olfactory Receptors.		AND FOTEOS MACRIDES (Worcester Foundation for Experimental Biology, Shrewsbury, MA 01545) Spatial Organization of Receptor Surfaces and Odorant Passageways in the Hamster Nasal Cavity: Morphometric Analyses Based on a Stereotaxic Atlas.
P21 - 95	W. T. NICKELL AND M. T. SHIPLEY (University of Cincinnati College of Medicine) Prolonged Inhibition of Anterior Commissure Input of the Olfactory Bulb by Stimulation of the Diagonal Band.	P34 - 108	E.MEISAMI (Physiology Dept., Univ. Illinois, Urbana, IL 61801) AND B. WENZEL (Physiology Dept., Univ. California, Los Angeles, CA 90024) Is the Northern Fulmar's Large Olfactory Bulb Designed for High Sensitivity in Odor Detection?
P22 - 96	CHARLES DERBY AND DAVID BLAUSTEIN (Department of Biology, Georgia State University, Atlanta, Georgia 30303) Morphological and Physiological Characterization of Individual Olfactory Interneurons in the Protocerebrum of the Crayfish.	P35 - 109	K. BHATNAGAR (Anatomy Dept., Univ. Louisville, Louisville, KY 40292) AND E. MEISAMI (Physiology Dept., Univ. Illinois, Urbana, IL 61801) Neuron Number and Topography in the Human Intrabulbar Anterior Olfactory Nucleus.
P23 - 97	DAVID P. WELLIS AND JOHN W. SCOTT	SATURDAY, MA	X 2 - Slide Presentations
	(Department of Anatomy and Cell Biology, Emory University School of Medicine	•	
		•	"ODOR AND TASTE MIXTURES"
	Atlanta, GA 30322) <u>Intracellular</u>		ODOR MID INGIL HILLOMID
<i>;</i> •	Recordings of Odor Induced Responses in the Rat Olfactory Bulb.		on IV - Hernando Desoto N & S or. S. Price, Medical College Virginia
F24 - 98	Recordings of Odor Induced Responses in		on IV - Hernando Desoto N & S
P24 - 98	Recordings of Odor Induced Responses in the Rat Olfactory Bulb. DONALD A. WILSON AND MICHAEL LEON (Department of Psychobiology, University of California, Irvine) Evidence of Lateral Synaptic Interactions in Olfactory Bulb Output Cell Responses to	Chaired by D	n IV - Hernando Desoto N & S r. S. Price, Medical College Virginia R. C. GESTELAND AND G. D. ADAMEK (Osphresiopolis, Dept. Anat. & Cell Biol., Univ. of Cincinnati) Adaptation and Mixture Component Suppression in
	Recordings of Odor Induced Responses in the Rat Olfactory Bulb. DONALD A. WILSON AND MICHAEL LEON (Department of Psychobiology, University of California, Irvine) Evidence of Lateral Synaptic Interactions in Olfactory Bulb Output Cell Responses to Odors. K. A. HAMILTON AND J. S. KAUER (Tufts-New England Medical Center, Boston MA) Responses of Mitral/Tufted Cells to Electrical Stimulation in the Olfactory	Chaired by D 8:00 - 110	n IV - Hernando Desoto N & S r. S. Price, Medical College Virginia R. C. GESTELAND AND G. D. ADAMEK (Osphresiopolis, Dept. Anat. & Cell Biol., Univ. of Cincinnati) Adaptation and Mixture Component Suppression in Olfaction. PAOLA F. BORRONI AND JELLE ATEMA (Boston University Marine Program, Marine Biological Laboratory, Woods Hole, MA) Cross Adaptation of Primary Chemosensory
P25 - 99	Recordings of Odor Induced Responses in the Rat Olfactory Bulb. DONALD A. WILSON AND MICHAEL LEON (Department of Psychobiology, University of California, Irvine) Evidence of Lateral Synaptic Interactions in Olfactory Bulb Output Cell Responses to Odors. K. A. HAMILTON AND J. S. KAUER (Tufts-New England Medical Center, Boston MA) Responses of Mitral/Tufted Cells to Electrical Stimulation in the Olfactory Bulb of the Tiger Salamander STEPHEN P. FRACEK, JR., LINDA E. EZISNY AND ROLLIE SCHAFER (North Texas State University, Denton, TX 76205) Olfactory Bulb Neurons Develop Rhythmic Activity	8:00 - 110 8:15 - 111	n IV - Hernando Desoto N & S r. S. Price, Medical College Virginia R. C. GESTELAND AND G. D. ADAMEK (Osphresiopolis, Dept. Anat. & Ceil Biol., Univ. of Cincinnati) Adaptation and Mixture Component Suppression in Olfaction. PAOLA F. BORRONI AND JELLE ATEMA (Boston University Marine Program, Marine Biological Laboratory, Woods Hole, MA) Cross Adaptation of Primary Chemosensory Neurons. SUSAN S. SCHIFFMAN, CAMILLA GRAHAM AND ZOE WARWICK (Department of Psychiatry, Duke Medical Center, Durham, NC 27710) Inosine-5'-monophosphate and Inosine Enhance Some Sweet Tastes. HARRY T. LAWLESS (S. C. Johnson & Son., Inc.) An Analogy to the Release From Mixture Suppression Effect in Taste
P25 - 99	Recordings of Odor Induced Responses in the Rat Olfactory Bulb. DONALD A. WILSON AND MICHAEL LEON (Department of Psychobiology, University of California, Irvine) Evidence of Lateral Synaptic Interactions in Olfactory Bulb Output Cell Responses to Odors. K. A. HAMILTON AND J. S. KAUER (Tufts-New England Medical Center, Boston MA) Responses of Mitral/Tufted Cells to Electrical Stimulation in the Olfactory Bulb of the Tiger Salamander STEPHEN P. FRACEK, JR., LINDA E. EZISNY AND ROLLIE SCHAFER (North Texas State University, Denton, TX 76205) Olfactory Bulb Neurons Develop Rhythmic Activity in Cell Culture.	8:00 - 110 8:15 - 111 8:30 - 112	R. C. GESTELAND AND G. D. ADAMEK (Osphresiopolis, Dept. Anat. & Ceil Biol., Univ. of Cincinnati) Adaptation and Mixture Component Suppression in Olfaction. PAOLA F. BORRONI AND JELLE ATEMA (Boston University Marine Program, Marine Biological Laboratory, Woods Hole, MA) Cross Adaptation of Primary Chemosensory Neurons. SUSAN S. SCHIFFMAN, CAMILLA GRAHAM AND ZOB WARWICK (Department of Psychiatry, Duke Medical Center, Durham, NC 27710) Inosine-5'-monophosphate and Inosine Enhance Some Sweet Tastes. HARRY T. LAWLESS (S. C. Johnson & Son., Inc.) An Analogy to the Release From Mixture Suppression Effect in Taste Occurs in Odor Mixtures. MICHAEL D. RABIN (Connecticut Chemosensory Clinical Research Center, University of Connecticut Health Center, Farmington, CT 06032) Experience
P25 - 99 P26 - 100	Recordings of Odor Induced Responses in the Rat Olfactory Bulb. DONALD A. WILSON AND MICHAEL LEON (Department of Psychobiology, University of California, Irvine) Evidence of Lateral Synaptic Interactions in Olfactory Bulb Output Cell Responses to Odors. K. A. HAMILTON AND J. S. KAUER (Tufts-New England Medical Center, Boston MA) Responses of Mitral/Tufted Cells to Electrical Stimulation in the Olfactory Bulb of the Tiger Salamander STEPHEN P. FRACEK, JR., LINDA E. EZISNY AND ROLLIE SCHAFER (North Texas State University, Denton, TX 76205) Olfactory Bulb Neurons Develop Rhythmic Activity in Cell Culture. "OLFACTION: ANATOMY" M. T. SHIPLEY, J. H. MCLEAN AND M. N. LEHMAN (Univ. of Cincinnati) Contrasting Patterns of Serotonergic Innervation of the Main and Accessory Olfactory Bulb in the Adult Rat. C. REYHER (University of Berlin, West Germany) J. H. MCLEAN AND M. T. SHIPLEY	8:15 - 111 8:30 - 112 8:45 - 113 9:00 - 114	R. G. GESTELAND AND G. D. ADAMEK (Osphresiopolis, Dept. Anat. & Ceil Biol., Univ. of Cincinnati) Adaptation and Mixture Component Suppression in Olfaction. PAOLA F. BORRONI AND JELLE ATEMA (Boston University Marine Program, Marine Biological Laboratory, Woods Hole, MA) Cross Adaptation of Primary Chemosensory Neurons. SUSAN S. SCHIFFMAN, CAMILLA GRAHAM AND ZOE WARWICK (Department of Psychiatry, Duke Medical Center, Durham, NC 27710) Inosine-5'-monophosphate and Inosine Enhance Some Sweet Tastes. HARRY T. LAWLESS (S. C. Johnson & Son., Inc.) An Analogy to the Release From Mixture Suppression Effect in Taste Occurs in Odor Mixtures. MICHAEL D. RABIN (Connecticut Chemosensory Clinical Research Center, University of Connecticut Health Center, Farmington, CT 06032) Experience Facilitates Olfactory Discrimination and Mixture Component Analysis by Humans.
P25 - 99 P26 - 100 P27 - 101	Recordings of Odor Induced Responses in the Rat Olfactory Bulb. DONALD A. WILSON AND MICHAEL LEON (Department of Psychobiology, University of California, Irvine) Evidence of Lateral Synaptic Interactions in Olfactory Bulb Output Cell Responses to Odors. K. A. HAMILTON AND J. S. KAUER (Tufts-New England Medical Center, Boston MA) Responses of Mitral/Tufted Cells to Electrical Stimulation in the Olfactory Bulb of the Tiger Salamander STEPHEN P. FRACEK, JR., LINDA E. EZISNY AND ROLLIE SCHAFER (North Texas State University, Denton, TX 76205) Olfactory Bulb Neurons Develop Rhythmic Activity in Cell Culture. "OLFACTION: ANATOMY" M. T. SHIPLEY, J. H. MCLEAN AND M. N. LEHMAN (Univ. of Cincinnati) Contrasting Patterns of Serotonergic Innervation of the Main and Accessory Olfactory Bulb in the Adult Rat. C. REYHER (University of Berlin, West Germany) J. H. MCLEAN AND M. T. SHIPLEY (University of Cincinnati) Evidence For Olfactory Inputs To The Septum.	8:00 - 110 8:15 - 111 8:30 - 112	R. C. GESTELAND AND G. D. ADAMEK (Osphresiopolis, Dept. Anat. & Ceil Biol., Univ. of Cincinnati) Adaptation and Mixture Component Suppression in Olfaction. PAOLA F. BORRONI AND JELLE ATEMA (Boston University Marine Program, Marine Biological Laboratory, Woods Hole, MA) Cross Adaptation of Primary Chemosensory Neurons. SUSAN S. SCHIFFMAN, CAMILLA GRAHAM AND ZOB WARWICK (Department of Psychiatry, Duke Medical Center, Durham, NC 27710) Inosine-5'-monophosphate and Inosine Enhance Some Sweet Tastes. HARRY T. LAWLESS (S. C. Johnson & Son., Inc.) An Analogy to the Release From Mixture Suppression Effect in Taste Occurs in Odor Mixtures. MICHAEL D. RABIN (Connecticut Chemosensory Clinical Research Center, University of Connecticut Health Center, Farmington, CT 06032) Experience Facilitates Olfactory Discrimination and Mixture Component Analysis by Humans. A. B. MARIN, T. E. ACREE AND J. BARNARD (Cornell University, Geneva) Variation
P25 - 99 P26 - 100 P27 - 101	Recordings of Odor Induced Responses in the Rat Olfactory Bulb. DONALD A. WILSON AND MICHAEL LEON (Department of Psychobiology, University of California, Irvine) Evidence of Lateral Synaptic Interactions in Olfactory Bulb Output Cell Responses to Odors. K. A. HAMILTON AND J. S. KAUER (Tufts-New England Medical Center, Boston MA) Responses of Mitral/Tufted Cells to Electrical Stimulation in the Olfactory Bulb of the Tiger Salamander STEPHEN P. FRACEK, JR., LINDA E. EZISNY AND ROLLIE SCHAFER (North Texas State University, Denton, TX 76205) Olfactory Bulb Neurons Develop Rhythmic Activity in Cell Culture. "OLFACTION: ANATOMY" M. T. SHIPLEY, J. H. MCLEAN AND M. N. LEHMAN (Univ. of Cincinnati) Contrasting Patterns of Serotonergic Innervation of the Main and Accessory Olfactory Bulb in the Adult Rat. C. REYHER (University of Berlin, West Germany) J. H. MCLEAN AND M. T. SHIPLEY (University of Cincinnati) Evidence For	8:15 - 111 8:30 - 112 8:45 - 113 9:00 - 114	R. C. GESTELAND AND G. D. ADAMEK (Osphresiopolis, Dept. Anat. & Ceil Biol., Univ. of Cincinnati) Adaptation and Mixture Component Suppression in Olfaction. PAOLA F. BORRONI AND JELLE ATEMA (Boston University Marine Program, Marine Biological Laboratory, Woods Hole, MA) Cross Adaptation of Primary Chemosensory Neurons. SUSAN S. SCHIFFMAN, CAMILLA GRAHAM AND ZOE WARWICK (Department of Psychiatry, Duke Medical Center, Durham, NC 27710) Inosine-5'-monophosphate and Inosine Enhance Some Sweet Tastes. HARRY T. LAWLESS (S. C. Johnson & Son., Inc.) An Analogy to the Release From Mixture Suppression Effect in Taste Occurs in Odor Mixtures. MICHAEL D. RABIN (Connecticut Chemosensory Clinical Research Center, University of Connecticut Health Center, Farmington, CT 06032) Experience Facilitates Olfactory Discrimination and Mixture Component Analysis by Humans. A. B. MARIN, T. E. ACREE AND J. BARNARD

SATURDAY, MA	Y 2 - Poster Presentations	P14 - 129	CHARLES N. STEWART, MARCUS W. THOMSEN
	"PLASTICITY AND DEVELOPMENT"		AND JOSEPH M. DALTON (Franklin & Marshall College, Lancaster PA 17604) Comparison of Aspartame and 4-
P1 - 116	M. B. VOGT AND C. M. MISTRETTA (Dept. Oral Biol., School Dent., and Center for Nursing Research, Univ. Michigan, Ann Arbor, MI 48109) <u>Adaptation</u>	P15 - 130	Chloroaspartame Taste Characteristics in Mice and Monkeys. A. C. SPECTOR (Univ. of Penn.) G.
	Characteristics of NST Taste Neuron Responses to Salts in Fetal and Postnatal Sheep.		SCHWARTZ (Monell Chem. Senses Ctr.) AND H. J. GRILL (Univ. of Penn.) Sucrose Detection Thresholds Before and After Peripheral Gustatory Nerve Transection
P2 - 117	P. R. PRZEKOP JR., AND D. L. HILL (University of Virginia) Environmental Effects Upon the Developing Gustatory System: Towards a "Sensitive Period".	P16 - 131	in Rats. ALAN C. SPECTOR AND HARVEY J. GRILL (Univ. of Penn.) Taste Quality Distinctions Between Maltose and Sucrose
P3 - 118	JOSEPH C. STEVENS AND WILLIAM S. CAIN (John B. Pierce Foundation Laboratory and Yale University) Smelling through the Nasopharynx; Effect of Aging.		in Rats: Issues Involving the Generalization of Conditioned Taste Aversions.
P4 - 119	RICHARD M. COSTANZO AND EDWARD E. MORRISON (Medical College of Virginia, Richmond, VA 23298) Olfactory Nerve Transection in the Aging Hamster: Neurogenesis and Axon Reconnection of the Olfactory Bulb.	P17 - 132	ADAM DREWNOWSKI (University of Michigan School of Public Health), FRANCE BELLISLE (College de France, Paris, France) AND PIERRE AIMEZ AND BRIGITTE REMY (Hotel-Dieu, Paris, France) Taste and Bulimia.
P5 - 120	HILARY J. SCHMIDT AND GARY K. BEAUCHAMP (Monell Chemical Senses Center) Hedonic Reactions to Odors in Three-Year-Old Children.	P18 - 133	Senses Center, Philadelphia, PA) Perceptual and Salivary Responses to Oral Chemical Irritants Among Frequent and Infrequent Consumers of Hot Spices.
P6 - 121	REGINA SULLIVAN AND MICHAEL LEON (University of California - Irvine) NE Modulation of One-trial Olfactory Conditioning and Olfactory Bulb Neural Responding to an Attractive Odor.	P19 - 134	SANDRA P. FRANKMANN AND BARRY G. GREEN (Monell Chemical Senses Center) Effect of Cooling on the Sweetness of Natural and Artificial Sweeteners.
P7 - 122	KUNIO YAMAZAKI, GARY K. BEAUCHAMP, DONNA KUPNIEWSKI, CATHI STAHLBAUM (Monell Chemical Senses Center) JUDY BARD, LEWIS THOMAS AND EDWARD A. BOYSE (Memorial	P20 -135	BARRY G. GREEN (Monell Chemical Senses Center) Spatial Summation of Chemical Irritation on the Tongue. ALIKI AKONTIDOU, ROSS CARTER AND ROBERT
	Sloan-Kettering Cancer Center) Relation of MHC-Related Mating Preferences to Postnatal Chemosensory Imprinting.		A. FRANK (Dept. of Psychology, Univ. of Cincinnati) Sweet and Bitter Perception in PTC Tasters and Non-tasters: A Multi-Measure Approach.
P8 - 123	HEATHER J. DUNCAN, GARY K. BEAUCHAMP AND KUNIO YAMAZAKI (Monell Chemical Senses Center) Relative Contribution of Different Genetic Regions to Urinary Odors Distinguishing Inbred Strains of Mice.	P22 - 137	DEBRA L. KORCHMAR, ALIKI AKONTIDOU AND ROBERT A. FRANK (Dept. of Psychology, Univ. of Cincinnati) A Preliminary Study of Chocolate Perception in PTC Tasters and Non-Tasters.
P9 - 124	CATHI C. STAHLBAUM (Monell Chemical Senses Center and Cornell University) The Influence of Olfactory Bulbectomy on the Mate Choice of Female Mice.	P23 - 138	TERESA ANNE VOLLMECKE AND SHARON VERDINELLI (Department of Psychology, University of Pennsylvania, 3815 Walnut St., Phila. Pa. 19104) Effects of Context on Sweetness Evaluation:
P10 - 125	DAVID BEGUN AND MIMI HALPERN (SUNY Health Science Center at Brooklyn) Conditioned Discrimination of an Airborne Odorant by Garter Snakes,	P24 - 139	Generalization Between Different Flavors. RICK BELL AND MABEL M. CHAN (New York
P11 - 126	Thamnophis sirtalis sirtalis. E. MEISAMI (Physiology Dept., Univ. Illinois, Urbana, IL 61801) A Possibly		University) The Relatonship between Carbohydrate and Simple Sugar Intake and Measures of Sweet Taste Preference in Free-living Adults.
	New Axon-Bearing Spiny Neuron in the Olfactory Bulb of Developing Rats.	P25 - 140	DEBORAH A. FROEHLICH AND ROSE MARIE PANGBORN (Department of Food Science and Technology, University of California,
P12 - 127	"INTAKE REGULATION AND NUTRITION" JAMES C. SMITH, LAURA WILSON AND DANA		Davis, CA 95626 USA) Effect of Mastication on Human Parotid Salivary Flow Rate and Alpha-amylase Secretion.
D13 - 128	MERRYDAY (The Florida State University) A Longitudinal Study of Sucrose Intake in the Fisher-344 Rat.	P26 - 141	SYED SHAMIL AND GORDON G. BIRCH (University of Reading) Structure, Taste and Solution Properties of 5-Membered
P13 - 128	RUDY A. BERNARD, TIMOTHY W. PRIEHS AND KAREN MOONEY (Dept. of Physiology, Michigan State University, East Lansing MI 48824) Salt Appetite in Hamsters is Linked to Escape from Retention Effects	P27 - 142	Rings. DAVID J. MELA (Moneli Chemical Senses Center, Philadelphia, PA) Sensory Effects of Fat Content in Food.
	of DOCA.	P28 - 143	DAVID J. MELA, RICHARD D. MATTES, SHUYA TANIMURA, MICHAEL ADAMS, AND CAROL CHRISTENSEN (Monell Chemical Senses Center, 3500 Market St. Philadelphia, PA 19104) Bitter Taste Sensitivity to Caffeine in Users and Non-Users.

P29 - 144	V. K. STONE, L. E. MARKS, J. C. STEVENS AND L. M. BARTOSHUK (John B. Pierce Foundation Laboratory) Chemosensory Magnitude Matching.	9:00 - 155	J. CARTER ROWLEY III, DAVID T. MORAN AND BRUCE W. JAFEK (University of Colorado School of Medicine) Tracer Studies of Microvillar Cells Suggest A Second Morphologically Distinct Class of
P30 - 145	M. ALBRIGHT (T. J. Lipton, Inc., Englewood Cliffs, NJ) <u>Effects of</u> Different Response Tasks on		Sensory Neuron Exists in Mammalian Olfactory Epithelia.
	Discrimination of Chemosensory Stimuli of Varying Complexity.	9:15 - 156	BERT Ph.M. MENCO (Dept. of Neurobiology & Physiology, O.T. Hogan Building, Northwestern University, Evanston, IL
	"VOMERONASAL, TRIGEMINAL AND TERMINAL NERVE CHEMORECEPTION"		60201) Tight-Junctions in Developing Rat Olfactory Epithelia.
P31 - 146	MICHAEL MEREDITH (Dept. Biol. Sci., Florida State University, Tallahassee, FL) Chronic Electrophysiological Recordings of Vomeronasal Pump Activation in Awake Animals.	9: 30 - 157	B. ZIELINSKI, M. L. GETCHELL AND T. V. GETCHELL (Dept. of Anatomy and Cell Biology, Wayne State University School of Medicine, Detroit MI 48201) Sustentacular Cell Ultrastructure: Evidence for Secretion and Absorption in the Salamander.
P32 - 147	WAYNE L. SILVER AND DIANNE B. WALKER (Department of Biology, Wake Forest University, Winston-Salem, NC 27109) Nasal Trigeminal Chemoreception: Response to Nicotine.	9:45 - 158	MARILYN L. GETCHELL, THOMAS E. FINGER AND THOMAS V. GETCHELL (Department of Anatomy & Cell Biology, Wayne State University School of Medicine, Detroit,
P33 - 148	M. R. GARCIA-MEDINA, J. E. COMETTO-MUNIZ AND A. M. CALVINO (Laboratorio de Investigaciones Sensoriales, CONICET - Esc. Sal. Publ., Fac. Medicina, UBA, C.C.53, 1453-Buenos Aires, Argentina) Trigeminal and Topographical Characteristics of Odors.		MI 48201 and Department of Cellular & Structural Biology, University of Colorado Medical School, Denver, CO 80262, USA) Localization of NGF-like, VIP-like and Substance P-like Immunoreactivity in the Olfactory Mucosae of the Salamander, Bullfrog and Grass Frog.
P34 - 149	J. RUSSELL MASON (USDA/APHIS/ADC Denver Wildlife Research Center, c/o Monell Chemical Senses Center) AND LARRY CLARK (Monell Chemical Senses Center, 3500 Market Street, Philadelphia, PA 19104) Behavioral Assessment of Olfactory and Trigeminal Responsiveness of Starlings (Sturnus vulgaris) to 9 Anthranilates.	Chaired by D School	"TASTE: PERIPHERAL AND CENTRAL ANATOMY" n V B - Hernando Desoto S r. D. T. Moran, Univ. Colorado Medical
P35 - 150	CELESTE R. WIRSIG AND SCOTT F. BASINGER (Department of Ophthalmology, Baylor College of Medicine, Houston, Texas 77030) The Terminal Nerve in the Bird.	8:00 - 159	C. M. MISTRETTA (Dentistry and Nursing, Univ of Michigan, Ann Arbor), S. GURKAN AND R. M. BRADLEY (Dentistry, Univ. of Michigan) Morphology of Chorda Tmpani Fiber Receptive Fields and Proposed Neural Rearrangements During Development.
SUNDAY, MAY	3- Slide Presentation	8:15 - 160	D. R. RIDDLE AND B. OAKLEY (University
	"OLFACTION: PERIPHERAL ORGANIZATION"		of Michigan, Ann Arbor, MI 48109) Some Effects upon Fungiform and Foliate Taste Buds of Condensing the Innervation by
	n V A - Hernando Desoto N r. M. L. Getchell, Wayne State School	8:30 - 161	the Chorda Tympani Nerve. R. J. DELANY AND S. D. ROPER (Department
8:00 - 151	DONALD A. LEOPOLD, DAVID E. HORNUNG, MAXWELL M. MOZELL, STEVEN L. YOUNGENTOB AND GEORGE R. PETRO (SUNY Health Science Center at Syracuse) The Relationship Between Nasal Anatomy and Human Olfaction.	0.50	of Anatomy, Colorado State University, Ft. Collins, CO 80523 and Rocky Mountain Taste and Smell Center, University of Colorado Health Science Center, Denver, CO 80262) Basal Cells and Synaptic Connectivity in Taste Buds of Necturus
8:15 - 152	THOMAS A. SCHOENFELD, ANDREW N. CLANCY AND FOTEOS MACRIDES (Worcester Foundation for Experimental Biology, Shrewsbury, MA 01545) Spatial Organization of Receptor Surfaces and Odorant Passageways in the Hamster Nasal Cavity: Relationship to the Spatial	8:45 - 162	THOMAS E. FINGER, MARY WOMBLE (Univ. of Colorado Med. Sch.) VAR L. ST. JEOR, JOHN C. KINNAMON (Univ. of Colorado, Boulder) AND TETSUFUMIL UEDA (Univ. of Michigan) Synapsin-like Immunoreactivity of Nerve Fibers in and around Lingual Taste Buds of the Rat.
8:30 - 153	Organization of Central Projections. M. MOZELL, P. SHEEHE, D. HORNUNG, P. KENT, S. YOUNGENTOB AND S. MURPHY (SUNY Health Science Center at Syracuse) Imposed and Inherent Mucosal Activity Patterns: Their Composite Representation of Olfactory Stimuli.	9:00 - 163	MARC C. WHITEHEAD (Dept. Oral Biology, Ohio State Univ., Columbus, OH 43210) MARION E. FRANK, THOMAS P. HETTINGER, LEIN-TUAN HOU AND HYUN-DUCK NAH (Dept. Biostructure and Function, Univ. of Conn. Farmington, CT 06032) Persistence of Taste Buds in Denervated Fungiform Papillae.
8:45 - 154	G. D. ADAMEK (Osphresiopolis, Dept. Anat. & Cell Biol., Univ. of Cincinnati) Regulation of Odorant Information Transmission Occurs within the Olfactory Nerve.	9:15 - 164	M. SCOTT HERNESS (The Rockefeller University, New York, NY 10021) Immunocytochemical Localization of Vasoactive Polypeptide (VIP) in Hamster Taste Cells.

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- 9:30 165

 JADMEET S. KANWAL AND THOMAS E. FINGER
 (Univ. of Colorado Medical School)

 Gustatory Information is Processed in

 Multiple Facio-spinal Pathways in the
 Catfish Brainstem.
- 9:45 166

 BARRY J. DAVIS AND SUSAN MOORE
 (University of Alabama at Birmingham)

 Tyrosine Hydroxylase-Like

 Immunoreactivity in the Gustatory Zone
 of the Nucleus of the Solitary Tract in
 the Hamster.

"OLFACTION: PLASTICITY AND DEVELOPMENT"

Slide Session VI A - Hernando DeSoto N Chaired by Dr. G. D. Burd, Univ. Arizona

- 10:30 167

 HARRIET BAKER (Cornell University Med.

 Coll.) Neonatal Olfactory Deprivation
 Results in a Dramatic Reduction of
 Tyrosine Hydroxylase Levels in Adult Rat
 Main Olfactory Bulb.
- 10:45 168

 V. McM. CARR, A. I. FARBMAN (Dept. Neurobiology & Physiology, Northwestern Uni., Evanscon, IL), AND J. I. MORGAN (Roche Institute of Molecular Biology, Nutley, NJ) Immunofluorescence Studies of Embryonic Development of the Rat Olfactory Bulb.
- 11:00 169

 J. H. MCLEAN AND M. T. SHIPLEY (Univ. of Cincinnati) Postnatal Development of Noradrenergic Afferents to the Olfactory Bulb.
- 11:15 170

 L. L. FRAZIER AND P. C. BRUNJES
 (University of Virginia) Unilateral Odor
 Deprivation: Changes in Olfactory Bulb
 Cell Number and Density.
- 11:30 171 P. C. BRUNJES (University of Virginia)
 Unilateral Odor Deprivation and
 Olfactory Bulb Development in the
 Precocial Mouse Acomys cahirinus.
- 11:45 172 C. J. WYSOCKI, G. K. BEAUCHAMP, H. J. SCHMIDT AND K. M. DORRIES (Monell Chemical Senses Center) Changes in Olfactory Sensitivity to Androstenone with Age and Experience.
- 12:00 173

 R. G. MAIR, C. SLADE (University of New Hampshire) AND P. J. LANGLAIS (Harvard Med) Monoaminergic Activity in Olfactory Bulb and Cerebellum During Olfactory and Motor Learning Tasks.

"TASTE: INTAKE REGULATION AND PSYCHOPHYSICS"

Slide Session VI A - Hernando Desoto S Chaired by Dr. T. Scott, Univ. Delaware

- 10:30 174 STEPHEN W. KIEFER, GLORIA J. LAWRENCE
 AND KARRY L. SCHWEIGER (Kensas State
 University) The Bitter-Sweet Taste of
 Alcohol: Aversion Generalization to
 VArious Sugar-Quinine Mixtures in the
 Rat.
- 10:45 175

 RHONDA OETTING DEEMS AND MARK T.

 FRIEDMAN (Monell Chemical Senses Center,
 Phila., PA) Preference for NaCl
 Solutions in an Experimental Model of
 Liver Disease: The Bile Duct Ligated
 Rat.
- 11:00 176 ROBERT J. HYDE (San Jose State University, San Jose, CA 95192) Anterior vs. Posterior Tongue: Taste and Hedonic Responses Differ.

- 11:15 177

 J. E. STEINER AND A. STEINBERGER
 (Department of Oral Biology, "HADASSAH"
 Faculty of Dental Med., The Hebrew
 University, Jerusalem (Israel) P. O. Bo.
 1172) Multidisciplinary Measures of
 Taste Hedonics.
- 11:30 178

 J. GRINKER AND K. BOSE (University of Michigan) Infant Sucking Responses to Sweet Solutions Reconsidered.
- 11:45 179

 GARY K. BEAUCHAMP, MARY BERTINO (Monell Chemical Senses Center) AND KARL
 ENGELMAN (Clinical Research Center,
 Hospital of the University of
 Pennsylvania) Factors Responsible for
 Changes in Salt Taste Preference
 Following Alterations in Dietary Sodium
 Intake in Human Adults.
- 12:00 180 RICHARD D. MATTES, CAROL M. CHRISTENSEN (Monell Center) AND KARL ENGELMAN (Hospof the Univ. of Penna.) Heightened Salt Taste Sensitivity in Amiloride-Treated Normotensive Adults.
- 12:15 181 STEVEN KELLING (Dept. of Psychology and Field of Physiology) AND BRUCE HALPERN (Dept. of Psychology and Section of Neurobiology and Behavior, Cornell University, Ithaca NY 14853) Taste Intensity Tracking of Aqueous Square Wave Trains.

2 Odor Detection Ability Decreases With Age.
DANIEL A. DEEMS & RICHARD L. DOTY (Smell and Taste Center, University of Pennsylvania)*.

ability to recognize and identify odors markedly declines in later life. For example, administration of the University of Pennsylvania Smell Identification Test (UPSIT) to 1,955 men and women indicated that more than half of those over 65 years have considerable difficulty identifying odors (Doty et al., Science, 1984, 226, 1441-Although there is evidence that olfactory sensitivity, per se, is also decreased in the elderly, detection threshold studies have not evaluated all age groups. Thus, the nature and degree of olfactory threshold changes across the entire age range have been, until the present time, unknown. Using a single staircase forcedtest we have demonstrated that choice detection thresholds for pheny ethyl alcohol -- an odorant relatively free of trigeminal stimulative properties -- decline with age in a manner analagous to UPSIT scores. As with odor identification, large individual differences were observed, and men evidenced an average earlier decline in detection ability than women.

*Suppoted by National Institute of Neurological and Communicative Disorders and Stroke Grant NS 16365.

1 Characteristics of Chemosensory Patients and a Comparison of Olfactory Assessment Procedures.
DAVID V. SMITH, ROBERT A. FRANK, MYLES L. PENSAK and ALLEN M. SEIDEN (Taste and Smell Center, University of Cincinnati Medical Center)

Patients presenting to the University of Cincinnati Taste and Smell Center with a primary complaint of chemosensory dysfunction were assessed using a battery of psychophysical procedures, mostly adopted from other existing centers. In addition, patients underwent thorough otolaryngological examination and, when necessary, neurological, dental, or radiological evaluation. Of the initial 54 patients, 79.6% were found to have olfactory dysfunction, with 25 showing anosmia and 18 hyposmia. Of these 43 patients, olfactory loss was attributable to: head trauma (12 patients), post-viral infection (10), nasal or sinus disease (6), idiopathic (5), congenital (4), multiple causes (4), toxic exposure (1) and malingering (1). The number of head injury cases primarily reflects attorney referrals. The majority of the other patients were referred by physicians and the remainder were mostly self-referred. Taste loss was seen in 29.6% of the patients, with 1 patient showing ageusia, 3 partial ageusia, and 12 hypogeusia. Taste losses were attributable to: head trauma (5 patients), toxic exposure (2), peripheral nerve damage (2), post-viral infection (2), stroke (1), medication (1), multiple causes (1), idiopathic (1) and malingering (1). Dysgeusia was seen in 3 additional patients and was associated with hypogeusia and/or hyposmia in 5 others. Olfactory function was measured using the University of Connecticut battery (Butanol and Odor ID) and the University of Pennsylvania Smell Identification Test (UPSIT). Across 50 patients, the UPSIT correlated +0.93 with Odor ID, +0.73 with Butanol, and +0.90 with their composite. Butanol scores were typically smaller than the corresponding Odor ID scores, with a correlation of +0.78.

3 Isolated Deficit for Recognition of Airborne
Chemosensory Stimuli in Early Alzheimer's Disease.
J. WEIFFENBACH (National Institute of Dental Research),
E. KOSS (National Institute on Aging).

Histological and neurochemical findings in Alzheimer's disease (AD) patients have suggested that "olfactory pathways may be the site of initial involvement in the disease." The corollary, "olfactory deficits may be one of the earliest symptoms", is supported by preliminary reports of impaired olfactory recognition. The nature and time of onset of these deficits have yet to be established.

Ten males in the earliest stages of AD were screened to exclude coexisting medical or psychiatric conditions. They were compared to ten similarly screened healthy men on multiple chemosensory tasks. Patients unable to perform 2 and 4 alternative forced choice visual tasks analogous to the taste and smell evaluations were rejected. Patients obtained significantly lower scores than controls on the University of Pennsylvania Smell Identification Test. Differences for the 4 items primarily dependent on trigeminal stimulation and for the 36 primarily olfactory items were each significant. In contrast, the olfactory detection thresholds for pyridine obtained by patients and controls did not differ. No significant differences were observed for taste detection thresholds for either sucrose or citric acid. For neither tastant did patients differ from controls on measures of intensity judgment (slope, repeatability, monotonic trend).

Our patients, in the earliest stages of AD, demonstrated clearly impaired recognition of airborne chemosensory stimuli but normal olfactory detection, taste detection and taste intensity judgments. The isolated recognition deficit suggests that the initial chemosensory impairment in AD is central rather than peripheral. While our data suggest that the olfactory nerve is not the "site of initial involvement," they do not rule out the possibility that it is the route of entry for an exogenous agent causing the disease.

4 Odor Detection Performance of Rats Following d-Amphetamine Treatment: A Signal Detection Analysis. Richard L. Doty and Mark Ferguson-Segall (Smell and Taste Center, University of Pennsylvania)

The effects of d-amphetamine sulfate (0.2, 0.4, 0.8, and 1.6 mg/kg sc) on the odor detection performance of sixteen adult male Long Evans rats was assessed using high precision olfactometry and a go/no-go operant signal detection task. Drug and saline tests were administered every third day in a counterbalanced order. Relative to saline tests, enhanced detection performance to the target stimulus (ethyl acetate), as measured by a non-parametric signal detection index (SI), occurred following administration of 0.2 mg/kg of the drug, whereas decreased detection performance occurred following administration of 1.6 mg/kg of the drug. Although the responsivity index (RI) was not significantly altered at any drug dosage when calculated for the entire 250 trials of a test session, analysis of this measure at each odorant concentration (10°0.0, 10°5.5, 10°5.0, 10°4.5 and 10°4.0 relative to saturation) revealed significant increases at the 10°5.5 concentration for the 1.6 mg/kg dosage and at the 10°6.0 concentration for both the 0.8 and 1.6 mg/kg dosages. In addition, small but statistically significant increases in the latency to respond in the presence of the odor (i.e., S+ response latency) were observed at the 0.8 and 1.6 mg/kg dosages. Overall, these data suggest that (a) odor detection performance is enhanced by low doses of amphetamine, (b) odor detection performance is decreased by moderate doses of amphetamine, and (c) drugrelated alterations in response criteria occur following the administration of moderate doses of amphetamine.

Supported by National Institute of Neurological and Communicative Disorders and Stroke Grant NS 16365.

Effects of HCL on Taste: Taste and Bulimia. L. BARTOSHUK (J.B. Pierce Foundation Laboratory), M. FRANK, T. HETTINGER (University of Connecticut), C. PETERSON, V. RAMIREZ, J. RODIN (Yale University).

Bulimics who purge expose their tongues and palates to the HCl in vomit (.001 to .1 M). Since this HCl can erode teeth as well as enlarge salivary glands and irritate oral tissue, one might expect to find damage to taste receptors in bulimics.

HC1 might also cause temporary taste changes short of damage. Frank and Hettinger (1983) found that a 10-30 seconds exposure of the hamster tongue to .01 to .1 M HC1 reduced responses to sucrose, enhanced responses to KC1 and HC1 but had little effect on responses to NaC1.

We examined taste function in 7 bulimic women. Al-

We examined taste function in 7 bulimic women. Although taste function was normal when evaluated by a whole mouth "sip and spit" test, localized testing (solutions were painted on the front, rear edges and palate with long handled Q-tips) showed a reduced ability to taste HCl and QHCl on the palate.

We exposed the anterior of the tongue of 10 non-bulimic volunteers to HCl (.0018 to .032 M) for thirty seconds (tongues were dipped into cups of HCl). Taste stimuli were then "painted" on the tongue tip within the area exposed to HCl. The HCl exposure reduced the sweetness of sucrose and the bitterness of quinine, but had little effect on the saltinesses of NaCl and KCl and the sourness of HCl. Effects lasted about 15 minutes.

We conclude that bulimics may experience some temporary taste loss immediately after purging as well as chronic taste loss on the palate.

We thank NIH Grants #NS 16993 and 21600

NOTES

6 Sucrose Taste Perception in Depressed
Patients. R.G. Settle, J.D. Amsterdam and E.
Abelman (Smell and Taste Center and Depression
Research Unit, University of Pennsylvania)*

Appetite disturbances are frequently associated with depressive illness, and some depressed patients report a craving for "sweets." 36 with major depression and 19 healthy patients rated the perceived intensity pleasantness of seven concentrations of sucrose (0.04 to 2.4 M) and deionized water using 10 cm visual analog scales. Patients rated the higher concentrations of sucrose as significantly less intense and more pleasant than did controls. factors were related to the differences pleasantness ratings between patients controls. 1) A higher proportion of patients preferred the two highest sucrose concentrations to lower concentrations than did controls (28%) 2) The pleasantness ratings of patients a craving for "sweets" (58% of were more similar to controls than patients reporting no craving for vs. 5%). reporting patients) those of In addition, a higher proportion of craving "sweets" reported a recent "sweets." patients increase in appetite (40%) and a lower proportion reported a decrease in appetite (25%) than did patients not craving "sweets" (7% and 67%, The relationship among sucrose respectively). pleasantness ratings, craving for "sweets" and in appetite is interesting since change improvement in a patient's appetite has been reported to be one of the earliest and most indications of recovery from depressive reliable illness.

* Supported in part by NINCDS grant NS 16365.

Odorant Identification as a Function of Concentration. H.N. WRIGHT (Clinical Olfactory Research Center and Department of Otolaryngology, State University of New York Health Science Center at Syracuse)

A 10 x 10 closed set odorant confusion matrix was presented to two normal control subjects over a 6step decreasing concentration series (log) to determine whether an anticipated decrease in percent correct identifications would be accompanied by an increase in the parosmic substitute identification of one odorant for another. The matrix was replicated five times at each concentration for each subject, resulting in 50 observations/cell. It was hypothesized that since odorant identification is a multidimensional event, a decrease in odorant concentration would cause a disturbance among the underlying dimensions thereby leading to parosmic substitutions. Such indeed was found to be the case, demonstrating that parosmic substitution can occur in clinical populations (such as those patients with nasal airway obstruction) without a receptor deficit.

This work was supported by PHS Program Project grant NS19658 from the National Institute of Neurological and Communicative Disorders and Stroke.

7 Intensity Effects on the Odor of the Enantiomers of Carvone. Lisa M. Pike, Melvin P. Enns, David E. Hornung (St Lawrence University, Canton, NY).

The purpose of the present study was to examine how intensity impacts on the previously reported differences in the odor qualities of the two optical isomers of carvone. In Experiment I, 18 college students, naive to the psychophysical techniques of scaling odorants, were asked to use the method of absolute magnitude estimation to judge the intensity of the smell of each of seven suprathreshold concentrations of R-(-) and S-(+)-carvone. Intensity estimates given to the S-(+)-carvone were significantly larger than the intensity estimates given to the R-(-)-carvone. The psychophysical functions (log-log coordinates) describing the intensity estimates were: R-(-), y=0.63, m=0.27, r=0.96; S-(+), y=0.85, m=0.34, r=0.97.

In Experiment II, solutions of R-(-) and S-(+) were paired either on equal concentrations or on the equal "subjective estimates of intensity" from Experiment I. On a given trial there was either one S-(+) paired with two identical R-(-) solutions or one R-(-) paired with two identical S-(+) solutions. Sixteen undergraduates were asked to sniff the three solutions and then indicate which one was "different". In all situations, including those matched for subjective intensity, subjects picked the odd stereoisomer as being different from the other two.

The results of both experiments further suggest that the perception of the stereoisomers of carvone is indeed dependent on the chirality of the incoming odorant molecules.

Supported by a General Foods Corporation grant.

Susceptibility to Low Renin Hypertension. FAY FERRELL and ANNETTE Z. DREITH (Dept. of Nutrition, Univ. of California, Davis, CA 95616).

Enhanced Ca appetite has been shown in the spontaneous hypertensive rat, which has lower serum Ca and higher urinary Ca than its WKY control, and responds to Ca supplementation with decreased BP. The HTN of the Dahl salt sensitive (S) rat differs from that of SHR. Dahl S has low renin activity, developing HTN quickly when fed high NaCl, and eventually becoming HTN regardless of diet. High dietary Ca lowers BP in Dahl S providing that NaCl is not excessive. Low renin HTN humans, who also respond to high excessive. Low renin film numers, who also respond to fight dietary NaCl with increased BP, reportedly have low blood Ca levels, with BP lowered by Ca supplementation. Dahl S and their NTN salt-resistant (Dahl R) controls allow comparative studies-in animals with HTN of differing etiologies-of appetite for minerals involved in BP regulation, and provide a rat model for human low renin HTN. Following group assignment based on baseline BP, 11 Dahl S and 11 Dahl R rats fed Ca replete diets were offered ascending concentrations of 0.001-0.1M CaCl, or Ca Lactate vs. water. Intakes of each solution were measured in two 48-hr 2-bottle tests. Dahl S exhibited higher preference ratios than Dahl R for both CaCl₂ and Ca Lac, and ingested more mEq of each per 100 g BW. Post test BPs of CaCl₂ and Ca Lac rats did not differ from each other or from unexposed ctrls, nor was MEq Ca in either form consumed by individual rats related to final BP or increase over baseline in BP. The heightened Ca preference and intake seen in Dahl S was of greater magnitude than we observed earlier in SHR over WKY. In SHR, in contrast, CaCl2 exposure via preference tests was associated with lower BP. Results suggest that Ca appetite differs not only between various HTN rat models and their NTN ctrls, but between rats with different etiologies underlying their HTN. Furthermore, efficacy of Ca (ingested in amounts consumed in 2-bottle tests) in lowering BP varies between SHR and Dahl S. In the latter, higher Ca levels, such as would be received via <u>dietary</u> supplementation, appear necessary to produce a significant beneficial effect on BP.

There is neuroanatomical evidence for damage to entorhinal cortex in Alzheimer's Disease. Recent reports have shown a decline in the ability to identify odors in patients with the disease (Doty, ISOT, 1986). Increased olfactory threshold might be expected from the neuroanatomical data, but has not been demonstrated. The present study investigated memory for odors and visual stimuli in ten patients who met the criterion for probable Alzheimer's Disease. Recognition memory was explored for various types of stimuli: common odors, faces of American presidents and vice-presidents, and electronic symbols. Ten of each were first presented in intermixed order for inspection and then subsets of five of each were presented again, along with an equal number of distractors, for recognition ten minutes later. The Alzheimer's patients' performance was compared to that of 16 young (mean age = 21 years) and 16 elderly (mean age = 72 years) persons whose performance in the same task (with twice as many trials) has been reported (Cain and Murphy, ISOT, 1986). To control for differences in the subjects' criterion bias, Analysis of Variance was conducted on the subjects' Az scores. Az scores are computed from hits, misses, false alarms and correct rejections. The patients with Alzheimer's Disease scored significantly lower than controls on all tasks and, showed a greater decrement in odor memory than in visual memory. Young subjects were equally facile with odors, faces, and symbols. Olfactory thresholds were determined on all subjects using a two-alternative, forced-choice, ascending series with butanol. When the subject was incorrect in his choice, concentration was increased. When he was correct, the same concentration was presented, to a criterion of four correct in a row. Results for odor memory will be discussed in light of information about olfactory threshold. This study suggests significant impairment in olfactory memory in patients with Alzheimer's Disease. This impairment is distinguished by its severity from that seen for odor memory in normal elderly persons and for visual memory in Alzheimer's patients.

Supported by NIH grant # AG04085to C.M. We thank Drs. R. M. Katzman, B. Lasker, and D. Salmon for access to patients who have been evaluated for Alzheimer's Disease at the UCSD Alzheimer's Disease Research Center.

1 1 Increased cerdiovescular reactivity in adult Spreaue-Dewley rats exposed per instally to a high salt diet. ROBERT J. CONTRERAS (University of Alabama et Birmingham, Department of Psychology, Birmingham 35294).

Dietary NaCl intake is known to be a factor in the development of hypertension. While this relationship has been accepted for many years, the mechanism by which excess dietary NaCl influences blood pressure is unknown. In this regard, our research program has focussed on the effects of early dietary NaCl on the blood pressures and taste preferences of adult rats and the mechanisms thereof.

Adult female rats consumed a diet containing either low (.12%), mid (1%), or high (3%) amounts of NeCl throughout pregnency and lactation. The offspring were kept on these same diets until 30 days of life, at which point, they were given a 1% salt diet. At 100 days of age, blood pressure was recorded directly from the cerotid entery of ewake, unrestrained rats. Baseline unstimulated blood pressure was recorded for 20-min; this was followed by two 60-min per iods in which we measured cardiovascular reactivity defined by the blood pressure response first to angiotensin II (100 ug/kg BW), and then to isoproterenol (15ug/kg BW).

We discovered that the cardiovascular reactivity measures uncovered differences between the animals raised on the low and high solt diets that were undetected with beseline blood pressure measurements. The baseline blood pressure levels of both groups were slightly less than 140 mm Hg. However, the high salt rats were more responsive to both the pressor effects of angiotensin II and the depressor effects of isoproterenol compared to the low selt rats. After angiotens in II, the blood pressure response of the high salt rats peaked about 200 mm Hg and took longer than 30-min to return to beselfne; the blood pressure response of the low salt rats peaked at about 170 mm Hg and returned to baseline 30-min after injection. Similarly after isoproterenol, the high selt groups' blood pressure dropped to a low of 105 mm Hg and took longer than 30-min to return to baseline; the low selt groups' dropped only to 120 mm Hg and returned to beseline 30-min after injection. These results were obtained after the imposition of an intervening period when the animals of both groups were fed the same 1% NaCl diet for two months. Furthermore, the results were obtained in rets with little genetic bies to hypertension, illustrating the fact that a relatively benign dietary manipulation may lead to enduring changes in cardiovascular function.

This research was supported by NIH, NHLBI grant HL38630.

NOTES

Age-related Differences in Short Term Memory for Olfactory and Visual Stimuli. R. BLAIR SKINNER, FRED E. ROSE, MAGDALENA M. JENSEN, CLAIRE MURPHY* (San Diego State University) and WILLIAM S. CAIN (Pierce Foundation, Yale University)

Recent work has demonstrated age-associated differences in long term memory for odors and graphic stimuli (Cain & Murphy, ISOT, 1986). We sought to determine whether such differences would also occur in short term memory (STM), thus suggesting interference in the encoding of odor memory. A total of 53 persons participated: 12 females aged 18-21 (M = 19), 13 males aged 18-25 (M = 20), 13 females aged 63-82 (M = 70), and 15 males aged 64-82 (M=73). All were active, community-dwelling persons with no hospitalizations within the past year. Short term memory was investigated for three types of stimuli: odors with commonly-known labels, odors with chemical names, and free-form visual line drawings. Ten odors and ten visual stimuli were presented to each subject with the provision that each subject receive three odors with chemical names. Upon presentation, subjects first rated familiarity of the stimulus. Twenty-six seconds later, they were presented with either a stimulus which had been previously presented (old) or one which had not (new). Az scores were computed from hits, misses, false alarms and correct rejections and these Az scores were subjected to multifactor ANOVA. Results indicate a significant effect of age; but, unlike our earlier work with long term memory, no interaction between age and type of stimulus. Elderly subjects had more difficulty regardless of stimulus type. The existence of a verbal label did not seem to differentially facilitate encoding in STM for young and old. It remains an interesting question at which point in the memory process verbal encoding becomes important for odor recognition memory performance.

Supported by NIH grant # AG04085 to C.M.

13 Suprathreshold Evidence for Quality-Specific Aging in the Taste System, MAGDALENA M. JENSEN (San Diego State University), and CLAIRE MURPHY (San Diego State University)*

Age-associated decrements in the taste system at both threshold and suprathreshold have been reported. Diminished sensitivity at threshold has been demonstrated to be quality-specific with the greatest age-related decrements associated with the perception of bitter stimuli and the least for sweet stimuli (Weiffenbach et al., 1982, <u>J. of Geront., 37, 372-377</u>). The present study was designed to test quality-specific aging at the suprathreshold level by comparing the relative perceived intensities of the individual components of two-component mixtures for elderly and young subjects. For each of the following three matrices, 48 subjects participated: 12 females and 12 males 65-83 years old (M = 72.5), and 12 females and 12 males 18-31 years old (M = 20.3). To produce three matrices of 16 possible mixtures each; three concentrations of sucrose and deionized water (.15, .30, .60 M) were mixed with three concentrations of caffeine and deionized water (.0025, .005, .01M); with three concentrations of citric acid and deionized water (.0015, .0030, .0060 M); and with three concentrations of NaCl and deionized water (.10, .20, .40 M). Using the method of magnitude matching with weights (20, 50, 100, 200, 400, 750 g), and the sip and spit method, subjects rated the intensity of both components in the mixture and the umixed state. A series of Multifactor Analyses of Variance were conducted. Age-related losses were associated with the perception of bitter and sour for unmixed components, but not for salty or sweet. For isointensity mixtures, elderly subjects perceived bitter as being significantly less intense than did the young subjects. Results will also be presented for non-isointensity mixtures. Quality-specific aging at the suprathreshold level may be indicative of peripheral dysfunction in the elderly and merits further investigation.

*Supported by NIH grant #AG04085 to C. M.

Post-Traumatic Anosmia: Ultrastructural Correlates. BRUCE W. JAFEK, M.D., PAMELA M. ELLER, BARBARA A. ESSES, M.D., DAVID T. MORAN, PH.D. (University of Colorado School of Medicine)

*Supported in part by NIH Grant #1-P01-NS-20486-01, Rocky Mountain Taste and Smell Center.

Although the first reference to the loss of the sense of smell due to head injury (post-traumatic or post-concussive anosmia) was in 1864, it was rarely recognized until a major series of war injuries were analyzed, which placed its incidence at 5-10% in concussed patients.

Anosmia was postulated to be due to shearing of the fine fila olfactoria as they pass through the cribiform plate as the brain accelerates or decelerates within the calvarium. While other mechanisms were considered, the few anatomic observations were based upon the most severe injuries (post-mortem) and detailed psychophysical analyses were rare.

In order to study this type of anosmia more completely, six patients with post-traumatic anosmia were studied in depth at the Rocky Mountain Taste and Smell Center at the University of Colorado. Following detailed historical review, psychophysical testing was undertaken and olfactory biopsies obtained. These will be presented along with speculation on the pathogenesis of the anosmia, prognosis, and therapy.

Histological and Ultrastructural Changes of the Taste Bud Following Radiation: A Pilot Study. BARBARA A. ESSES, M.D., BRUCE W. JAFEK, M.D., DANIEL J. HOMMEL, M.D., PAMELA M. ELLER (University of Colorado School of Medicine)

It is a well established fact that taste dysfunction (dysgeusia) occurs following a routine course of radiation therapy to the head and neck. Several authors have attempted to demonstrate the changes which take place in response to radiation treatment.

In the study presented, mice (CH57BL6/J from Jackson Laboratories) were used as a model. They were 20-25 grams in size and each underwent radiation exposure to the head and neck region. There were three distinct groups: 1) mice given fractionated doses of radiation over the course of several days (180cGy/day at 5 day/wk.) with immediate sacrifice, 2) mice given fractionated doses of radiation with sacrifice four and seven days following therapy, and 3) mice given single dose therapy with sacrifice at different intervals following therapy. Their circumvallate papillae were harvested at the time of sacrifice.

The specimens were set up for light microscopy in all cases. They were studied for taste bud number, taste bud size, and cells per bud. The last group was also studied using electron microscopy with attention directed towards the light and dark cells. A decrease in bud number, size, and cellularity were seen as a function of time and dosage. The specific changes are reported.

Charting Odor and Irritation of Formaldehyde at Environmentally Realistic Concentrations. WILLIAM S. CAIN, LAI-CHU SEE, and TARIK TOSUN (John B. Pierce Foundation Laboratory and Yale University)

Subjects seated in an environmental chamber judged the perceived irritation and odor of formaldehyde during exposures to concentrations ranging from 0.25 to 2.0 ppm. Prominent characteristics of the sensations included growth of irritation with time for the lower concentrations and decay for the highest. This pattern proved true across the three sites of irritation (eye, nose, throat). These sites also proved roughly equivalent in sensitivity. The timecourse of irritation suggested the operation of coincident processes of potentiation and adaptation. When described in terms of two multiplicative exponential processes, the rate of potentiation was faster than the rate of adaptation by about an order of magnitude. In order to probe the existence of these simultaneous processes, an irritating stimulus of carbon dioxide was presented briefly to the eye, followed by test stimuli of lower and higher concentrations. Whereas lower concentrations were depressed in irritation, higher concentrations were enhanced, thereby revealing that adaptation and potentiation could occur coincidentally. In an experiment on possible interactions between odor and irritation, the odorous substance pyridine was injected into an environment containing I ppm formaldehyde. The irritation from the formaldehyde decreased. Such sensory interactions may also occur in environmentally realistic situations.

This research was supported by grants #ES00354 and #ES00592 from the National Institute of Environmental Health Sciences.

7 The Effect of an Antidepressant Drug, Amitriptyline, on Growth of Axons from Olfactory and CNS Neurons in Explant and Cell Cultures. FEDERICO GONZALES AND ALBERT I. FARBMAN (Dept. of Neurobiology, Northwestern Univ., Evanston, IL 60201).

Amitriptyline is a commonly prescribed drug used in the treatment of depression. We have recently observed (Farbman et al., Soc. Neurosci. Abstr., 1986) that this drug inhibits the growth of neurites in explant cultures of chick embryo olfactory epithelium. In this study, we report that In this study, we report that amitriptyline has an identical effect on cell cultures of the central nervous system of chick embryos and on neurons from olfactory epithelium and CNS of mammalian (rat) embryos as well. Fragments of E15 rat olfactory epithelium or olfactory bulb were explanted in tissue culture in the presence of 0 to 16.0 µM amitriptyline. After 2 and 4 days, the cultures were examined and evaluated for neurite outgrowth. Inhibition of neurite outgrowth was seen at concentrations of 2.0 µM and higher. A similar dose inhibited neurite outgrowth from chick CNS neurons that had been dissociated and plated in cell culture. The results indicate that amitriptyline is toxic to growing neurons in doses lower than those considered toxic to healthy adult humans. The observations have special significance in olfaction because receptor neurons are continually produced in adults and a portion of this cell population would be growing axons at any given time.

Supported by NIH grant #NS 06181.

NOTES

Impairment of Olfactory Function in Workers
Chronically Exposed to Cadmium Fumes. CECILE S.
ROSE, PETER G. HEYWOOD, and RICHARD M. COSTANZO
(Medical College of Virginia, Richmond, VA 23298).

Workers exposed to cadmium fumes are at increased risk for chronic kidney and lung damage. These disease effects are related to the cumulative body burden of cadmium as measured by the urinary concentration of cadmium. The presence of low molecular weight proteins in urine (beta-2-microglobulinuria) is the first sign of cadmium-induced end organ damage. A few studies suggest cadmium-related effects on olfaction. However, olfaction deficits have never been quantified as a function of cadmium body burden. We administered a standardized quantitative test of olfactory function to a group of factory workers (n=44) with high and low cadmium body burdens as measured by urinary cadmium levels. Cadmium damage to the urinary cadmium levels. Cadmium damage to the kidney was measured by the presence of beta-2microglobinuria. No other significant occupational exposures occurred in this population. Our results revealed that the cadmium-exposed workers had significantly lower olfactory function scores compared to a control population. The workers with high urinary cadmium levels (>10 ug/L) and evidence of cadmium-induced beta-2-microglobulinuria (beta-2 >370 ug/L) had significantly lower olfactory function scores (p<.01) compared with those workers with lower urinary cadmium levels. These results provide the first quantitative data demonstrating significant impairment of olfactory function following chronic exposure to cadmium fumes.

Supported by VCU Grants-in-Aid to CSR and RMC.

A Possible Role for Ca** in turnover of taste cell membrane binding sites for the sweet protein, thaumatin. ALBERT I. FARBMAN, LYNN M. COLLETTI (Dept. of Neurobiology, Northwestern Univ., Evanston, IL 60201), GÖRAN HELLEKANT, R. ALBRECHT AND H. VAN DER WEL, (Dept. of Veterinary Sciences, Univ. of Wisconsin, Madison, WI. 53706).

recently shown that when the sweet protein, We have thaumatin, is conjugated to colloidal gold and applied to the tongue of Rhesus monkeys, gold particles can be visualized on the microvillus tips of Type II cells of foliate taste buds, and on small (50-100 nm diameter) membrane bounded blebs in the pore. We proposed that these blebs are released from the cell as an apocrine secretion and represent a means by which the cell rids itself of some stimulus-binding site complexes (Farbman et al., Scanning Microscopy, 1:351-357, 1987). In the present study, we have examined the effect of pre-rinsing the tongue surface (for 30 minutes) with a calcium-free Ringer solution on the stimulus-induced production of blebs in monkey foliate and vallate taste buds. We used thaumatin and sucrose solutions as stimuli, immediately after the rinse. In control (no rinse, no stimulus) and in sucrose-stimulated taste buds, only 20% of pores contained small blebs. In thaumatin stimulated buds, that were not pre-rinsed with Ca^{++} -free Ringer, 90% of the taste pores contained the blebs. After pre-rinsing with Ca++-free solution, stimulation with thaumatin produced blebs in only 8% of taste pores. Prerinsing with amiloride, a sodium channel blocker, had no effect. The results suggest that the production of small (50-100 nm) blebs in the taste pore is a response of taste cells to thaumatin, but not to sucrose, and support the suggestion that they may be involved in ridding the cell of large molecular stimuli. Blocking the sodium channels has no effect on this phenomenon, but depletion of external Ca** inhibits it. It is possible that external Ca** is important in the taste cell response to protein sweeteners.

Supported by NIH grant #NS 17021

20 <u>Ligand Binding Specificity of the Olfactory L-Alanine</u>
Receptor of Channel Catfish. R. DINO RULLI and RICHARD C.
BRUCH (Monell Chemical Senses Center)

Neurophysiological cross-adaptation studies have shown that odorant L-amino acids for the channel catfish (Ictalurus punctatus) can be classified into four groups, corresponding to acidic, basic, shortchain neutral, and long-chain neutral amino acids (Caprio and Byrd (1984), J. Gen. Physiol.). The short-chain neutral amino acid L-alanine is a potent olfactory stimulus for the catfish. The ligandbinding specificity of the olfactory L-alanine receptor site in isolated cilia preparations was therefore investigated by competitive binding methods. Approximately 40 amino acids and derivatives were tested for their ability to compete with radiolabeled L-alanine for common binding sites. Acidic and basic L-amino acids, and imino acids, did not compete effectively for the L-alanine binding site. Several neutral L-amino acids, including glycine, leucine, serine, threonine and cysteine, competed as effectively as L-alanine itself for the receptor. Although long-chain neutral L-amino acids, such as norleucine, methionine and phenylalanine, competed nearly as effectively as L-alanine itself, a significant (p < 0.002) difference was obtained between long-chain and shortchain neutral ligands. L-Alanine derivatives containing a blocked amino (e.g., N-acetyl) or carboxyl group (e.g., amides and esters) were less effective competitors than L-alanine. In combination, these results are consistent with previous electrophysiological data in catfish and support the previously defined receptor classes. These results are also consistent with previous structure-activity relationships determined electrophysiologically that indicate a requirement for unblocked amino and carboxyl groups for maximal electrophysiological potency.

This work was supported by BRSG S07-RRG5825-07 from the Biomedical Research Support Program, NIH and a grant from the Veterans Administration (to J.G. Brand).

21 Calmodulin Blockers and Chemosensory Responses in the Cilate, Tetrahymena. M. LEVANDOWSKY and DELMA BRATWOLD (Haskins Labs of Pace University, N.Y., NY 10038)

Swimming behavior in ciliate protozoa has been shown to be largely controlled by a calcium-dependent membrane potential, and internal calcium levels. We have shown previously that the chemosensory response of the ciliate Tetrahymena is also calcium-dependent. Since many calcium-dependent cellular processes involve the calcium-binding protein, calmodulin (CAM), we studied the effects of several groups of CAM-blooking agents. Both the pheothiazines (chlorpromazine, trifluoperazine) and the naphthalene sulfonamides (W7) inhibit chemosensory responses at levels where other effects on swimming behavior are not apparent. These two groups have relatively little in common other than their antagonism to CAM. Furthermore, the naphthalene sulfonamide W5, structurally very similar to W7 but not a CAM blocker, did not inhibit chemosensory responses.

Voltage-Dependent Ionic Currents in Isolated Taste Receptor Cells of the Tiger Salamander. KUMIKO SUGIMOTO (Monell Chemical Senses Center) and JOHN TEETER (Monell Chemical Senses Center and University of Pennsylvania)

Action potentials have been recorded from mudpuppy taste receptor cells in response to depolarizing currents (Roper, 1983) and certain taste stimuli (Kinnamon et al., 1985). The significance of regenerative responses in taste transduction, however, remains unclear. Taste cells of larval tiger salamanders also generate impulses and we have used the whole cell configuration of the patchclamp technique to partially characterize voltage-dependent ionic currents in taste receptor cells isolated from this species. Taste cells were dissociated from isolated pieces of lingual epithelium using enzyme (0.5 mg/ml collagenase and 0.5 mg/ml hyaluronidase) and EGTA (2 mM) treatments and plated on plastic culture dishes in normal Ringer. Isolated taste cells usually displayed both a transient inward current and a delayed outward current in response to depolarizing voltage steps from a holding potential of -60mV. The transient inward current activated at -20 to -30 mV and reached peak amplitude at 0 to 10 mV, with a latency of about 0.6 ms. This current was completely eliminated by substitution of choline † for in the bath solution or by the addition of 1 µM TTX, indicating that this was a Na+-current. The delayed outward current activated at -40 to -30 mV, reached peak amplitude in several ms and usually displayed some inactivation within the 25 ms voltage step. Most of the outward current (85-90%) was eliminated by addition of 5 mM TEA and 5 mM BaCl, to the bath, or substitution of Cs for K in the pipette solution. In some cells, the outward current was partially suppressed by addition of 2 mM CoCl2 to the bath. These results indicate that most of the outward current was carried by K and that some of the K current was activated by Ca -influx. In a few cells, a slow long-lasting inward current was observed at membrane potentials more depolarized than -30 mV and may have represented a Ca⁺⁺-current. These results suggest that there are at least four types of voltage-dependent ionic channels in tiger salamander taste cells; two types of K+-channels, Na+-channels and -channels. Although the functional significance of the observed macroscopic currents remains to be elucidated, it is likely that they control the excitability of the taste cells.

This work was supported by NSF Grant BNS-8609555 and BRSG S07-RRG5825-07 from the Biomedical Research Support Program, NIH.

The molecular ontogeny of pheromone reception in the gypsy moth, <u>Lymantria dispar</u>. VOGT,R.G. (SUNY, Stony Brook); KOEHNE,A.C. (University of Wurzburg, FRG); DUBNOW,J. (SUNY, Stony Brook); PRESTWICH,G.D. (SUNY, Stony Brook).

A male gypsy moth antenna possesses about 40,000 sensory hairs, each containing two neurons specifically sensitive to opposite optical enantiomers of the epoxide C1s-7,8-epoxy-2 methyloctadecane. The sensory hairs contain two pheromone binding proteins (PBP) (15,000 daltons) which show 50% sequence homology to each other. These proteins show cross reactivity to antisera prepared against PBP of Antheraga polyphemus and, in combination, show 50% sequence homology with the A. polyphemus PBP. The sensory hairs also possess strong epoxide hydrolase activity, unique to the antennae, which degrades the pheromone to its corresponding three 7,8-diol.

Adult development occures during the pupal phase, requiring approximately 10 days. We have established criteria for selecting animals at specific, visually recognizable stages during the last half of adult development. We have learned, for example, that the PBPs first appear on day -3 before adult eclosion, and are fully expressed by day -2. In this poster we will present data on the electrophoretic profiles of male and female antennae during the final 5 days of adult development. We will also present data on the ontogeny of hydroxylase activity, and on the time course of transcript availability (mRNAs) for the PBPs. Structural analysis of the pre- and post-processed forms of the PBPs will also be presented.

Support from USDA 85CRCR11736 (GDP) and Deutscher Akademischer Austauschkienst (DAAD) (ACK).

NOTES

24 Use of Covalently Crosslinked Ligand and Antiligand Antibody to Identify Chemoreceptors in Paramecium. J. MICHAEL SASNER (Univ. of Vermont) and JUDITH VAN HOUTEN (Univ. of Vermont)

Folic acid is specifically bound by paramecia and this binding has been shown to correlate with chemoresponse. Two membrane proteins that fit criteria for chemoreceptors have been identified by affinity chromatography, Con-A sepharose chromatography, and 1251 surface labeling. We are now using covalent crosslinking of an n-hydroxy-succinimide derivative of folate to its receptor as another method of identifying candidates for the receptor. This method will circumvent the problems of applying affinity chromatography to low affinity binding proteins. Cells crosslinked with folate show no attraction to folate (I che = 0.54 +/- 0.06 whereas control is 0.72 +/- 0.06) and normal attraction to acetate (0.61 +/- 0.06 whereas control is 0.67 +/-0.06). Therefore, the folate chemoreceptor should be among those proteins crosslinked. We have produced polyclonal anti-folate antibody and are now using this to locate folate cross-linked proteins of the membrane. The antifolate antibody is made against KLH-folate as immunogen and purified using protein A and folate affinity chromatography. The antibody is more reactive to KLH-folate than to KLH alone and more to BSA-folate than to either BSA or BSA-glycine in ELISAs. Therefore, the crosslinker is not recognized as an epitope. Immunodevelopment of nitrocellulose blots of cross-linked membrane proteins identifies four proteins of the same molecular weight as the folate binding proteins identified by affinity chromatography, including the two candidates for chemoreceptor. The immunodetection of blots is being pursued, but to improve sensitivity, we presently are examining $^{35}\mathrm{S-labelled}$ crosslinked membrane proteins that immunoprecipitate with the antifolate antibody. The precipitation is both antibody and protein concentration dependent.

25 Cyclic AMP and Chemoreception in Paramecium. JUDITH VAN HOUTEN (Univ. of Vermont), ROBIN R. PRESTON (Univ. of Vermont), MICHAEL GAGNON (Univ. of Rochester), and NARK WRIGHT (Univ. of Vermont)

Cyclic AMP figures into the behavior of Paramecium as i) an external chemoattractant and as ii) a second messenger in the control of ciliary beating (Bonini et al., Cell Motil & Cytoskel. 6:256, 1986; Schultz et al., FEBS Lett. 176:113, 1984). i) Paramecia have low affinity, saturable cAMP binding sites that are likely to include chemoreceptors (Smith et al., Biochim. Biophys. Acta in press). Both cyclic AMP binding and chemoresponse are inhibited by 5'AMP but not by cGMP. Externally applied cAMP hyperpolarizes the cells. Hyperpolarization is known to increase ciliary beating frequency and could account for the chemoresponse swimming behavior. The hyperpolarization does not require cilia; deciliated cells show normal membrane potential response and the large majority of the binding sites. There is one major cAMP binding protein of the cell body membrane and binding of this protein to cAMP affinity columns is inhibited by 5'AMP but not by cGMP. This protein of 48 kd is not immunologically related to known fragments of the large surface glycoprotein of Paramecium and it is missing in a cAMP chemoresponse mutant. ii) Hyperpolarization induced by changes in external K or Ca causes a 2-3 fold increase in internal cAMP (Bonini et al., 1986; Schultz et al., 1984). Assays of cAMP show no increase of nucleotide when cells are hyperpolarized by attractants although they show a 2 fold increase, as expected, with the same size hyperpolarization in low K. Therefore, there is the interesting possibility that the hyperpolarizations measured by microelectrodes in attractant or low K do not have identical effects on a potential dependent adenylate cyclase and that the ionic mechanism of the hyperpolarization matters in the modulation of adenylate cyclase activity.

Supported by NSF and Whitehall Fdn.

In Vivo and In Vitro Studies of the Localization of Antiqens recognized by Monoclonal Antibodies Neu-4, Neu-5 and Neu-9 in Rat Olfactory Epithelium, MICHAEL S. LIDOW, ALBERT I. FARBMAN (Dept. of Neurobiology & Physiology, Northwestern University, Evanston, IL 60201) JAMES I. MORGAN and JAMES HEMPSTEAD (Roche Institute of Molecular Biology, Nutley, NJ 07110).

A panel of monoclonal antibodies (MAb) vs. rat olfactory epithelium was recently developed (Hempstead & Morgan, J. Neurosci., 5:438-449, 1985). Three MAbs of this group, Neu-4, Neu-5 and Neu-9, were strongly immunoreactive with olfactory receptor cells and their axons. This study was an attempt to determine the cytological distribution and possible function of antigens recognized by these antibodies in olfactory receptor cells, in vivo and in vitro. For the in vivo studies, olfactory mucosa of rat embryos, in the 21st and 22nd day of gestation (E21 and E22), was fixed and processed for immunocytochemical staining. For the <u>in vitro</u> studies, explants were made of El6-rat embryo olfactory mucosa, grown for two days, fixed, and processed for immunocytochemistry. MAD Neu-5 recognized a surface antigen situated along the entire surface of the receptor cells, from the apical dendritic knob and cilia to the axon terminal and growth come. Supporting cells were unstained. MAbs Neu-4 and Neu-9 selectively recognized antigens on the lateral surfaces of both receptor and supporting cells, but the dendritic knob and cilia of receptor cells and the luminal surface of supporting cells were not stained. In addition, Neu-4 and Neu-9 were immunoreactive with axonal and growth cone surfaces. All three MAbs labelled the entire surface of respiratory epithelial cells, including their apical microvilli and cilia. The data suggest that Neu-4 and Neu-9 stain antigens that are probably not important in stimulus reception because they are absent from the dendritic knobs and cilia. All three antigens may be important in cellular interactions in olfactory epithelium.

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27 Structure-Activity Relationships in the Arginine Receptive
Taste Pathways of the Channel Catfish, Ictalurus punctatus.
BRUCE P. BRYANT (Monell Chemical Senses Center) and JOSEPH
G. BRAND (Monell Chemical Senses Center and Veterans
Administration)

Separate transduction pathways exist for alanine and arginine in the taste system of the channel catfish (Caprio, 1982). To characterize the molecular-structural requirements for activation or antagonism of the arginine pathway, structure-activity studies were performed using integrated multiunit responses and cross-adaptation. Of all the guanidinum-containing compounds tested, only L-arginine, L-AGPA α-amino-β-guanidino-propionic acid), and L-arginine methyl ester were strong stimuli with L-arginine being the most effective. Results of functional group substitutions and modifications of the arginine parent molecule indicate the following: 1) stereospecificity is observed with D-arginine being a much less effective stimulus than L-arginine; 2) an L-amino group must be present and unblocked. a-Chloro-γ-guanidino valeric acid and N-acetyl L-arginine were weak stimuli; 3) a free carboxylic acid group is not necessary for stimulatory activity. Certain esters are active stimuli; 4) the distance between the amino acid and the guanidinium group is not critical. L-AGPA, having 2 methylene groups less than arginine is a strong stimulus; 5) modification or substitution of the guanidinium group (L-citrulline, L-ornithine and L-lysine) result in the loss of stimulatory efficacy. Cross-adaptation experiments indicate that within the arginine analogs tested, the capacity to cross-adapt responses to L-arginine is proportional to their stimulatory effectiveness in multiunit preparations. These studies provide a basis for the design of inhibitors against the arginine taste receptor binding site.

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28 Monoclonal antibodies directed against catfish taste receptors: immunocytochemistry of brain, olfactory epithelium and oral taste buds. JOAN YONCHEK, and THOMAS E. FINGER (Univ. Colorado Medical School, Denver, CO).

A monoclonal antibody (G-10) raised against a plasma membrane fraction of catfish taste epithelium appears to bind to the alanine receptor protein (Goldstein & Cagan, 1982). Previous studies (Yonchek, et al, 1986) utilizing this antibody have shown that the antigen is present within taste receptor cells of the barbel epithelium. The present experiments examine the distribution of G-10 immunoreactivity elsewhere in the fish. Fixed tissue was obtained from the brain, olfactory mucosa, and oral cavity as well as from the barbels. Following sectioning, the tissue was exposed to the G-10 antibody, or to a non-specific mouse ascites fluid. All tissues then were prepared for standard peroxidase-antiperoxidase staining. The control ascites fluid never produced specific immunostaining. The G-10 antibody routinely stained barbel taste buds as well as cells in other tissues. Taste buds on the palatal organ stained similarly to taste buds on the barbels. Each taste bud contained immunoreactive receptor cells, the immunoreactive cells being located in the outer half of each taste bud. Immunoreactivity in the olfactory mucosa was quite strong, but is confined to the supporting cells which are situated along the outer half of each lamella. A few immunoreactive supporting cells also were present between the receptors. In no case, however, were any receptors immunoreactive. Similarly, no neurons in the brain, including the olfactory bulbs and gustatory lobes, exhibited G-10 immunoreactivity. Some cells in the pituitary and pituitary stalk did exhibit immunoreactivity. These immunoreactive cells, however, are tentatively identified as supporting cells since their histologic characteristics are unlike those of the secretory cell populations. The pattern of G-10 immunoreactivity in the pituitary and olfactory mucosa suggests that the antibody may be crossreacting with a protein functionally unrelated to the alanine taste receptor, but which shares a common sequence. Further immunochemical studies will be required to resolve

29 Hodulcin Suppresses Behavioral and Receptor Responses to Sucrose in the Blowfly. D.E. KOLODNY and L.M. KENNEDY. Dept. of Biology, Clark Univ., Worcester MA 01610

Hodulcin (from <u>Hovenia dulcis</u> leaves) selectively suppresses sweetness perception in humans (Saul et al,1985). Receptor cell neurophysiological studies of hodulcin actions in comparison with the similar taste modifiers, gymnemic acids and ziziphins, could elucidate transduction mechanisms. We tested the effects of hodulcin on behavioral and neurophysiological responses of Phormia regina to determine the suitability of this fly for such studies.

A just-suprathreshold sucrose concentration (Kennedy et al,1975) for the behavioral proboscis extension response to tarsal stimulation was found for each of 28 flies (median 30 $(\pm 10$ SIQR) mM sucrose)(in 50mM NaCl). Using the same hodulcin preparation (aqueous extract, 1% w/v) and temporal parameters as in the previous human studies, we treated each fly for 2 min with hodulcin and then stimulated for 5 sec with just-suprathreshold sucrose at 20 sec post-hodulcin and at subsequent 1 min intervals until a positive response occurred. At 20 sec post-hodulcin, the response was inhibited in 75% of the flies (p=0.006, Sign Test). The median recovery was 2 (1-8 95% CI) min post-hodulcin.

In neurophysiological tests, single taste hairs in isolated proboscises were stimulated (5 sec) with 50 mM sucrose (in 50 mM NaCl) at 1 min intervals for 10 min after 2 min hodulcin or distilled water. At 1 min post-hodulcin, firing was significantly suppressed (median 8%, range 0-67%, of pre-treatment rates) and then increased over the test period (p<0.05). The median time to firing \geq pre-treatment rates was 4 (range 1-7) min post-hodulcin. In contrast, post-water firing did not vary (p>0.05)(Kramer ANOVAs).

The fly behavioral and receptor cell neurophysiological recovery times are comparable to the median 2.5 (range 1-4) min post-hodulcin recovery found in human perception (Saul et al,1985,unpubl.). Hodulcin selectivity will be tested using a NaCl stimulus in neurophysiology experiments as above. These data suggest that <u>P. regina</u> is an appropriate model for receptor cell studies with hodulcin.

NOTES

Abstract .

Withdrawn

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31 Ion Channels from Lobster Olfactory Receptor Neuron Somata. TIMOTHY S. MCCLINTOCK and BARRY W. ACHE (C.V. Whitney Lab, University of Florida).

Single channel activity was recorded from the somata of olfactory receptor neurons identified by morphological criteria. Sections of cuticle having aesthetasc sensilla with innervating dendrites and somata attached were isolated by dissection and successively treated with papain and trypsin to clean the membranes. Three types of steady-state channels with slope conductances of 320 pS, 100 pS, and 60 pS were observed in inside-out patches. The 320 pS channel is equally permeable to sodium and potassium and is blocked by 20 mM tetraethylammonium but not by cesium. This was the most common channel observed in inside-out patches, but was never observed in cell-attached patches. This channel showed no inactivation but some voltage dependence: the percent open time increased from 50% at -60 mV to 90% at 20 mV. The 60 pS channel is characterized by short open dwell times (< 20 ms) and infrequent openings. Its reversal potential suggests that it is permeable primarily to chloride. The 100 pS channel opens frequently in long, flickering bursts at negative holding potentials. were observed in approximately 25% of the inside-out patches and infrequently in cell-attached patches, as might be expected from the several gigaohm input resistance previously measured during whole-cell clamp of these somata.

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32 Effects of different ionic environments on the receptor potential of olfactory receptor cells in the spiny
lobster. I. SCHMIEDEL-JAKOB, P. A. V. ANDERSON, B. W. ACHE. (C. V. Whitney Laboratory, University of Florida).

Receptor potentials were recorded intracellularly from olfactory neurons of the lobster in situ using patch pipettes in the whole cell confingration. When stimulated chemically (extract of crab muscle or Tetramin) these cells produced prolonged, transient, dose-dependent depolarizations. The contributions of potassium, sodium and calcium ions to the receptor potential were tested by modifying the ionic composition of the saline superfusing the dendrites. The magnitude of the receptor potential evoked by a chemical stimulus was measured in normal and in ion depleted saline. Reducing K⁺ from 13.4 mM to 1.34 mM led to a reduction in the receptor potential or to its complete elimination. The effects were reversible; the receptor potential recovered within 3-6 min to its original value. External Na was reduced from 480 mM to 48 mM by substitution with TMA or choline. In approximately 50% of the cells tested there was no detectable effect while in the remaining cells the receptor potential was abolished or reduced in amplitude. Partial recovery occurred within 6 min. TTX applied in the superfusate had no effect on the receptor potential, while superimposed action potentials were blocked. The amplitude of the receptor potential was decreased or abolished in approximately 50% of the cells where the Catt concentration was reduced by one-half. The effect was long lasting and poorly reversible. From these findings it appears that the receptor potential is produced by the movement of several cations, including K⁺, Na⁺, and Ca⁺⁺. It must now be determined whether each of these ions acts directly as a current carrier or contributes to the ion- or voltage-dependency of other current carriers.

Supported by NSF Award BNS-85-11256 and a grant from the Whitehall Foundation.

Evaluation of Odor Plume Parameters For Positional Information Relative to the Source of the Plume. PAUL A. MODRE, and JELLE ATEMA (Boston University Marine Program)

Animals use odor plumes to locate food, mates, and other objects, but we know little of how they locate the source of odor plumes. In principle, the fluid dynamic processes that disperse odors are the same in air and water, but since size scales are smaller in water, the aquatic environment is easier to model. In addition acuatic plume tracers, such as salt or dye, are easter to measure. We used a salt water plume in a fresh water carrier flow. The plume was sampled at four sites down current: 12.5, 25, 34.5, and 50 cm, located in the plume center line. Odor intensity approached a stable (Gaussian) spatial distribution only after extensive time averaging, 0.1 Hz sampling frequency. Thus, slow-sampling chemoreceptors will detect an increasing concentration gradient when moving toward the odor source. At each site the patchy distribution of coor appears as a series of pulses varying in strength and duration. We analyzed five parameters that characterize odor pulses: height, length. maximum rising slope, and time between peaks (intermittency). We analyzed these at sampling frequencies of 40, 10, and 1 Hz. Probability distributions of these parameters showed that the distribution of relative pulse heights did not change with distance from the source. Similarly, pulse length, and intermittency did not provide positional information in the down current axis of this odor plume. Only maximum pulse slope provided reliable positional information. It is important to note that a gradient in the slopes of stimulus onset would be measured best by a series of receptor cells with different rates of adaptation and disadaptation. The glutamate receptor cells of lobster legs show this type of temporal filtering diversity (Voigt and Atema, ISOT IX, 1987).

Spatial-Temporal Filtering in Olfactory Chemoreceptor Cells. RAINER VOIGT and JELLE ATEMA (Boston University Marine Program, Marine Biological Laboratory, Woods Hole,

MA 02543).

The lateral antennular flagellum of the American lobster functions as an olfactory organ, and is of critical importance for orientation in odor plumes. Intermittent sampling by flicking the antennules and the patchy nature of odor plumes make it likely that the receptor cells of this organ detect pulsed stimuli in a chemically noisy background. A major cell population of these receptors is narrowly tuned to taurine. We have started to characterize the time courses of adaptation and disadaptation of receptor cells using a series of standard ls stimulus pulses with varying signal-to-noise ratios (SNR). The temporal stimulus pulse profile was determined by measuring the change of conductivity of flowing deionized water (20 ml/min) after injection of 100 ul of lM NaCl solution over 300 ms. The stimulus chamber allowed 5s interpulse intervals without interference from the previous stimulus. Single cells were identified electrophysiologically with $10^{-4}\mathrm{M}$ taurine. A series of five pulses was applied in 10s intervals for one of several concentrations $(10^{-3}-10^{-6}\mathrm{M})$ in different backgrounds $(10^{-4}-10^{-7}M)$. Combinations of stimulus and noise background concentrations provided similar SNR at different absolute stimulus concentrations and vice versa.

The results suggest that regardless of the background noise levels the same SNR resulted in similar responses, including similar cumulative adaptation. Greater SNR caused stronger responses and showed greater cumulative adaptation while smaller SNR caused weaker responses and less adaptation. Thus, SNR ratios and not absolute stimulus levels were predictive of the responses of receptor cells in different backgrounds. This feature predisposes the receptor cells to extract information on spatio-temporal fine structure of odor plumes and not on the time-averaged concentration gradients.

Supported by NSF grant BNS 8512585

35 Response Characteristics of Pheromone Receptor Neurons on Cabbage Looper Moth Antennae. R. W. MANKIN and M. S. MAYER (Insect Attractants, Behavior, and Basic Biology Research Laboratory, Agric. Res. Serv., U.S. Dept. Agric., Gainesville, FL) and A. J. GRANT (Worcester Foundation for Experimental Biology, Shrewsbury, MA).

A subset of olfactory receptor neurons on male cabbage looper moth antennae have high sensitivity and temporal fidelity of response to (2)-7-dodecen-1-ol acetate (27:12Ac), the major sex pheromone component of this insect. The stimulus-response relationship for the receptor neurons is described by a power function similar to the behavioral stimulus-response relationship obtained in flight-tunnel bioassays. Analysis of the neuronal response function yields an estimate of the electrophysiological threshold for Z7:12Ac detection and the threshold for discrimination of differences in stimulus intensity, ADI. As in many other sensory systems, the Weber fraction, ADI/I (the discrimination threshold divided by the stimulus intensity), decreases as the stimulus intensity increases.

Because these neurons are highly sensitive to 27:12Ac they appear to be well adapted for detecting pheromone plumes long distances downwind from calling female moths. Furthermore, because they respond quickly to small changes in stimulus intensity at levels expected in female gland emissions, the neurons appear also to be adapted for tracking the rapidly fluctuating concentrations that occur as the pheromone plume breaks up in the turbulent wind stream just downwind of the female.

NOTES

An Analysis of the Courtship Display Behavior in the Blue Crab (Callinectes sapidus). D.E. WOOD, CHARLES D. DERBY, DONALD H. EDWARDS (Department of Biology, Georgia State University, Atlanta, Georgia 30303), and RICHARD A. GLEESON (C.V. Whitney Laboratory, University of Florida, Rt. 1, Box 121, St. Augustine, Florida 32086).

Previous studies have described the courtship behaviors of male blue crabs elicited by the presence of a pheromone in the urine of pubertal females (Gleeson, 1980, Mar. Behav. Physiol., 7, 119-134). The features of this behavior include a lateral cheliped spread, standing high above substrate on walking legs, and the rhythmic waving of the swimming appendages. The striking feature of the courtship display which clearly differentiates this behavior is the rhythmic waving of the swimming appendages above the carapace. The focus of this examination is a description of this rhythmic waving using videotape analysis electromyograms. On the basis of simultaneous extracellular recording from several leg muscles and videotape analysis, the overall pattern of joint movement and muscular coordination underlying the behavior is fully described. These methods provided a route to investigation of the in vivo operation of the oscillators controlling motor output.

Neurophysiological Responses in Female Utetheisa ornatrix (Lepidoptera: Arctiidae) to a Male produced Pheromone. ALAN GRANT, ROBERT O'CONNELL (The Worcester Found. for Exp. Biology, Shrewsbury, MA 01545) and THOMAS EISNER (Sec Neurobiology and Behavior, Cornell University, Ithaca, NY 14853).

Many insects produce and release complex chemical signals that modulate various aspects of their reproductive behavior. Although little is known about the signals released by males, it is clear that, in a substantial number of moth species, males have specialized glands that produce chemical signals which are involved in intraspecific communication. In Utetheisa ornatrix, males possess eversible abdominal coremata, which release hydroxydanaidal (HD). HD is derived biochemically in adult males from the pyrrolizidine alkaloids (PAs) which are found in the larvas primary host plants, Crotalaria spp. The alkaloids impart to this aposematically-colored insect a "distastefulness" that serves to protect them from predation. During copulation the male donates a portion of his PA to the female along with the ejaculate. This PA along with some of the PA that she sequestered as a larva is subsequently transferred to the eggs. Therefore, the amount of HD released by a "displaying" male could provide the female with information about the male's competitiveness and his potential for assisting in the chemical protection of their offspring. We recorded responses from sensilla on the antenna of females to stimulation with graded doses of HD and related compounds. The array of sensory structures on the antenna include long trichoid and short basiconic sensilla. In the female, the receptor neurons specialized for the detection of HD are housed in short basiconic sensilla. The sensitivity and dynamic range of these receptor neurons in response to stimulation with graded doses of HD was similar in females raised on artificial, alkaloid-free diets and in those raised on diets with added Crotalaria. Receptor neurons in female trichoid sensilla, although insensitive to HD and extracts of male coremata, were responsive to some of the other biological extracts tested, suggesting a complex communication system.

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Further Isolation and Purification of the Garter Snake
Chemoattractant in Earthworm Wash. DALTON WANG, PING CHEN,
MIMI HALPERN (SUNY Health Science Center at Brooklyn)

Warm water washes of earthworms (Lumbricus terrestris) prepared according to established protocols (e.g. Halpern et al., Pharmacol. Biochem. Behav. 21:655, 1984) contain a potent chemoattractant for garter snakes (e.g. Burghardt Psychon. Sci. 4:37,1966). An improved behavioral assay and modified sample handling revealed chemoattractant activity in lower molecular weight fractions than previously described in publications from this laboratory. Earthworm wash separated on an AcA 44 column yielded two peaks as previously described. If polymerization was prevented by adding the reducing agent dithioerythritol (DTE), both peaks had chemoattractant activity. Fresh preparations of earthworm wash contained considerable amounts of sulfhydryl-containing compound(s) (4.86 X E⁻⁵ mmoles sulfhydryls/ml wash) as determined by titration at 324 nm with 4,4'-dithiodipyridine. These compounds were found to polymerize through oxidative formation of disulfides that could be prevented by addition of the reducing agent DTE. A relatively pure and highly active preparation of chemoattractant(s) was obtained by means of a combination of covalent (activated Affi-gel 401) and permeation (Bio-gel P-2) chromatographies. The resulting material showed a major protein band on SDS PAGE under reducing conditions with a MWr of approximately 20,000

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39 <u>Isolation and Purification of Earthworm Alarm Pheromone and Garter Snake Chemoattractant from Electric Shock - Induced Earthworm Secretions. XIAN-CHENG JIANG, DALTON WANG, MIMI HALPERN (SUNY Health Science Center at Brooklyn)</u>

Earthworms (Lumbricus terrestris), when shocked with electric current, secrete substances that conspecifics avoid on contact and act as chemoattractants to garter snakes (Thamnophis sirtalis) (Halpern, J. et al., Chem. Senses 11:607, 1986). The secretion, when chromatographed on an AcA-44 column, yielded three peaks. Peak 1 (P1) and peak 2 (P2) contained both chemoattractant for garter snakes and alarm pheromone for earthworms. Peak 3 (P3) contained only alarm pheromone. When material from P1 was rechromatographed on an AcA-34 column, it separated into four peaks (P1.1 to P1.4). Only P1.2 contained snake chemoattractant. When P3 was rechromatographed on a Bio-gel P-2 column, it separated into 4 peaks (P3.1 to P3.4). Only P3.4 contained alarm pheromone. The ratio of sugar to protein of P3.4 is greater than those of any other peak derived from P3. Using 4, 4'-dithiodipyridine we determined that there were thiol groupcontaining peptides in the earthworm secretion at a concentration of $^{\rm h}.2$ X 10^{-5} mmoles/mg protein. Such peptides might be responsible for the polymerization which we have observed. Polymerization could be prevented by using a reducing reagent (DTE). Affi-gel 401 (for thiol groupcontaining compounds) was used to isolate the peptides and the resulting product could be further resolved into 4 peaks with a Bio-gel P-2 column. Only the first peak contained snake chemoattractant. SDS PAGE showed a major 20K band in the first peak.

These findings suggest that there are thiol group- containing peptides (perhaps glycopeptides) which are responsible for the snake chemoattractant and polysaccharides or aminopolysaccharides which are responsible for the alarm properties of the irritation-induced earthworm secretions.

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Feeding Enhancement in the Herbivorous Fish Tilapia zillii by Amino Acids. MICHAEL A. ADAMS and PETER B. JOHNSEN (Monell Chemical Senses Center, University of Pennsylvania, 3500 Market Street, Philadelphia, PA 19104)

For most animal species, feeding behavior is controlled to a substantial degree by chemical signals received by the feeding animal. Food selection is mediated by two factors: chemical constituents of the food item and the animal's own chemosensitivity. Taste generally plays a key role in determining acceptance or rejection of a food item. Our previous work with the herbivorous fish Tilapia zillii (Comp. Biochem. Physiol., 83A: 109-112, 1986) showed that the amino acids glutamic acid, aspartic acid, serine, lysine, and alanine were powerful stimulators of feeding in this fish. This study examined the ability of these stimulatory substances to enhance the consumption of a model food item (10% extract of Romaine lettuce in a disk-shaped 2% agar matrix). A feeding enhancer is defined as a substance which, when added to a food item, will increase the consumption of the food above the amounts of the unenhanced food normally consumed, as measured in a two-choice behavioral feeding bioassay.

It was determined that only glutamic acid, aspartic acid, and lysine produced enhancement of feeding when added in a concentration of 0.01 M to test food items. Addition of glutamic acid caused a 4.5 times increase in consumption of the food disk over an unflavored agar reference. For aspartic acid the enhancement was four times the reference, and lysine induced a two-fold enhancement. In an experiment designed to delineate possible mechanisms of action, plain agar was prepared with acetate buffer at four different pH values: 3.4, 4.0, 5.0, and 6.0. When these samples were tested with reference to unflavored agar, T. zillii in all cases preferred the agar containing acidic acetate buffer.

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Chemical Communication in the Saddle Back Tamarin: Do
Non-Volatiles Play a Role? KAY L. GREENFIELD (Monell Chemical
Senses Center, Philadelphia); GISELA EPPLE (Monell Chemical
Senses Center; German Primate Center, Göttingen); ANNE M.
BELCHER (Monell Chemical Senses Center; German Primate
Center); IRMGARD KÜDERLING (German Primate Center); JOSEF
SCHNEIDER (German Primate Center); AMOS B. SMITH, III (Monell
Chemical Senses Center; Chemistry Department, University of
Pennsylvania)

Scent marking with specialized skin glands is a common behavior in the saddle back tamarin, Saguinus fuscicollis. The scent marks identify species, subspecies, gender, and individual, and they also contain information on the social position and hormonal condition of an animal. The marks are chemically complex, containing numerous compounds, including volatiles as well as components of higher molecular weight. Analysis by means of gas chromatography/mass spectrometry has identified 16 major volatile components (squalene and 15 esters of butyric acid). However, behavioral studies have shown that mixtures of butyrates and squalene alone do not encode information on subspecies and gender. Bioassays show that the attractiveness of organic solvent soluble preparations of scent marks can be increased if the non-soluble portion is added. This increase in attractiveness is apparent only when the animals are allowed direct access to the sample with their muzzles, suggesting that high molecular weight material may also be important to the chemical code. Recent electrophoretic studies document the presence of several proteins (major band ~ 66,000 daltons) in aqueous scent mark suspensions from both males and females. Behavioral studies show that these aqueous suspensions contain cues for gender discrimination. Tamarins presented with a choice between samples from males and from females preferentially investigate the material from males. This response is similar to that shown to natural scent marks. However, incubation of aqueous pools of marks with protease for 1 hr at room temperature appears to destroy gender cues. Test subjects did not discriminate protease treated aqueous suspensions of male scent marks from similarly treated material from females. The results of these electrophoretic and behavioral studies suggest that proteins may play a role in providing cues for gender identification in this species.

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42 Olfactory and gustatory responses of the channel catfish, Ictalurus punctatus, to nucleotides. W. MICHEL, J. J. ROBINSON II AND J. CAPRIO. (Department of Zoology and Physiology, Louisiana State Univ., Baton Rouge, LA. 70803).

Studies of chemoreception in the aquatic environment have focused primarily on amino acid stimuli; however, recent evidence from fish and crustacea indicate that nucleotides are also important chemosensory stimuli. The objective of this study was to determine the basic response characteristics of olfactory receptors and facial taste nerves in the channel catfish to nucleotides. The electroolfactogram (EOG) and integrated neural activity were simultaneously recorded from the olfactory epithelial surface, and integrated multiunit and single unit taste responses were recorded from mandibular and maxillary nerves. Both olfactory and gustatory thresholds to the more effective stimulatory compounds were estimated at 10 The relative stimulatory effectiveness (RSE) for olfactory receptors to nucleotides (mM) was most like that for P2 purinoreceptors, in that the RSE pattern was pyrophosphate (PPi) = triphosphate nucleotides > diphosphate nucleotides = monophosphate nucleotides - nucleosides > water control. ATP, ITP and PPi often caused synchronous oscillations ("peripheral waves"). These three compounds and ADP also consistently evoked positive EOGs irrespective of whether peripheral waves were generated or not. Two distinct RSE patterns of taste responses to nucleotides obtained from different facial nerve twigs were observed: (Pattern 1), nucleosides > monophosphate nucleotides > diphosphate nucleotides > triphosphate nucleotides, and (Pattern 2), $triphosphate > diphosphate \ge monophosphate \ge nucleosides.$ Patterns (1) and (3) are somewhat similar to those of P1 and P2 purinoreceptors, respectively. Further, nucleotide receptors appear relatively independent of those for amino acids, since responses to nucleotides were obtained during continuous presentation of L-alanine and L-arginine to the gustatory receptive fields.

Mackie and Adron, 1978. <u>Comp. Biochem. Physiol.</u> 60A:79-83 see Carr <u>et al.</u>, 1986. <u>J. Comp. Physiol.</u> 158:331-338.

Burnstock, 1980. <u>Prog. Biochem. Pharmocol.</u> 16:141-154.
(Supported by NIH grant NS14819)

Mixture Interaction Analysis: A Polynomial Response Summation Model Which Incorporates the Beidler Equation. PETER C. DANIEL and CHARLES D. DERBY (Department of Biology, Georgia State University, Atlanta, Georgia 30303)*

Response summation models have been used in the analysis of mixture interactions in systems where each component of a mixture adsorbs to a different receptor site type. In using response summation models, it is often assumed that the simple sum of responses to individual components can predict the response to a mixture of these components. Such an application of the response summation model can result in the value of the summed response exceeding that of the maximal response for the system being measured, be it an individual neuron or organism. This problem can be avoided by using a polynomial model:

$$1 - (1 - P_a) (1 - P_b) (1 - P_c) \cdot \cdot \cdot (1 - P_i) = P_{abcani}$$

where $P_a,\ P_b,\ P_c,\ P_i$ are responses to single components a, b, c, and i, each expressed as a proportion of the maximum possible response and P_{abc} , i is the predicted response to the mixture consisting of these components. The Beidler receptor kinetics equation has previously been used to construct a mixture model based on the presence of a single type of receptor. The Beidler equation can also be incorporated into the above polynomial equation through substitution of $K_x C_x/(1+K_x C_x)$ for P_x where K_x is the association constant and C_x is the concentration for each respective component (x). Consequently, it is possible to develop single receptor and multiple receptor predictions of responses to mixtures based on the same kinetic parameters determined from responses to single components.

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Visualization of a putative pheromone receptor-protein using a tritium-labeled photoaffinity analog. VOGT, R.G. (SUNY, Stony Brook); PRESTWICH, G.D. (SUNY, Stony Brook); RIDDIFORD, L.M., (U. of Washington).

Insect pheromone detection has long been noted for its remarkable sensitivity. However, the molecular mechanisms underlying this phenomenon have been somewhat elusive. We have previously described a pheromone binding protein and a pheromone degrading esterase from the antennal sensory hairs of the silk moth Antheraea polyphemus. We have proposed that these soluble and extracellular proteins control the movement of pheromone molecules within the sensory hair lumen (*).

We now report the first visualization of a putative membrane-bound pheromone receptor protein. Using a tritium-labeled photoaffinity pheromone analog with reported physiological activity, we have specifically labeled a protein which is associated uniquely with the dendritic membrane of the olfactory neurons. This 67,000 dalton protein satisfies criteria which allows us to suggest that it is responsible for binding pheromone at the membrane and subsequently initiating the primary transductory events associated with pheromone detection.

*Vogt & Riddiford (1981) Nature 293:161-163. Vogt et al. (1985) Proc. Natl. Acad. Sci. USA 82:8827-8831. Vogt (1987) in Pheromone Biochemistry (eds. Prestwich & Blomquist) (Academic Press, NY).

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45 Clonal Cell Strains from Rat Olfactory Epithelium and MonocTonal Antibodies that Help Characterize Them. HAYDEN G. COON, ROBERTO NISINI and FRANCESCO CURCIO (NIH, National Cancer Institute, Laboratory of Genetics, Bethesda, MD)

We have used an artificial basement membrane substrate and a complex medium (containing: 150µg/ml bovine hypothalamus and 15µg/ml pituitary extracts, 6% fetal calf serum, insulin and other hormones) to grow mass cultures and clonal strains from olfactory epithelium (OE) of newborn rats. Most cells in the clones are blastematic, multipolar cells that we take to be basal cells or derivatives of them. Other cells in the cultures are bipolar and may have an axon-like process >200 μ long at one pole and a more blunt structure with fine projections (either cilia or microvilli) at the other. Cell morphology appears to be a good marker, well correlated with staining by monoclonal antibodies (Mabs) specific for differentiated sensory cells as seen in fresh or fixed frozen sections (FFFS) of rat pup noses. Apart from morphology, reasons for tentatively concluding that the cultures contain at least partially differentiated sensory cells are: 1) the cell strains stain with anti-OMP (olfactory marker protein), kindly supplied by Dr. F. Margolis, 2) they stain with our own Mabs specific to sensory cells in FFFS, 3) a filamentous network is stained with polyclonal antiserum thought to be specific for neurofilaments, 4) staining is strongest in, and often localized to, one pole (golgi) of a bipolar cell.

Using standard methods we have immunized mice by repeated injections (IP) of strips of intact rat pup OE. The Mab

Using standard methods we have immunized mice by repeated injections (IP) of strips of intact rat pup OE. The Mab supernates were screened on FFFS followed by FITC-2nd antibody. Of 4000 Mabs tested we found 3 anti-OMP-like Mabs and about 6 anti-brush border (anti-sustentacular cell) activities, and many "anti-luminal" Mabs that might be anti-mucin. We have concentrated on a group of 8 Mabs that react exclusively with the distal-most surface of the sensory neurons (olfactory vesicle). In unfixed sections these Mabs stain sensory cilia too. Serial FFFS reveal at least 3 patterns of staining within the sensory epithelium. None of these cross-reacts with hamster FFFS. At least one precipitates a strong 97kD band from OE incubated with 35S-met. We shall try to characterize these cells further with electrophysiology and biochemical assays for receptor function and more Mabs.

46 Cloning and Sequence Analysis of Two cDNAs Encoding Rat Odorant-Binding Protein. JONATHAN PEVSNER (Dept. of Pharmacology, Johns Hopkins Sch. Med., Baltimore MD 21205), RANDALL R. REED (Dept. Mol. Biol. & Genetics, Howard Hughes Med. Inst., Johns Hopkins Sch. Med.) and SOLOMON H. SNYDER (Dept. Neuroscience, Johns Hopkins Sch. Med.)

Odorant-binding protein (OBP) is a soluble 20 kDa protein that binds several structurally unrelated odorants and is secreted from glands in the nasal mucosa (Pevsner et al., P.N.A.S. 83:4942, 1986). We obtained the N-terminal sequence of 14 amino acids from rat OBP and synthesized a pool of 21-base oligonucleotide probes corresponding to the first seven amino acids. We used the probes to screen a lambda gt10 complementary DNA (cDNA) library derived from rat olfactory epithelium (provided by Ernest Barbosa, Johns Hopkins U.). Two clones (800 and 850 base pairs) which were recognized by the oligonucleotide probes were isolated and partially sequenced. Initial results suggest that the connactor encode OBP because amino acid sequences of the protein adjacent to the region used to generate the oligonucleotide probes are identical to those determined from DNA sequencing of that region. Minor sequence differences among the OBP clones and the protein suggest that multiple forms of OBP may exist. The predicted partial amino acid sequence indicates that OBP is homologous to \propto -2-microglobulin, an 18 kDa serum protein homologous to retinol-binding protein, B-lactoglobulin and protein HC. Recently, the gene for a secreted 20 kDa protein has been isolated from frog olfactory epithelium (Lee et al., Science, in press). This protein could represent frog OBP or, like OBP, a member of the larger binding protein family.

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Immunohistochemical Localization of GTP-binding Protein in Rat Olfactory Epithelium during Prenatal Development.

BARBARA MANIA-FARNELL, ALBERT I. FARBMAN (Dept. of Neurobiology, Northwestern Univ., Evanston, IL 60201)

It has recently been proposed that guanine nucleotide (GTP) binding regulatory proteins (G-proteins) are involved in the transduction of stimuli in olfactory receptor neurons (Pace et al., Nature 316:255, 1985; Sklar et al., J. Biol. Chem., in press). If this is the case, G-protein should be expressed at the time when receptor cells become functionally selective for odorants, i.e., in the rat fetus at about E17 (Gesteland et al., Neuroscience, 7:3127, 1982). In this study, we have examined the developing rat olfactory epithelium to determine when in ontogeny the ß subunit of Gprotein is first expressed. Rat fetuses, from E15-E22 were used in this study. Fetal heads were fixed in 4% paraformaldehyde in phosphate buffer and frozen sections prepared for indirect immunofluorescent staining. primary antiserum was a monospecific polyclonal generated in rabbits by immunization with a synthetic peptide, the amino acid sequence of which matches a portion of the ß subunit of G-protein (Mumby et al., PNAS, 83:265, 1986). The results showed that this subunit is first demonstrable at the ciliary surface of fetal rat olfactory epithelium at E20 and in axon bundles several days earlier. Localization of Gprotein in rat embryos is identical to that seen in adult frogs (Anholt et al., Biochemistry, in press). However, it is not demonstrable by immunofluorescence until 3 days after cells have become functionally selective for odorants. The reasons for this are not clear. One possibility is that the antigenicity of G-protein is attenuated by fixation.

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48 <u>Isolation and Characterization of Five G-Protein Encoding cDNA Clones from Rat Olfactory Epithelium</u>. DAVID T. JONES* and RANDALL R. REED (Howard Hughes Medical Institute, Johns Hopkins University).

In an effort to understand olfaction at the molecular level, we are identifying the components that may mediate odorant recognition and signal transduction. Recently, Pace et al. have shown that a crude preparation of the olfactory cilia (extensions of the dendritic bulb of the primary olfactory neurons) possess considerable odorant-. stimulatable, GTP-dependent, adenylate cyclase activity. This finding is reminiscent of the visual system where, in the rod outer segment, photoactivated rhodopsin activates a retinal specific GTP-binding protein (G-protein), transducin, which in turn activates cGMP phosphodiesterase. We hypothesized that, in analogy to the visual system, the olfactory system may have evolved its own distinct CDNA library was screened with a mixed oligonuclectide probe generated from a highly conserved region of known G-protein sequences and five independent G-protein encoding clones were isolated. Analysis of their primary structures identified them as $G_{\rm S}$, $G_{\rm O}$, and three types of $G_{\rm I}$. The predicted amino acid sequences show that all five G-proteins are highly homologous to each other and to the two forms of transducin. Transducin (I & II), G_0 , and three G_1 proteins all bear greater than 58% identity. G_5 , the most divergent, bears less than 45% identity with any of the other groups. Northern analysis demonstrated that G_5 and one form of G_1 (G_{118}) are the most abundantly expressed and that the remaining three G-clone messages (G_0 , G_{116} , and G_{127}) are expressed at relatively low levels in olfactory tissue. Western analysis (personal communication) indicated that the G_1 species are evenly distributed throughout the tissue, whereas, G_S , like adenylate cyclase, is highly enriched in the cilia. We suggest, therefore, that the initial events of olfactory signal transduction, for at least some odorants, are mediated by odorant receptors coupled to adenylate cyclase via the stimulatory \mathbf{G} -protein, $\mathbf{G}_{\mathbf{S}}$. *National Science Foundation Fellow

Phospholipid Turnover in Catfish Barbel (Taste) Epithelium with Special Reference to Phosphatidylinositol-4,5-bisphosphate.

T. HUQUE*, J.G. BRAND*,+, J.L. RABINOWITZ+ and D.L. BAYLEY*. (*Monell Chemical Senses Center, 3500 Market Street, Philadelphia, PA 19104; and +Veterans Administration Medical Center and University of Pennsylvania, Philadelphia, PA 19104).

The amount and type of lipid may affect the function and stability of taste receptors. In addition, the metabolism of certain phospholipids, in particular the polyphosphoinositides, may be involved in signal transduction in taste, as they are in olfaction (Huque and Bruch, Biochem. Biophys. Res. Commun. 137, 36-42, 1986). A total lipid analysis of catfish barbel epithelium revealed that approximately 40% of the total lipid was recovered in the neutral fraction, while 60% was recovered in the polar fraction. neutral fraction, while 60% was recovered in the polar fraction. The phosphatidylcholines (PC), -ethanolamines and -serines accounted for the majority of the phospholipids and were all present to nearly the same extent (10-15%). The phosphatidylinositols (PI) accounted for approximately 5% of the total lipid. Using ³²PO₄, phospholipid turnover studies revealed that PC and lyso-PC turned over to the greatest extent and reached a peak of ³²P incorporation after 10-15 mins of incubation. PI turnover was next active. Persentation of phosphotically incubated 5-biss. next most active. Degradation of phosphatidylinositol-4,5-bis-phosphate (PIP₂) to the putative second messengers inositol triphosphate (IP₃) and diacylglycerol (DAG) was studied with radio-labeled PIP₂ using the procedure of Downes and Michell (Biochem. J. 198, 133-140, 1981). Anion-exchange chromatography separated the various inositol phosphates. The barbel epithelium exhibited PIP₂-phosphodiesterase (PIP₂-PDE, E.C. 3.1.4.11) activity. The primary labeled product formed was IP3. Enzyme activity was calcium-dependent but not calcium-regulated. When the medium contained 2 mM EGTA the enzyme was rendered inactive, but at free Ca $^{2+}$ concentrations spanning the range 0.1-12 μ M, enzyme activity was relatively constant. Degradation of PIP, was pHdependent, showing maximal activity at pH 7.1. The activity of the enzyme was enhanced by alanine, a known taste stimulus for the catfish. When ventral skin of the catfish was used as a control tissue (since it has few taste buds), PIP2-PDE activity was observed, but with no sensitivity to alanine.

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Olfactory L-Amino Acid Receptor Specificity and Stimulation of Potential Second Messengers. RICHARD C. BRUCH (Monell Chemical Senses Center), R. DINO RULLI (Monell Chemical Senses Center) and ARDITHANNE G. BOYLE (Monell Chemical Senses Center; Veterans Administration Medical Center)

Electrophysiological assays have shown that L-alanine and L-arginine are effective olfactory stimuli for the channel catfish, Ictalurus punctatus (Caprio and Byrd (1984), J. Gen. Physiol). Since these amino acids do not cross-adapt significantly, it was proposed that these odorants interacted with separate receptor sites and/or transduction pathways. The specificity of the binding of these odorants to receptor sites was therefore evaluated in isolated cilia preparations from the olfactory epithelium. Since these receptors interact with signal-transducing GTP-binding proteins (Bruch and Kalinoski (1987), J. Biol. Chem.), the ability of these odorants to stimulate the formation of the potential second messengers inositol triphosphate (IP₃) and cAMP was also investigated. Although L-alanine and L-arginine exhibited similar binding affinities, they did not compete effectively for common binding sites, suggesting that these stimuli bound to separate receptors. In addition, the interactions of these ligands with receptors were selectively and differentially inhibited by lectins (Kalinoski et al. (1987), Brain Res.). Both odorants rapidly (within 15 sec) stimulated IP3 formation by 2-3-fold over basal levels. Similar degress of adenylate cyclase stimulation were also observed in response to both stimuli after 10 min. Both odorant-stimulated IP₃ and cAMP formation were GTP-dependent, implicating GTP-binding protein involvement in mediating activation of these second messenger pathways. In combination, these results suggest a role for both phosphoinositidederived and cyclic nucleotide second messengers in olfactory transduction. These results also support the hypothesis that odorantactivation of some olfactory receptors may stimulate both phosphoinositide turnover and adenylate cyclase by mechanisms mediated by G-proteins. These results further suggest that receptor specificity, rather than alternative transduction pathways, may account for the previous electrophysiological cross-adaptation data.

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Biochemistry and Physiology of Purinergic and
Taurinergic Chemosensory Systems. HENRY G. TRAPIDOROSENTHAL, RICHARD A. GLEESON, WILLIAM E. S. CARR (C. V. Whitney Laboratory and Dept. of Zoology, University of Florida, St. Augustine, FL 32086)*

The antennule of the spiny lobster, <u>Panulirus argus</u>, is an olfactory organ that has chemosensory sensilla containing the dendrites of large numbers of primary sensory neurons. Electrophysiological studies have revealed that populations of these neurons are differentially activated when either purine nucleotides (AMP, ADP, or ATP) or taurine (Tau) are present in sea water bathing the sensilla. At least three populations of purinergic receptors have been identified that are strongly activated by nucleotides but not by the nucleoside adenosine (Ado). Two populations of taurinergic receptors are also known.

Biochemical studies show that the chemosensory sensilla contain ectonucleotidases that dephosphorylate, and thereby inactivate, the excitatory nucleotides, AMP, ADP and ATP, to yield the far less active nucleoside Ado. Ado is then internalized by a specific uptake system. Both the uptake system and the ectonucleotidases have marked similarities to analogous systems found in the internal tissues of vertebrates. The rate of dephosphorylation of AMP by the sensilla is very rapid. Kinetic measurements are being made to evaluate the correlation between the inactivation rate of AMP and the antennular flicking mechanism that is used by the lobster to obtain intermittent samples of the external chemical environment.

Whereas stimulatory nucleotides are inactivated in sensilla by dephosphorylation, stimulatory Tau molecules are cleared from the receptor environment by a specific uptake system similar to that found in internal tissues.

Collectively, our results show that certain of the receptors and the inactivation systems present in olfactory sensilla, have marked similarities to analogous systems in internal tissues where some of the same substances have neuroactive effects.

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 $52 \underbrace{\begin{array}{c} \text{Intracellular Recordings from Ganglion Cells} \\ \text{of the Elasmobranch Nervus Terminalis.} \end{array}}_{\text{And MICHAEL MEREDITH (Dept. of Biological Science,}}$ Florida State University, Tallahassee, FL 32306).

Data from in vivo electrophysiological experiments in the bonnethead shark (Sphyrna tiburo) suggest that action potentials from the brain suppress nervus terminalis (NT) ganglion cell activity, probably via inhibitory synapses (White and Meredith, ISOT/AChems, 1986). To further investigate this inhibition, as well as other aspects of NT physiology, intracellular recordings were made in vitro from cells in the NT ganglion of the (Dasyatis sabina). After removal from the animal and brief treatment with collagenase, the NT ganglion with short lengths (3 - 5 mm) of peripheral and central trunks attached was pinned out in a chamber filled with Ringer's solution and the nerve ends drawn into suction electrodes for electrical stimulation. Using glass microelectrodes, cells were found in NT ganglia of both species which responded to electrical stimulation of the central and/or peripheral nerve trunk with long hyperpolarizing potentials, supporting the in vivo extracellular electrophysiology. In some of these cells, central trunk stimulation elicited an action potential which preceded the hyperpolarizing potential. Cells were also found which exhibited action potentials without hyperpolarizations when the central trunk was stimulated and tions when the central trunk was stimulated, and others which responded to peripheral trunk stimulation with simple or complex depolarizing potentials. The variety and complexity of membrane potential changes observed following nerve trunk stimulation suggests that the NT ganglion serves a function more complex than that of a simple group of sensory cell bodies or a simple efferent relay.

Supported by NSF Grant BNS 8412141.

Effects of Vomeronasal and Main Olfactory Lesions on Communicative Behaviors and Hormone Responses in Hamsters. Robert E. Johnston, Cheryl Pfeiffer (Cornell University)

In a continuing effort to characterize the functions of the main and accessory olfactory systems we investigated the effects of peripheral lesions of these systems on hormonal responses and on odor-facilitated communicative behaviors in male and female golden hamsters (Mesocricetus auratus). To eliminate sensory function vomeronasal organs were surgically removed while the olfactory mucosa was treated with ZnSO4. The behaviors studied were ultrasonic calling and two scent marking behaviors, flank marking and vaginal marking. In male hamsters circulating levels of testosterone were measured following exposure to females or their odors.

Vomeronasal organ removal had no effect on odor-elicited scent marking behaviors by either males or females, but it did significantly reduce ultrasonic calling by females. Zinc sulfate treatment of the olfactory mucosa reduced flank marking frequency by both males and females and also decreased vaginal marking frequency. In addition such treatment reduced ultrasonic calling by females. Thus scent marking behaviors are mediated primarily by main olfactory system input whereas ultrasonic calling is influenced by both olfactory and vomernasal systems.

Lesions of the vomeronasal organ in both sexually experienced and sexually naive males eliminated increases in testosterone levels caused by exposure to vaginal secretions; lesions of the main olfactory system had no effect. Lesions of either system alone did not influence increases in testosterone caused by contact with a female, but combined lesions of both systems in sexually naive males did eliminate testosterone responses. Thus the vomeronasal organ is necessary for testosterone responses to vaginal secretions whereas the main olfactory system has little influence. Sexually inexpereinced males require some olfactory/vomeronasal input to show testosterone responses to females themselves.

These results are consistent with the previously proposed hypothesis that the main olfactory system is important for making discriminations (species recognition in the case of flank and vaginal marking, male vs female in the case of vaginal marking and ultrasonic calling) whereas the vomeronasal system is concerned with more basic biological processes (testosterone responses and, in the case of ultrasonic calling, mating).

This research was supported by NSF grant BNS 84-10040.

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The Origin Of Luteinizing Hormone-Releasing Hormone (LHRH) In The Vomeronasal System Of The Hamster. M.N. LEHMAN (Univ. Cincinnati Coll. Med.), S.W. NEWMAN (Univ. Michigan) and A.J. SILVERMAN (Columbia Univ. P.& S.)

Immunoreactive LHRH fibers in the hamster are found in the central targets of the vomeronasal sensory system, specifically in the accessory olfactory bulb (AOB), and the medial nucleus of the amygdala (M). It is not known whether these fibers arise from LHRH neurons located along the course of the terminal nerve or within its ganglion, or from LHRH cells located more caudally in the diagonal band, medial septum, and preoptic area. We have used a combination of retrograde tract tracing and immunocytochemistry to determine which LHRH cells project to AOB and M, and compared their distribution with that of LHRH cells which project to the median eminence (ME) Stereotaxic microinjections of fluorescent tracers (100-200 nl of either 4% Fluoro-Gold in 0.9% saline or 10% rhodamine-coated microspheres in the same vehicle) were made into either AOB, M, or ME of pentobarbital anesthetized male hamsters. Following survival periods of 3-7 days, hamsters were perfused with Zamboni's fixative and their brains processed for LHRH immunocytochemistry using an avidin-fluorescein procedure (Vectastain). Sections were examined with fluorescence microscopy to determine which LHRH cells were retrogradely labelled. Analysis revealed that (1) LHRH input to the AOB arises in part from a very few cells in the medial portion of the anterior olfactory nucleus and in the anterior hippocampal rudiment; (2) LHRH fibers in M originate from a small number of scattered cells in the preoptic area beneath the anterior commissure; and (3) LHRH input to the ME arises from a larger number of cells (approximately 50% of all LHRH neurons) distributed throughout the diagonal band, medial septum, and preoptic area. LHRH cells in the intracerebral portion of the terminal nerve do not appear to project to either the AOB, M, or ME. Results suggest heterogeneity in the projections of individual LHRH cells.

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Vomeronasal Chemoreception, Sexual Experience and Reproductive Behavior of Male House Mice and Prairie Voles.

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The concept that adult male mammals remain in a static condition of reproductive competency has been challenged by recent evidence that interactions with females result in gonadotropin and androgen surges. These endocrine responses to females are proposed to augment the potency of the male to subsequently stimulate reproduction in females, and in some situations can be elicited by chemosignals associated with females. We sought to determine whether social/sexual experience and vomeronasal chemoreception are important determinants of behavioral and endocrine responses of male house mice and prairie voles to certain types of stimuli.

Males were subjected to surgical removal of vomeronasal organs (VNX) or sham surgery (SHAM). SHAM and VNX male house mice were given social experience with females that were anointed with perfume or ethanol, and then tested for behavioral (ultrasonic vocalizations, USV) and androgen responses to male or female urine or perfume. SHAM males had higher androgen levels only in reponse to female urine, in spite of their social experience. However, USV responses to female urine persisted in some males despite VNX, in contrast to previous reports.

Sexually experienced and inexperienced male prairie voles were subjected to VNX or SHAM surgery. We then recorded USV and androgen responses to anesthetized female conspecifics. Preliminary results corroborate those for mice: androgen responsiveness, but not always behavioral responsiveness, depend on an intact vomeronasal system.

Histological verifications are incomplete, but these results suggest that chemosensory-induced endocrine responses may be hard-wired in their dependence on vomeronasal chemoreception. In contrast, behavioral responses to chemical cues may show considerable plasticity.

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Inhibition of Chorda Tympani Taste Responses in a Model System. B. OAKLEY, D. R. RIDDLE, C. R. BELCYZINSKI, C. L. DESIBOUR, AND S. E. HUGHES (University of Michigan, Ann Arbor, MI 48109 and Washington University, St. Louis, MO 63119).

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In the gerbil, Meriones unguiculatus, we crossed the right chorda-lingual nerve to the left lingual nerve. Both the left (native) and right (foreign) chorda tympani (CT) nerves responded vigorously to taste stimulation of the left side of the tongue where the foreign CT innervated many of the fungiform taste buds, as subsequently confirmed by anatomical analysis. We frequently observed putative inhibitory interactions while simultaneously recording from both CT nerves during gustatory stimulation. We confirmed the existence of potent inhibition by using electrical stimulation (e.g., 1 msec, 4V pulses for 2 min at 50 Hz) of the native CT to inhibit taste responses of the foreign CT in 11 of 16 animals. Specifically, a significant reduction occurred in both the phasic and tonic responses to 0.5 M sucrose (41% and 32% inhibition, respectively) and in the phasic and tonic responses to 0.3 M NaCl (52% and 23% inhibition, respectively). Inhibition of the taste responses was long-lasting: 50% recovery required 11.5±2.0 minutes. The inhibition must have occurred at the level of the taste buds as connections with the brain were not necessary for the inhibitory effects. The short latency of inhibition ruled out ischemia as an explanation for reduced taste responses. Spontaneous activity in the foreign CT was normal during and after electrical stimulation of the native CT. No significant changes occurred in the taste responses of the electrically stimulated nerve itself, or in the responses of the native nerve when the foreign CT was stimulated. Nor were changes observed following electrical stimulation of the CT in normal or control animals. Inhibitory interactions, as revealed in this model system by the dominance of the native CT, may act to sharpen beststimulus profiles.

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57 Quality Coding in Olfaction by Spiny Lobsters:
Behavioral Discrimination. JACQUELINE B. FINE-LEVY and
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University, Atlanta, Georgia 30303)*

We are using the Florida spiny lobster, $\underline{\text{Panulirus}}$ $\underline{\text{argus}}$, as a behavioral model for the study of quality coding in olfaction. It has been demonstrated, using a discriminative conditioning paradigm, that spiny lobsters are able to behaviorally differentiate between members of a set of chemical mixtures, and between members of a set of single compounds; all chemicals appearing as components of the animals' natural food (Fine et al, 1986, Chem. Senses 11,597). Two sets of experiments were conducted, one using chemical mixtures and the other using single chemicals. In each set of experiments, lobsters were distributed into groups, each group being trained to a different member of the set. Comparisons of changes in responses following conditioning indicate that crab mixture and shrimp mixture are perceived as being more similar to each other than to mullet mixture and oyster mixture, with oyster mixture being the most dissimilar from crab mixture and shrimp mixture. Within the single chemical set, AMP and glycine appear to be perceived as being more similar to taurine than are betaine and glutamate. Comparisons between these behavioral data and neurophysiological data (Girardot and Derby, abstract at this meeting) provide an indication of the type of chemical coding which allows the animals to behaviorally discriminate between these chemicals.

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Neural Discrimination of Odorant Quality in the Spiny
Lobster: Multivariate Analysis. NADIA GIRARDOT and CHARLES
D. DERBY (Department of Biology, Georgia State University.

Atlanta, GA 30303)*

Multivariate analyses (cluster and factor analyses, and multidimensional scaling) were used to test the hypothesis of a neural population code for quality discrimination among chemicals presented to the antennule of the spiny lobster,
Panulirus argus. The data resulted from extracellular
recordings in response to chemical stimulation of spike frequency in response to chemical stimulation of either receptor cells or output neurons projecting from the main olfactory center of the Brain. The chemical stimuli used were a set of 8 compounds normally found in the lobster's natural food sources (taurine=tau, glycine=gly, betaine=bet, L-glutamate=glu, L-proline=pro, L-alanine=ala, adenosine 5' monophosphate=amp, and hydroxy-l-proline=hyp. In one experiment, the stimuli were extracts of natural food (crab, mullet, oyster, shrimp), and the synthetic mixtures of these extracts based on their chemical composition. The analysis provided evidence that evaluation of relative similarities of across-neuron patterns generated by olfactory stimuli results in qualitative discrimination among them. For instance, the 8 single components may be clustered into four group 3=tau; group 4=gly). This discrimination is established at the receptor level and in general persists up to the output neuron level. For the complex odorants (extracts and mixtures) the patterns generated by crab and shrimp are more similar to each other than to those produced by oyster and mullet. Increasing the concentration by a 10 or 100-fold produced a proportional shift in the amplitude of the across-neuron pattern without altering it, suggesting that quantity of olfactory stimulus is coded by comparing shifts in amplitudes of the neural population responses, while quality is coded by comparing their patterns.

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59 Real-time video recording of olfactory bulb evoked potentials in the salamander using a voltage-sensitive dye.

J.S. KAUER. (Tufts-New England Medical Center, Boston. MA)

A prevalent hypothesis about how quality and concentration of single odor stimuli are represented, suggests that the encoding of molecular information at each relay in the olfactory pathway takes place within many simultaneously activated neural elements. There is good evidence that such 'parallel' representation of an odor occurs at receptor, bulbar and prepyriform levels. To examine such distributed activity in the olfactory bulb simultaneous multiple-site electrical recording, and 2-deoxyglucose (2-DG) and cytochrome oxidase metabolic

mapping methods have been used.

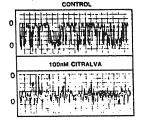
In the present study, series of 16 sequential real-time video images (128 x 128 pixels) of changes in dye fluorescence from the olfactory bulb have been generated after electrical stimulation of the olfactory nerve (ON). Using voltage sensitive dye RH414 (obtained from L.B. Cohen, Molecular Probes Inc., and A. Grinwald) images acquired via a Dage TV instrumentation camera (with a Newvicon tube), were enhanced using computerized processing. The time course of the fluorescence changes correlated well with the time course of simultaneously recorded field potentials. Signals were not seen in unstained tissue or in the absence of electrical stimulation. The signals were abolished after treatment of the bulb with 10 um. tetrodotoxin. Stimulation of the whole ON elicited widely distributed fluorescence changes, first throughout the glomerular layer, and then throughout the deeper bulbar layers. Stimulation of two widely separated ON fascicles (medial and lateral) also elicited widespread fluorescence changes in each of the other layers. This method allows the observation of changes in neuronal activity with higher spatial resolution than simultaneous electrode recordings or dye recordings using diode detectors and with higher temporal resolution than 2-DG mapping.

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60 <u>Ion Channels of Olfactory Cilia</u>. PEDRO LABARCA, SIDNEY A. SIMON and ROBERT R. H. ANHOLT (Department of Physiology, Box 3709, Duke University Medical Center, Durham, NC 27710)

We have detached chemosensory cilia from the olfactory epithelium of the bullfrog, Rana catesbeiana, and fused the isolated ciliary membranes with preformed planar lipid bilayers. Single channel records revealed spontaneous openings of at least three different cation selective channels with conductances of 40 pS, 100 pS and 230 pS, respectively, in 5 mM HEPES, 200 mM KCl, pH 7.0. Only the 100 pS channel was activated (opened) by odorants. Activation was observed at nanomolar concentrations of the bellpepper odorant, 3-isobutyl,2-methoxypyrazine, and the citrus odorant, citralva (3,7-dimethyl-2,6-octadiene-nitrile). The lifetime of the channel in the presence of odorants was in the millisecond range. Channel activation was reversible upon removal of the odorant from the bilayer chamber via perfusion. The odorant channel exhibited a linear current-voltage relationship between -70 mV and +70 mV. The probability of the channel being in the closed state was greater at negative applied voltages. The channel was cation-selective with similar permeability to sodium and potassium ions. These observations suggest that activation by odorants of cation channels at the chemosensory membrane may play an important role in olfactory reception.



Records of the odorant sensitive channel before (upper trace) and approximately 1 min after (lower trace) the addition of 100 nM citralva at a holding potential of -40 mV. o, open state; c, closed state.

A cyclic nucleotide-gated conductance in olfactory receptor cilia. TADASHI NAKAMURA AND GEOFFREY H. GOLD (Dept. of Physiology, Yale U. Sch. of Med., New Haven, CT 06510)

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An odorant-sensitive adenylate cyclase has recently been reported in olfactory receptor cilia (Pace et al., Nature 316, 255). If cAMP is an intracellular messenger for olfactory transduction, there must be a conductance in the olfactory transduction, there must be a conductance in the ciliary plasma membrane that is regulated by cAMP. We studied the conductive properties of olfactory receptor cilia by use of the patch clamp technique. With patch pipettes that were fire-polished to resistances of 30-50 Mohm, we obtained gigaohm seals on cilia of enzymatically dissociated receptor cells from the olfactory mucosa of the road. Bufo Marinus In excised, inside-out patches, we toad, <u>Bufo Marinus</u>. In excised, inside-out patches, we observed a conductance which was reversibly induced by the bath application of cAMP. This conductance was observed in the absence of added NTP's, indicating that it is gated directly by cAMP, rather than resulting from protein phosphorylation. The timecourse of the ciliary conductance changes followed step changes in bath cAMP concentration with a lag of <-200 msec. With Ringer's solution in the patch pipette and a pseudo-intracellular solution in the bath, the reversal potential for this conductance was -5+3 mV. The value of the reversal potential is similar to reported values for the reversal potential of the odorant response, suggesting that this conductance mediates olfactory transduction in vivo. The steady-state concentration dependence of the ciliary conductance was measured for cAMP, cGMP and cCMP; the K1/2 values were 3.4±1.0, 1.5±0.6, and 57±10 uM, respectively. The slightly higher affinity for cGMP than for cAMP raises the possibility that the intracellular messenger may be cGMP, or a mixture of cyclic nucleotides, rather than cAMP alone. The ciliary conductance exhibits voltage-dependent block by divalent cations from both sides of the membrane. In the absence of divalent cations, single channel events of ~50 pS are observed. The similarities between the ciliary conductance and the cGMP-gated conductance that mediates visual transduction in rod and cone photoreceptors, suggest considerable similarity between the mechanisms of olfactory and visual transduction.

Electrical Properties of Olfactory Receptor Cells Isolated from Coho Salmon (<u>Oncorhynchus kisutch</u>). <u>Gabrielle Nevitt</u>, Department of Zoology, University of Washington, Seattle, Washington, 98195

Salmon offer a unique opportunity to study the biophysical basis of olfaction as it relates to behavior. Their keen ability to discriminate between specific chemicals by smell has been shown to play a key role in homing as well as predator avoidance. Here, I present a characterization of ionic currents present in olfactory receptor cells of Coho salmon using the whole-cell recording technique of Hamill, et al (1981).

Receptor cells were isolated from 18 month to 2 year old fish by gently agitating rosette tissue in 0 Ca / 0 Mg ringer, followed by an incubation in papain (.5 mg/ml / 30 mtn). Elongate, ciliated receptor cells retained their morphology and were easily recognizable. 10-20 G ohm seals were routinely obtained, and in cell-attached configuration, spontaneous action potentials were frequently observed.

Under voltage clamp recording conditions, both inward and outward voltage-dependent currents were seen. Transient inward currents occurred in response to depolarizations from holding potentials of -60 to -110 mV. Amplitudes ranged from 200 to 500 pA and peak currents occurred at voltages of -10 to -40 mV. Peak currents activated within 1 ms and 1/4 inactivation times were of the order of 3 ms. I assume this current to be carried by Na ions because it was blocked by TTX, and absent when choline replaced Na in the external ringer. The inward Na current was followed by a delayed outward current which was blocked by the replacement of internal K with Cs in Na-free ringer. With identical internal solution and 10 mM Sr substituting for 3 mM Ca externally, a second inward current was observed during depolarizations from a holding potential of -40. Peak current occurred at steps to a -10 to +10 mV range. This current did not inactivate during a 100 ms pulse, and completely disappeared within 3-6 min following the establishment of whole-cell configuration. I believe this current to be Sr ions passing through a Ca channel. Further investigation will focus on elucidating the role of these currents in olfactory cell response to odorants. (Supported by NIH grant HD17486 and a Graduate Research Award from the University of Washington)

63 Effects of Injection of cyclic-GMP into the Mouse Taste Cells. Keiichi Tonosaki and Masaya Funakoshi (Dept. of Oral Physiology, Sch. of Dent., Asahi Univ., Hozumi, Hozumi, Motosu, Gifu 501-02, Japan)

The mouse taste cells were morphologically and electrophysiologically classified into two types, H- and D-type, according to their sucrose responses. From the results indicated the possibility that the H-type response is due to a decrease of potassium and/or chloride conductance and D-type response is due to an increase of sodium conductance. Still the taste transduction mechanism is unclear. It is natural that there are several complex intracellular taste transduction mechanisms in the taste cell. Since cyclic-GMP and cyclic-AMP might be postulated as a candidate for an intracellular transmitter in a variety of cells.

In the present experiments, in order to examine the possibility that cyclic-GMP is an internal transmitter in the process of taste transduction of H-type taste cell (The results of D-type are discussed elsewhere.). Cyclic-GMP was iontophoretically injected into a mouse taste cell from one barrell of a double or tri-barrelled microelectrode with the other barrell monitoring membrane potential and resistance changes. Cyclic-GMP induced a depolarization membrane potential accompanied by an increase in membrane resistance. The depolarization potential change was also caused by cyclic-AMP injection, though it was always smaller than that by cyclic-GMP. The results suggest that cyclic-GMP is involved in the taste transduction process in the mouse taste cell. However, uncertainty still remained as to whether the cyclic-GMP directly correlates with the taste cell membrane potential.

Transmucosal Inward Currents Coincide With Neural Excitation and Adaptation to NaCl in the Rat. John A. DeSimone, Gerard L. Heck, and Krishna Persaud (Dept. of Physiology, Virginia Commonwealth University, Richmond, VA 23298)

Electrophysiological studies on the in vitro lingual epithelium as well as the effect of amiloride on the neural salt response have shown the likelihood that a sodium channel serves as a taste receptor. Recent studies in T. Sato's laboratory confirm earlier results, and provide direct evidence for the presence of apical membrane sodium channels in taste cells and a basolateral membrane sodium pump. Both taste bud cells and non taste bud cells will contribute to the stimulus-evoked transepithelial electrical response. As a measure of the commonality of mechanism it would be valuable to correlate transepithelial potentials or currents under voltage clamp across a defined lingual area with the integrated neural response from the same area. We have done this in the rat using a single-sided Ussing chamber. We find that under short-circuit a sodium chloride stimulus evokes an initial fast inward current and conductance increase followed by a slow inward current and a These currents and slow conductance increase. conductance changes coincide respectively with the rising phase and the adaptation phase of the neural response. The slow phase current rises to its asymptote with approximately the same time course as neural adaptation. Both phases of the course as neural adaptation. Both phases of the current are reduced by amiloride to the same extent as the neural response. The slow component is blocked by ouabain in vitro. The results indicate that receptor cells: 1) are depolarized by an inward sodium current from the apical side, and 2) repolarized by an analysis of the contraction of the contracti electrogenic pump on the basolateral side.

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Effects of Potassium Channel Blockers on Rat Chorda Tympani Nerve Responses. M. KIM and C.M. MISTRETTA (Nursing and Dentistry, Univ. Michigan, Ann Arbor, MI 48109)

demonstrated results that aminopyridine (4-AP), a potassium channel blocker, depresses chorda tympani nerve responses to KCl in rat. To characterize further the effect of 4-AP and learn whether other potassium channel blockers alter the KCl taste response, we used five K-channel blockers. Taste responses were recorded electrophysiologically from the chorda tympani nerve and measured in the presence of each blocker at various concentrations. each blocker at various concentrations. Ten ml of 0.05M KCl dissolved in distilled water was applied to the tongue and followed immediately with an application of 10 ml 0.05M KCl in blocker. A ratio was calculated of the response to KCl in the presence of the blocker, relative to the response in water alone. As a control, this ratio was compared to the ratio obtained from 2 consecutive applications of 0.05M KCl in water. 4-AP was about 80% effective as a blocker at 5mM concentration. Responses to KCl concentrations lower than 0.25M were reduced more than those to higher concentrations. 5mM 4-AP produced a 40% reduction in the responses to 0.05M KCl and KBr, a 30% reduction for CsCl and KH₂PO₄, and 20% for RbCl and NH₄Cl. Responses to 0.05M NaCl and 0.5M sucrose were not affected. These results indicate a similar ion permeability sequence for the proposed channel as that reported in other excitable cells. Responses to 0.05M KCl apparently were not reduced by BaCl₂ (1 0.05M KCl response by about 30%, but also reduced the 0.05M KCl response by about 30%, but also reduced responses to NaCl. These data suggest that 4-AP sensitive K-channels have a role in transduction of taste responses to potassium. (Supported by N.S.F. Grant BNS 8311497.)

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Transport of Physiology), V.F. HOLLAND (Dept. of Physiology), Duke University, Durham, NC 27710.

Electrical and morphological measurements on isolated dog tongue between the tip and circumvallate papillae were performed bilaterally up to 44 days after unilateral tran-section of the CT nerve. The electrical studies were performed in an Ussing chamber in which the mucosal and serosal surfaces were bathed in solutions of 0.5 M NaCl and Ringers respectively. The electrical measurements showed that both sides were affected with the same time course, and to about the same extent. Over a period of 44 days the change in the short circuit current density, Isc, is Isc = I^{T} sc + I^{T} sc, where I^{T} sc = $97.5 \exp[(3-t)/15.5]$ for days 3 to 44 and I^{T} sc = $97.5[1 - 2.7 \exp(-t/16.5)]$ for days 17 to 44. Over this entire period, 0.1 mM amiloride inhibited Isc about 50%. After 17 days, Isc is minimum (about 40% of control) whereupon it increases to its control value at 44 days. These two processes represent the epithelial responses to CT (and TB) decay and regeneration, respectively. This interpretation is consistent with previously measured degeneration times (for taste buds) and regeneration times (for CT nerves) for fungiform papillae having a nerve stump of about 3cm. A plot of the open circuit potential vs t has a maximum at 34mV at 24 days. Morphological studies indicate that no pronounced changes were seen in either the number or diameter of the taste buds up to 18 days after transection. However, at 21 days, fewer and smaller taste buds were found on the ipsilateral side and none were present at 29-31 days after the lesion. After 21-31 days, taste buds on the contralateral side were fewer in number but normal in appearance. At 36 days postsurgery, several normal looking taste buds reappeared on the ipsilateral side and at 44 days their number had returned to that of control levels. These observations suggest that in dog tongue Na is transported across amiloride inhibitable cells that need not be organized into taste buds. This work was supported by grant NS20669.

Identification of the Apical Specialization in Dissociated Taste Cells by a Selective Staining Procedure. SUE C. KINNAMON and STEPHEN D. ROPER (Department of Anatomy, Colorado State University, Ft. Collins, CO 80523 and Rocky Mountain Taste and Smell Center, Denver CO 80262.)

We recently developed a technique for isolating taste cells from mudpuppy lingual epithelium. Isolated cells have membrane properties similar to taste cells of intact epithelium and are suitable for studying mechanisms of taste transduction using the patch-clamp recording technique (Kinnamon and Roper, ISOT IX, in press.) A limitation of the procedure is that after isolation, taste cells lose their characteristic morphology and apical membrane can no longer be distinguished from basolateral membrane. We now report that apical membrane can be identified in the isolated cells by incubating the surface of lingual epithelium with fluorescein-5-isothiocyanate conjugated wheat germ agglutinin (FITC-WGA) prior to dissociation. Isolated strips of lingual epithelium were washed with 100 mM ammonium chloride to remove mucus. After a 30 min treatment with collagenase (1.5 mg/ml), the apical surface of the epithelium was incubated in FITC-WGA (1 mg/ml) for 10 min. Taste buds were isolated by gently separating the non-gustatory epithelium from the the underlying connective tissue, and individual cells were isolated in Ca⁺⁺-free Ringer. Fluorescence was observed at the tips of elongate processes, presumably representing the apical region of the taste cells. Only a single patch of fluorescence was ob-served on the taste cell membrane. Several cells were unstained by the lectin; these were usually small spherical cells morphologically resembling basal cells. After incubating overnight at 40 C, taste cells became spherical. However, fluorescence was still restricted to a small patch of membrane. These results suggest that there is little redistribution of receptor membrane in isolated cells maintained at 40 C for several hours. This procedure will allow us to identify the apical specialization of taste receptor cells, so that taste stimuli can be directly applied to the receptor membrane.

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Behavioral and Physiological Responses to ATP Suggest Mediation by P₂- Like Chemoreceptors. RICHARD K. ZIMMER-FAUST (Marine Science Institute, University of California, Santa Barbara, CA 93106), RICHARD A. GLEESON, WILLIAM E. S. CARR (C. V. Whitney Marine Laboratory, University of Florida, St. Augustine, FL 32086)*

When released from the tissues of injured prey organisms, the purine nuclotide ATP serves as a potent stimulant of feeding-related behavior in the California spiny lobster, Panulirus interruptus (Zimmer-Faust. 1987. Biol. Bull. in press). ATP-sensitive olfactory cells with physiological properties similar to P₂-type purinergic receptors are known to exist in another species of lobster, the Florida spiny lobster, P. argus (Carr et al. 1986. J. Comp. Physiol. 158: 331-338). The current study employed behavioral assays of P. interruptus to determine if the responses to ATP analogs suggested mediation by P₂-like chemoreceptors. These assays were complemented by electrophysiological studies to identify ATP-sensitive cells in the olfactory organ of this species, and to determine if these cells had proposities similar to those identified previously in P. argus.

similar to those identified previously in P. argus.

The following results indicate that the behavioral response of P. interruptus to ATP may be mediated by P2-like chemoreceptors: 1) behavioral stimulation shows a potency sequence of ATP > ADP > AMP or adenosine; 2) behavior is evoked by nucleotide triphosphates with modifications in either the purine or ribose moieties; and 3) behavior is evoked by the slowly degradable ATP analogs, &, Y-imido ATP (AMPPNP) and \$, Y-methylene ATP. Extracellular recordings from single chemosensory cells show that ATP-sensitive cells do indeed exist in the antennule of \underline{P} . Interruptus. Like the ATP-sensitive olfactory cells identified earlier in \underline{P} . argus, these cells have a potency sequence of ATP > ADP > AMP or adenosine, exhibit brief response durations with low response intensities, and are activated by the slowly degradable analog AMPPNP. The above results suggest that the behavioral response of P. interruptus to ATP is mediated by a P2-like chemoreceptor, and that this particular type of chemosensory cell is common to two geographically isolated congeneric species of decapod crustaceans. *Supported by NSF Grant BNS-8607513.

PEPTIDE PHEROMONES SYNCHRONIZE CRUSTACEAN EGG HATCHING AND LARVAL RELEASE. Dan Rittschof, Richard B. Forward, Jr. and Mona C. DeVries. (Duke University Marine Laboratory, Beaufort, NC 28516.)

Egg hatching of an estuarine crab is precisely timed with respect to environmental cycles. At the time of hatching, pheromones are released from hatching eggs which induce the female to perform stereotypic larval release behavior in which she vigorously pumps her abdomen. This action breaks open the attached unhatched eggs and results in the synchronized release of larvae. The pheromones are a group of small peptides. Exposure to synthetic peptides induces the female to perform her larval release behavior. Concentrations of synthetic peptides evoking responses (thresholds from 1.5 x 10⁻¹⁰ to 1.8 x 10⁻⁶ M) are consistent with those predicted from compositional analysis of the native pheromones. The response to the pheromones is specific and graded. Assay of a systematically varied compositional series of peptides evokes responses that varied in threshold based upon the nature of the amino acid residues. Lowest thresholds were evoked by neutral-basic peptides with arginine at the carboxy terminal. without the general neutral basic form were inactive. The composite amino acids of active peptides initiate pumping behavior only at pharmacological concentrations (above 10 M). Thus, these results indicate that specific small peptides act as crustacean pheromones. These peptides are similar to the active site of leucocyte attractant peptides generated by the mammalian complement system.

High olfactory sensitivity of mature male goldfish to prostaglandins, presumed spawning pheromones. PETER W. SORENSEN (Department of Zoology, University of Alberta, Edmonton, Alberta, Canada), TOSHIAKI J. HARA (Department of Fisheries and Oceans, Freshwater Institute, Winnipeg, Manitoba, Canada), and NORMAN E. STACEY (Department of

In teleost fish and other vertebrates prostaglandins are synthesized during ovulation to promote follicular rupture. Titres of prostaglandin F2 α are elevated in both the serum and ovarian fluid of recently ovulated goldfish. High circulating levels of prostaglandin F2 α are believed to enter the brain to trigger female spawning behavior in goldfish, synchronizing sexual receptivity with the presence of ovulated eggs. It now appears that prostaglandin F2 α plays an additional role in synchronizing postovulatory reproductive events in goldfish: it "escapes" to the water in ovarian fluid to serve as a potent pheromone stimulating

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the male's olfactory system.

The olfactory sensitivity of mature male goldfish to a variety of prostaglandins and their metabolites was measured by underwater electro-olfactogram (EOG). All prostaglandins were detectable, but only prostaglandin F2cc and its metabolite, 15-keto-prostaglandin F2cc, evoked notable responses. The detection threshold for prostaglandin F2cc was approximately $10^{-9}\,\mathrm{M}$ and responses to it saturated at $10^{-9}\,\mathrm{M}$ at a magnitude 3-4 times that elicited by $10^{-9}\,\mathrm{M}$ L-serine. 15-keto-prostaglandin F2cc had a detection threshold of $10^{-11}\,\mathrm{M}$ and responses to it saturated at $10^{-9}\,\mathrm{M}$ at a response magnitude 1-2 times that elicited by $10^{-9}\,\mathrm{L}$ -serine. Cross-adaptation experiments demonstrated that these compounds are detected by different receptor sites and that these sites differ from those which recognize the pre-ovulatory pheromone 17cc,20p-dihydroxy-progesterone, as well as a bile acid and L-serine. Peripheral responses to prostaglandin odors were confirmed by electrophysiological recording from the medial olfactory tract.

Funded by the Alberta Heritage Foundation for Medical Research, Department of Fisheries and Oceans Canada, and the Natural Sciences and Engineering Research Council of Canada.

Pheromone Components and Active Spaces: What do Male Moths Smell and When Do They Smell It? C. E. Linn Jr., and W. L. Roelofs (Dept. Entomology, NYS Ag Expt Station, Cornell Univ., Geneva, NY 14456).

Studies were conducted in a sustained-flight tunnel with the redbended leafroller, cabbage looper, and Oriental fruit moths, and in the field with the latter species to determine whether male response downwind of a female is initiated by the major, or most abundant, component of the $% \left(1\right) =\left(1\right) \left(1\right) \left$ pheromone, or by the complete female-released blend. In the flight tunnel males of all three species initiated upwind flight in significantly higher percentages to the complete blend of components, at all dosages, compared to the major component alone. In the field male Oriental fruit moths responded in greater numbers and at significantly greater distances to the 3-component blend of this species, than to the major component alone. Our studies support the hypothesis that the active space of a multicomponent pheromone is a function of male perception of the female-released blend of components, rather than the major component alone, and that so-called minor components have a greater impact on male behavior far downwind of a female than previously thought. This conclusion is of particular importance because it represents a reversal from that presented in earlier studies with each species. The reversals came about after reinvestigations showed that the female produced blends were different and more complex than those previously identified and used in behavioral studies. We now realize that these were partial blends or ones containing different ratios of components than those released by females, but that had proven to be very attractive in field screening trials. It is clear that whereas these partial blends can elicit certain behaviors, the responses are inferior, both qualitatively and quantitatively to those exhibited with the femalereleased blends. Our conclusion represents an important general principle, and one that is most consistent with the paradigm concerning the function of multicomponent pheromones as species-specific mating signals.

NOTES

72 "Tuning" and "Noise" in Chemoreception: the Acoustic Analogy. JELLE ATEMA (Boston University).

In chemoreception research a great deal of attention has been given to the determination of effective stimulatory compounds and their mode of action on the receptor. This first aspect of chemoreception draws on models developed in pharmacology and, recently, molecular biology.

Far less effort has gone into the study of chemical signal detection. As in all other receptor systems, chemoreceptors must filter relevant signals from a noisy environment. This analysis of the function of chemoreceptor cells and organs requires thorough understanding of the natural distribution of chemical stimuli. (Atema, J. 1987. Distribution of Chemical Stimuli. In J. Atema, R.R. Fay, A.N. Popper, W.N. Tavolga, eds. Sensory Biology of Aquatic Springer-Verlag, New Animals. York. 29-56.) Specifically, to appreciate how signals are embedded in noise, one must have a quantitative model of the dynamics of spatio-temporal stimulus patterns. Since the stimulus environment places important constraints on receptor function, receptor filter properties should match stimulus features by which signals can be discriminated from noise. chemoreception can draw on models developed in acoustics, particularly with regard to "noise" and "tuning". However. parallels must be considered carefully to avoid confusing terminology.

I will discuss the following concepts for comparison between chemical and acoustic reception: environmental background noise, signal-to-noise ratio, spectral tuning, temporal tuning, stimulus-response function, effect of noise on stimulus-response function, and mixture suppression.

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73 Difference Thresholds for Odor Detection by an Aquatic Animal. RICHARD K. ZIMMER-FAUST and GORDON SMYTH (University of California at Santa Barbara)

Substances naturally attractive to aquatic animals frequently occur at high ambient concentrations. Olfactory competency may have evolved to detect slight differences in odor relative to background "noise". We report, for the first time, behavioral thresholds of an aquatic animal to a feeding stimulant taking into account natural background levels measured at the time of testing. Chemical determinations were made using high pressure liquid chromatography. The behaviors assayed were those indicating odor detection and appetitive feeding by the spiny lobster, Panulirus interruptus. Dilution associated with stimulus introduction was monitored by injecting fluorescent dyes, then recording fluorescence using fiber optic probes attached to olfactory (antennules) and taste (pereiopod dactyls) appendages of unrestrained animals. Recorded fluorescence was converted to an analog voltage signal, digitized and stored on a microprocessor at 10 msec intervals. This is roughly the latency of "real" animal chemoreceptors. Dose-response functions were then modelled using maximum likelihood procedures and logistic regression. For these conditions, we found: (1) lobsters to detect stimulant concentrations that were $\le 1\%$ greater than ambient, and (2) depending on whether feeding is assumed initiated by receptors on olfactory or taste appendages, lobsters begin feeding at concentrations two to five times greater than ambient. The difference threshold for detection by lobsters is below those reported for terrestrial animals and compares favorably to the competency of vertebrate visual and auditory systems. Threshold for lobster feeding was further reduced by mixing attractants structurally dissimilar. We present mathematical arguments that feeding thresholds may be related to the degree of reduction in uncertainty of a chemical signal, given that behavior has evolved under conditions of environmentally stochastic · chemical "noise".

Supported by a grant from the Whitehall Foundation.

When is Chemoreception Useful in the Nutrition of Grazing Microzooplankton? M. LEVANDOWSKY (Haskins Labs of Pace University, N.Y., NY 10038), J. KLAFTER and B. S. WHITE (Exxon Research and Engineering Co., Annandale, NJ 08801)

It is argued that, in still water, the best searching strategy for a swimming microzooplankter (e.g., a ciliate or a phagotrophic flagellate) is based on (a) a random walk described by a Levy distribution (instead of a normal distribution), coupled with (b) a chemosensory response in the immediate vicinity of food particles. In isotropically, homogeneously turbulent waters, on the other hand, it is concluded that (a) the kinetic details of swimming behavior are relatively unimportant, as long as some kind of random swimming occurs, and (b) chemosensory responses are much less useful than in still water. Some preliminary data with ciliates are presented that are consistent with these predictions.

75

Palato-Lingual Taste Responses of the Mongolian
Gerbil. MOHSSEN S. NEJAD and LLOYD M. BEIDLER
(Department of Biological Science, The Florida State
University, Tallahassee, FL 32306)

We previously reported that the rat greater superficial petrosal (GSP) nerve, contrary to the chorda tympani (CT) nerve is highly responsive to sucrose and reverse is true for sodium chloride. The hamster GSP and CT nerve responses to sucrose and NaCl were shown to be relatively similar (Nejad and Beidler, '85; '86). For these reasons, we electrophysiologically studied the palato-lingual (GSP and CT) taste responses of 12 male and female Mongolian gerbils (Meriones unguiculatus) to some chemical stimuli. Our observations indicated that the GSP nerve of the gerbil like the other two species of Rodents (rat and hamster) was extremely active and responsive to chemical stimulants. However, the order of the relative integrated response magnitudes of the GSP and CT nerves of the gerbil to the tested chemical stimuli was different than those of the rat and the hamster. For example, in the gerbil CT nerve an equimolar concentration of sucrose and NaCl solutions induced approximately the same integrated neural responses (sucrose/NaCl = 1). However, the GSP nerve of the gerbil was more responsive to 0.1M sucrose than to 0.1M NaCl (sucrose/NaCl ≈ 5). In comparison, the GSP nerve response ratio of 0.1M sucrose to 0.1M NaCl of the three Rodents were: gerbil (~ 5) > rat (~ 2) > hamster (1). The order of the palato-lingual response magnitudes of the gerbil to the four basic taste qualities was as follows: GSP, 0.1M sucrose > 0.01M citric acid >, 0.02M Na-saccharin > 0.1M NaCl >, 0.01M Q.HCl; CT, 0.02M Na-saccharin >, 0.1M sucrose > 0.1M NaCl > 0.01M citric acid > 0.01M Q.HCl. The relative order of the integrated response magnitudes to a series of 0.3M chloride salts was: GSP, LiCl >, NaCl > NH4Cl > CaCl₂ > KCL; CT, LiCl >, NaCl > NH4Cl ≅ CaCl₂ > KCl.

Responses in Single Fibers of the Hamster Chorda
Tympani Nerve. THOMAS P. HETTINGER & MARION E.
FRANK (University of Connecticut Health Center,
Farmington, CT 06032).*

Amiloride, a potent inhibitor of sodium transport in epithelial cells, has been found to be an effective probe for studying mechanisms of taste reception. Of the three major electrophysio-logically defined classes of neurons in the hamster chorda tympani nerve, namely, those responding best to sucrose, NaCl or HCl applied to the tongue, only the NaCl-best neurons inhibited by amiloride. Complete inhibition of responses to NaCl is rapid, competitive, and achieved by concentrations of amiloride in the micromolar range, properties consistent with the hypothesis that activation of NaCl-best fibers is mediated by the conductive sodium transport pathway occurring in the apical membrane of the receptor cells. The Na/H antiporter is not likely involved in the inhibition, since it requires millimolar concentrations of amiloride. Responses to LiCl and HCl in NaCl-best fibers are also suppressed by amiloride, while responses to HC1 and NaC1 in HC1-best fibers are not. Effective stimuli for NaCl-best fibers all appear to use a common receptor, probably equivalent to a sodium channel, while HC1-best fibers employ a different receptor system. The single-fiber results are consistent with the previously obtained wholenerve data, which showed extensive, but incomplete suppression by amiloride of integrated responses to NaCl (Hettinger and Frank, AChemS 8, 1986). The current single-fiber data are, however, much easier to interpret because of the clearly divergent properties of the NaCl-best and HCl-best and lend further support for the classification of fibers as "natural" types.

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77 The Correct Method of Signal Processing For Whole-nerve Recordings. HARRY WMS. HARPER (Stauffer Chemical Co.), B. W. KNIGHT (Rockefeller University).

The use of "integrated" whole-nerve recordings to estimate response magnitudes has been widespread in taste research. However, the common practice of rectifying and averaging the AC-amplified nerve potential leads to serious, systematic underestimation of the mean firing rate of the neural population in the nerve. The correct underestimation of the mean fiffing face of the neural population in the nerve. The correct procedure is to average the squared nerve potential, to obtain the variance of the signal. These results follow directly from treatment of the whole-nerve potential as a Poisson shot-noise, to which the Central Limit Theorem and Campbell's Theorem may be applied. High-precision analogue simulations demonstrate the validity of this monolithic analogue analysis. Commercial practical basis multipliers form the for laboratory instrumentation with adequate dynamic range and more than adequate bandwidth. To a good approximation (at all but very low response levels), data obtained by rectifying and averaging can be corrected by simply squaring them.

Research conducted at Rockefeller University.

NOTES

78 Concentration-Response Functions of Single Chemosensory Fibers in the Hamster Superior Laryngeal Nerve. TAKAMITSU HANAMORI and DAVID V. SMITH (University of Cincinnati College of Medicine)

In several mammalian species, including humans, the laryngeal surface of the epiglottis contains a large number of taste buds. In hamsters, epiglottal taste buds represent 9.7% of the total (Miller & Smith, 1984). These chemoreceptors have an important role in the reflexive activities of coughing, swallowing and apnea. Electrophysiological studies of superior laryngeal nerve (SLN) fibers in a number of species show that these taste buds respond well to KCl, NH₄Cl, HCl, urea and water (Bradley, Stedman & Mistretta, 1983; Dickman & Smith, 1985). In the present study we investigated the responsiveness of hamster SLN fibers to a range of stimulus concentrations. Stimuli were 0.01 - 1.0 M sucrose, 0.01 - 1.0 M NaCl, 0.0003 - 0.03 M HCl, 0.0003 - 0.03 M quinine hydrochloride (QHCl), presented in 1/2 log step increments via a push-pull cannula at 0.1 ml/sec. All stimuli were dissolved in 0.154 M NaCl, which also served as the rinse. SLN fibers responded well to distilled water, to HCl and to NaCl at the lower and higher concentrations. Sucrose and QHCl were not very effective stimuli even at strong intensities. NaCl was the most effective stimulus at the lowest concentration level, HCl at the middle of the range, and NaCl and HCl were equally effective at the highest concentrations. These response characteristics are different from those in the hamster chorda tympani (CT) and glossopharyngeal (IXth) nerves. NaCl and sucrose are the most effective stimuli for CT fibers and QHCl and HCl for IXth nerve fibers. The acidity and tonicity of stimuli effective for SLN fibers may be significant for the reflexes triggered by chemical stimulation of the epiglottis.

79

Electrical Stimulation of the Autonomic Supply of von Ebner's Glands in the Rat. SUAT GURKAN and ROBERT M. BRADLEY (Dept. Oral Biol., School Dent., Univ. Michigan, Ann Arbor, MI 48109)

Von Ebner's lingual salivary glands drain into the clefts of the circumvallate and foliate papillae, which contain the majority of the lingual taste buds. Recently we have shown that injection of β -adrenergic and cholinergic agonists causes a 50% depletion of the secretory granules when administered alone, and total depletion when given simultaneously (Brain Res. 1987). Since these drugs are administered systemically, they can be effective at a number of locations. We have used electrical stimulation of the parasympathetic and sympathetic nerve supply to von Ebner's glands to determine whether the drugs are acting at the gland level. Rats were starved overnight to cause accumulation of the secretory granules in gland acini and then anesthetized. In one group either the sympathetic trunk (3 rats) or the glossopharyngeal nerve (3 rats) was stimulated electrically (20 Hz, 1msec, 4V for sympathetic and 50 Hz, 1 msec, 8V for glossopharyngeal) for 20 minutes. In a second group of 4 rats these nerves were stimulated for 1 hour. In each group two rats were sham operated as controls. In addition in 2 rats both the glossopharyngeal and sympathetic trunk were stimulated simultaneousely for 1 hour. Following stimulation the von Ebner glands were rapidly removed, fixed, embedded in plastic and sectioned at 1 μ M. Examination of the sections from all electrically stimulated glands revealed no apparent reduction in secretory granules. These results indicate that there are fundamental differences between the effectiveness of drugs and electrical stimulation on gland activity. Supported by N.I.H. Grant NS21764.

Electrical Stimulation of Rat Medullary
Neurons Controlling von Ebner's Glands THERESA A.
HARRISON and ROBERT M. BRADLEY (Dept. Oral Biol.,
School Dent., Univ. Michigan, Ann Arbor, MI
48109)

Injection of HRP into the circumvallate papilla produces retrograde labeling of a group of cells located just medial and dorsal to the rostral (gustatory) solitary nucleus (NST). It has been suggested that these are preganglionic control parasympathetic neurons that the postganglionic secretomotor output to the lingual von Ebner's glands (Bradley et al, Brain Res., 1985). We now have demonstrated that electrical stimulation of these neurons produces salivation. Maps were constructed of brain loci at which electrical stimulation (20 Hz, 0.5msec, for 10 s) with a low-impedance tungsten microelectrode did or did not produce saliva from the circumvallate and/or foliate papillae in Nembutal-anesthetized rats. Lesions were made at circumval the most saliva and/or lease at sites showing the most saliva and/or lowest salivation threshold. Saliva was evoked at low salivation threshold. Saliva was evoked at low intensities from loci in a limited brain region. At these "best" sites, the amount of saliva released was voltage dependent. Anatomical reconstruction of lesions and electrode tracks showed that "best" sites corresponded to the region containing labeled parasympathetic neurons after HRP injection in circumvallate papilla. Sites outside this area, even within the NST, did not evoke saliva, or did so only at higher intensities. Our results indicate that these neurons may function in central control of salivary secretion by the von Ebner's glands, and thus the fluid environment of taste buds in thus the fluid environment of taste buds in circumvallate and foliate papillae. The location of these neurons suggests that there may be important reciprocal interactions between taste and salivary neurons. Supported by N.I.H. Grant NS21764.

81 Cholecystokinin administration does not influence taste-evoked activity in rat NTS.
Barbara K. Giza and Thomas R. Scott (Department of Psychology and Institute for Neuroscience, University of Delaware, Newark, DE 19716)

Physiological factors associated with satiety have been shown to influence taste-evoked neural activity at several levels of the nervous system. Gastric distension, hyperglycemia and moderate hyperinsulinemia are all associated with a decrease in responsiveness to sugars and, less reliably, to other taste qualities. Cholecystokinin (CCK) is a gut hormone and putative neurotransmitter that has been found to induce satiety in a variety of species, including the human. While its effects are thought to be vagally mediated, the mechanism and site of CCK's action are unknown. There has been speculation that the satiety effect induced by its exogenous administration could be mediated by alterations in taste sensitivity. We monitored multiunit taste-evoked activity in the nucleus tractus solitarius (NTS) of anesthetized rats during intravenous administration of either plasma (controls) or 2.0 µg/kg CCK-8, a quantity sufficient to depress feeding significantly in behavior tests. Taste stimuli were 1.0 M glucose, 1.0 M fructose, 0.1 M NaCl, 0.03 M HCl and 0.01 M quinine HCl. We found no significant changes in responsiveness to any stimulus in the 30 min period following CCK administration. Possible interpretations of this result include 1) the dose of CCK may have been too small. 2) There could be an effect on taste responses that is not apparent This is improbable in the rat where NTS is an obligatory synapse and also receives centrifugal projections from both thalamo-cortical and ventral forebrain taste areas. 3)CCK may be a satiety factor whose mechanism does not involve the taste system. 4) CCK may not be a true satiety factor.

Taste Processing in the Rabbit: Generalization of Learned Taste Aversions. LESLIE L. WIGGINS (Oxford University), DAVID V. SMITH and ROBERT A. FRANK (University of Cincinnati)

Previous electrophysiological work on rabbits has demonstrated their potential for the study of taste mechanisms. The present experiment examined the behavioral similarities among several stimuli presented to rabbits. Fifteen stimuli served as both conditioning (CS) and test stimuli (TS): 0.1 M sucrose and fructose, 0.01 M Na-saccharin, 0.1 M NaCl, NaNO3, Na2SO4, KCl, NH4Cl and CaCl2, 0.001 MHCl, tartaric acid and citric acid, 0.003 Mquinine hydrochloride (QHC1), 0.3 M urea, and 0.0001 M sucrose octaacetate (SOA). Each of 45 animals was strongly (> 90% reduction in licks/10 sec) conditioned to avoid one of the stimuli and was then tested for its response to each of the others. Citric and tartaric acid and SOA were eliminated from the experiment because the animals never reached the 90% conditioning criterion. The mean number of licks to each of the stimuli was compared to values from control animals to derive mean percent suppression scores. Similarities among the stimuli were examined using hierarchical cluster analysis (BMDP1M) and multidimensional scaling (KYST). Strong mutual suppression occurred among the 3 sodium salts, among the 3 nonsodium salts, and between the 2 sugars. Stimuli with mixed tastes to humans, like Na-saccharin and urea, fell between the others in a 2-dimensional space representing these similarities. relationships varied to some extent depending upon whether the similarities among the test stimuli (TS) or among the conditioning stimuli (CS) were considered, primarily among those stimuli with mixed tastes. For example, when Na-saccharin was a TS it was located between the sugars and sodium salts in a 2-dimensional similarity space but was between the sugars and bitter-tasting stimuli (e.g., QHCl) when it was a CS.

83 A Freeze Fracture Study on the Apical Specializations of Taste Cells in the Mudpuppy, Necturus maculosus. T.A. CUMMINGS and S.D. ROPER. (Dept. of Anatomy, Colorado State University, Fort Collins, CO 80523).

The initial excitation in taste chemoreception is elicited by contact between chemical stimuli and apical specializations of taste cells. Integral membrane proteins, revealed in freeze-fracture replicas as intramembranous particles (IMPs), possibly serve as specific receptor sites and/or as ionophores which mediate this initial process in chemosensory transduction. We have demonstrated that light cells and dark cells possess morphologically distinct apical specializations and postulate that these differences may impart chemoreceptor specificity (Cummings, Delay, and Roper, 1987, submitted). To further substantiate this hypothesis, we are studying the membrane morphology of the apical specializations using the freeze-fracture replica technique.

The lingual epithelium from adult mudpuppies was fixed in 2.5% glutaraldehyde and individual taste papillae were dissected from the lingual epithelium under 25X magnification. Samples were cryoprotected with glycerin and frozen in liquid freon. Samples were then fractured in a Balzers 301 freeze-fracture device at -130°C and 10⁻⁷ mbar, replicated with platinum at a 40° angle, and rotary coated with carbon. Preliminary results indicate that taste cell types can be distinguished in freeze-fracture replicas. Experiments are underway to identify apical specializations belonging to dark and light cells and to determine whether important differences in IMP size, density, and distribution can be observed. These data may further elucidate the correlation between structure and function and thus enhance our understanding of the mechanisms involved in chemosensory transduction.

Supported by NIH grants NS20382, NS20486, and AG03340.

NOTES

84 <u>Ultrastructure of Gerbil Fungiform Taste Buds.</u> VAR L. ST. JEOR and JOHN C. KINNAMON. (Dept. MCD Biology, Univ. of Colorado, Boulder.)

As part of a comparative ultrastructural study of taste buds in vertebrates, we are examining gerbil fungiform taste buds. We are using the combined techniques of high voltage electron microscopy (HVEM) and computer 3-D reconstructions from serial micrographs.

Like mouse fungiform taste buds, the general shape of gerbil fungiform taste buds resembles a garlic bulb. Four basic cell morphologies have been observed: dark (type I), intermediate, light (type II), and basal or undifferentiated. Although the ultrastructural features of these taste buds are generally similar to other rodent taste buds. certain differences are conspicuous. Specifically, the electron-density of gerbil dark cells is much more extreme compared with dark cells in mouse fungiform, foliate and vallate taste buds. Gerbil dark cells also seem to exhibit less heterochromatin adhering to the inner leaflet of the nuclear envelope than is present in mouse foliate and vallate taste buds. Secondly, we have yet to observe any examples of the atypical mitochondria at gerbil taste cellnerve fiber appositions that are common in murine taste buds.

Nerve fibers contain large varicosities resembling those seen in mouse fungiform taste buds. In each taste bud several of these nerves reach to within 15 um of the taste pore. There is a striking paucity of synapses in these gerbil taste buds compared with murine foliate and vallate papillae.

We are currently reconstructing the nerve fiber arborizations and studying the patterns of synaptic connectivity within these taste buds.

This work was supported in part by grants from the Procter & Gamble Co. and NIH grants NS21688, RR-00592, PO1-NS30486. We wish to thank Dr. Bruce Oakley for providing gerbils for this study.

Relations Between Planar Dendritic Growth and Cytochrome Oxidase Activity in the Pontine Taste Area of Developing Rats. PHILLIP S. LASITER (Dept. Psychol., Florida Atlantic Univ.), DAVID L. HILL (Dept. Psychol., 85 Univ. of Virginia, Charlottesville), and DIANE A. MOORMAN (Dept. Psychol., Florida Atlantic University)

Dendrites of pontine taste area (PTA) neurons in rats exhibit extensive radial growth during the first five weeks of life. For instance, a threefold increase in average length of PTA dendrites is observed between the ages of ll days and 35 days. We have previously shown that dendritic growth in the PTA is well-correlated with the development of adult-like electrophysiological responses in that nucleus. The present studies evaluated the development of cytochrone oxidase (CO) activity in the PTA of developing rats. The cytochrome systems promote cellular respiration, electron transport and oxidative phosphorylation, and contribute to protein synthesis, vesicle formation, maintenance of resting membrane potentials, and rapid axoplasmic transport. Thus, CO reactivity in our histochemical studies was presumed to be at least one metabolic marker for developing neuronal activity in the PTA region. Histochemistry was performed on tissue derived from animals aged 8 - 120 days, and microscopic densitometric measurements were subsequently performed within the PTA region. A monotonic relationship was obtained between CO activity and age. Activity was lowest in animals aged 8 and 18 days, as compared to all other age groups, and did not differ reliably between these Conversely, CO reactivity age groups. significantly between the ages of 18 and 46 days. Activity was asymptotic, and not reliably different, between the ages of 46 days and 120 days. Comparison of data between studies indicate that there is a pronounced relationship between (1) the development of CO reactivity in the PTA, (2) radial growth of PTA dendrites, and (3) development of electrophysiological responses within the PTA. These data indicate that dendritic development and maturation of cytochrome systems serve a fundamental role in the ontogeny of taste responsiveness in PTA neurons.

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Organization and responses of facial lobe subnuclei in the channel catfish. Ictalurus punctatus. T. HAYAMA and J. CAPRIO (Department of Zoology and Physiology, Louisiana State University, Baton Rouge, LA. 70803).

The medullary facial lobes (FL) in freshwater ictalurid catfishes are the primary gustatory nuclei which receive facial nerve input in a somatotopic manner from taste and tactile neurons innervating the external body surface. Finger (1) reported three subnuclei in the FL of bullhead catfishes, Ictalurus natalis and I nebulosus, and Marui and Caprio $^{(2)}$ correlated the somatotopic organization of the FL in I. punctatus (Ip) with FL substructure in terms of three subnuclei. Recently, a precise somatotopy was described in the Japanese sea catfish, <u>Plotosus lineatus</u> (formerly anguillaris). Each FL of <u>Plotosus</u> is clearly divided into five subnuclei: four roughly equal, longitudinal columns which have input from four similarly-sized barbels, respectively, and a fifth larger column representing the face-flank(3). Anatomical and electrophysiological re-examination of FL substructure of Ip was performed in order to correlate subnuclei structure with somatotopy. The results indicated that rostral to intermediate portions of the FL in Ip were divided into five subnuclei representing four barbels and the face-flank as seen in Plotosus, although the subnuclei of Ip were less distinct. The subnucleus receiving maxillary barbel input was relatively larger and the subnucleus receiving nasal barbel input was relatively smaller than the corresponding subnuclei of Plotosus. These findings correlate well with the difference in relative sizes of these barbels in the two species. Electrophysiological examination, further showed that amino acid sensitive regions within the maxillary barbel subnucleus were generally confined to the dorsal half of the subnucleus, while somatosensory responses were observed throughout with maximal activity in the ventral subnuclear portions.

(1) T. Finger (1976) J. Comp. Neurol., 165:513-526

(2) T. Marui and J. Caprio (1982) <u>Brain Res.</u> 231:185-190 (3) T. Marui et al. (1985) Achems VII Abstr. #125

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Fungiform Taste Pore Quantification in Living Rabbits. INGLIS MILLER, JR. (Bowman Gray Sch. Med., Wake Forest Univ.)

Visualization of pores in living fungiform papillae permits quantification of the same taste receptors over time. Both humans and rabbits have fungiform papillae with 0-15 or more taste buds. Our objective is to develop a procedure to identify and quantify taste buds in living subjects which can be used both with humans and laboratory animals. Male, New Zealand rabbits are anesthetized with Halothane through an endotracheal tube, and the ventral surface of the tongue tip is visualized through a videomicroscopy system. Methylene blue dye is applied to the tongue surface, and an array of papillae is mapped. Individual papillae are identified by their size, shape and position in the array, and they are assigned an identification number. Finite dots can be identified on the surface of individual fungiform papillae. The image of each papilla is stored on videotape, and a digitized map of papillae and their putative pores are stored on a computer. Three series of experiments are underway. In acute experiments, an array of papillae and their putative taste pores are mapped in living rabbits; the animal is sacrificed; and the tongue is prepared for light microscopy. The region of tongue which was mapped is reconstructed, and the location of taste buds is compared with the configuration of dots from the living state. Overall, the rabbit tongue contains a mean total of 100 ± 13 (R=12 rabbits, S.D) fungiform papillae. One half of the ventral tongue tip contains $25.9 \pm 9.6(R=11)$ papillae or 1.27 ± 0.5 pap/mm² (N=285 pap, R=11 rabbits). Papillae contain a mean of 5.46 ± 1.2 (N=285 pap, R=11, S.D) dots/papilla or 6.51 dots /mm² (N=730 dots, R=6). Histological reconstruction is in progress, but preliminary results show a correlation of .94 between dots (putative pores) and confirmed taste buds. In chronic experiments, living rabbits are to be examined at intervals of 6 weeks to determine if a change in the count or position of putative pores occurs. In another series, taste pores are examined at weekly intervals after unilateral crush of the chorda tympani nerve.

(Technical assistance was provided by Frank Reedy, Jr. work is supported by NIH Grant NS 20101 from NINCDS.)

An Assessment of Behavioral Olfactory Discrimination in the Spiny Lobster Using an Habituation Paradigm. PETER C. DANIEL and CHARLES D. DERBY (Department of Biology., Georgia State University, Atlanta, Georgia 30303)*

A non-associative habituation conditioning paradigm was usedas an alternative to an associative aversion conditioning paradigm (see Fine et al, 1986, Chem.Senses, 11, 597; Fine-Levy and Derby, abstract at this meeting) in order to examine behavioral aspects of quality coding in olfaction in the Florida spiny lobster (<u>Panulirus argus</u>). Magnitude of search response to two concentrations of four artificial mixtures that mimicked natural food extracts of crab, oyster, mullet, and shrimp and to artificial sea water (control) was measured in five lobsters before and immediately after habituation to the crab artificial mixture. A 66 to 100% decrease in response to the crab mixture was accomplished through 2-min presentations of 5 ml of alternating concentrations (0.05 and 0.5 mM) of stimulus, repeated every 5 min for a total duration of 3 to 3.5 h. Lobsters were able to differentiate between mixtures, although responses to all chemicals decreased following habituation by at least 42%. At the 0.5 mM concentration, habituation was greatest to crab (88%), followed by shrimp (55%), oyster (48%) and mullet (42%). A similar pattern emerged at the 0.05 mM concentration, although habituation was higher for all mixtures: crab oyster (67%), and mullet (64%). crab (94%), shrimp (76%), t (64%). These patterns of those reported for aversion discrimination parallel conditioned lobsters. We are currently using the habituation paradigm to test discrimination between single compound stimuli.

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Contribution of Salivary Level of Glutamate to the Cause of Umami Taste of Ribonucleotides.

Shizuko YAMAGUCHI, Mika KIMURA & Yasushi KOMATA (Central Reseach Laboratories, Ajinomoto Co., Inc.)

It is well known that the intensity of umami of nucleotides is synergistically enhanced by MSG. Since human saliva contains small amount of glutamic acid, it would be possible that the apparent umami of nucleotides is a consequence of salivary glutamate. The purpose of this study is to examine how does the salivary level of glutamate affect the intensity of umami of nucleotides. The detection threshold (RL) and the differential threshold (DL) were measured in simple aqueous solution and various concentrations of MSG solutions.

RL of IMP was greatly lowered and DL was decreased (steepened) with increasing concentration of MSG. The concentration of MSG of the solution which yielded the same RL of IMP as that in aqueous solution was estimated as 10^{-5} M. It was exactly the same as that yielded the same DL in MSG solution as that in aqueous solution, i.e., RL and DL of IMP in aqueous solution were regarded as those caused under the presence of 10^{-5} M of glutamate. The content of glutamate in saliva was determined as 1.8 X 10^{-5} M (0.33mg% as MSG). This suggested the possibility of a peripheral effect of salivary glutamate on the mechanism underlyning the taste of IMP.

NOTES

THE SWEETNESS OF BIHARY MIXTURES OF SUCROSE AND STEVICSIDE: AN INFORMATION INTEGRATION ANALYSIS. Susan Hubay & Robert A. Frank (Dept. of Psychology, Univ. of Cincinnati).

Angerson's (1981) information integration approach was used to evaluate the sweetness, additivity of sucrose and stevioside, a low calorie sweetening agent derived from the leaves of <u>Stevia rebaudiana</u>. The commercial potential of stevioside is currently being explored by mixing it with other sweeteners in soft arinks, chewing gums and other foods. It has been reported to act synergistically with several sweeteners (i.e., the observed sweetness of the stevioside mixture is greater than what would pe predicted based on the compined sweetness of the mixture components). This claim was evaluated by assessing the sweetness of 0.02, 0.05 and 0.09% w/v steviosice (Maruzen Chemicals, Osaka, Japan), 0.1, 0.25 and 6.5 M sucrose and the factorial combinations of these two sweetners. The subjects rated the sweetness of the stimuli on a 21 point category scale. Ail stimuli were rated twice per session using the sip and spit method (stimulus volume = 5 ml). A tap water rinse and 30 sec intertrial interval followed the presentation of each stimulus. A psychological integration function was constructed for the stevioside/sucrose mixtures, and this function revealed a tendency for the lower concentrations of the sweeteners to produce greater than expected sweetness (i.e., synergy), while the nigher concentrations showed a tendency toward less than expected sweetness (i.e., suppression). The observed synergy and suppression were defined in terms of the predictions of a simple additive sweetness model. However, since a substance mixed with itself does not exhibit perfect additivity, but rather, psychophysical compression, one must evaluate the sweetness of the mixtures following a correction for the compression expected in the psychophysical functions of stevioside and sucrose. Following this correction, it was found that the stevioside/sucrose mixtures exhibited synergy at low concentrations, but only the expected compression at high concentrations. We are presently using the information integration approach to assess the interactions of other sweeteners.

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91 An Introduction to Charm Analysis. T. E. ACREE, A. B. MARIN & J. BARNARD (Cornell University, Geneva)

Charm analysis is a technique for the determination of odor detection thresholds of individual compounds in complex mixtures. The technique presents pure aroma compounds in a stream of cool, humidified air for subjects to smell. During the procedure, the subject sits at a video terminal and records the instant that an odor is detected, the quality of the odor perceived, and the instant that the smell disappears. The aroma compounds are very pure because they are added to the air stream as gas chromatographic effluents. Furthermore, the chromatograph completely volatilizes the compounds so that the amount injected relates directly to the concentration of the odor stimuli in the air stream.

Repeated analyses at several serial dilutions of a stimulus mixture yields a quantitative measure of odor intensity called charm. It can be shown that the charm data integrated over the elution time of an aroma compound is a kind of odor unit, that is, the amount of compound injected in the gas chromatograph divided by the odor detection

threshold for the subject.

Using a single subject, we can quantify the changes in concentration of odor-active compounds. For example, charm analysis was used to quantify β -damascenone at the femtogram level during the isolation of its precursor from grapes and apples. Using multiple subjects, a single mixture of aroma chemicals was used to study variation in response to odor (Marin et al., AChemS 9, 1987). Furthermore, the associations between odor descriptors and elements of the charm chromatogram made by the various subjects can relate language to perception and stimulus concentration. For example, "off-odor" was used as a descriptor in a charm analysis to direct the quantification of off-odor causing chemicals in a commercial beverage.

Perceptual Properties of Pungent Odorants. COMETTO-MU NIZ, J.E.; GARCIA-MEDINA, M.R.; CALVIÑO, A.M. and HERNANDEZ, S.M. (Laboratorio de Investigaciones Sensoriales, CONICET Esc. Sal. Públ., Fac. Medicina, UBA, C.C.53, 1453 Buenos Aires, Argentina.

Supported in part by a grant to project #9082-03 from the CONICET, República Argentina.

A series of concentrations of each of two pungent odorants were presented alone or in the presence of different backgrounds of the other irritant, in order to explore functional properties of the olfactory and common chemical senses.

Stimuli comprised formaldehyde (at 1.0; 3.5; 6.9 and 16.7 ppm), ammonia (at 210; 776; 1,172 and 1,716 ppm) and their 16 possible binary mixtures. The stimulus-response functions (approximated by power functions) for each substance showed fairly constant exponents regardless of whether the irritant was presented alone or with various fixed levels of the other irritant. The average exponent value (\pm S.D.) for formaldehyde in the various conditions was: 0.23 (\pm 0.04), while for ammonia was: 0.63 (\pm 0.05). The latter showed, in logarithmic coordinates, a consistent upward concavity. At low concentrations, the total perceived intensity of the binary mixtures showed hipoadditivity, i.e. the total perceived intensity of the mixtures was significantly lower than the sum of the perceived intensities of the perceived intensity of the perceived inten ceived intensities of their components. At intermediate concentrations the mixtures showed simple additivity and at high concentrations, hiperadditivity. When subjects made estimations of the olfactory and common chemical components of the evoked sensations it was seen that perceived pungency presented higher exponents than perceived odor. Furthermore, odor was always hipoadditive while pungency was, mainly, ad ditive and, also, hiperadditive. It seems that the total perceived intensity of the mixtures is processed like odor mixtures at low intensities and like pungent mixtures at high intensities.

3 STIMULUS RESPONSE FUNCTIONS OF RAT OLFACTORY BULB NEURONS. T.A. Harrison*, J. W. Scott, R.K. Conine and D.P. Wellis. (Department of Anatomy & Cell Biology, Emory University, Atlanta, Georgia 30322).

To study sensory processing by the olfactory bulb, we recorded single mitral/tufted cells and interneurons while presenting graded series of odorant concentrations. odorant most often used was amyl acetate in concentrations of 0.04 to 4 micromoles per liter. Series of six concentrations were completed for 61 cells. The stimulus series were repeated for 27 cells and concentration series were run with two or more odors for 11 cells. These neurons showed systematic response changes over the stimulus concentration series as assessed by the method of Harrison and Scott (J. Neurophysiol. 1986, 56, 1571-1590). stimulus-response functions are expressed as degree response change relative to the variability during stimulus These stimulus-response functions were stable repetition. with repeated stimulus series. The analysis is sensitive to temporal pattern, so that orderly response functions were obtained even when the total spike count did not vary systematically with stimulus concentration. responds to several odors, the responses to different odors often differ only in threshold or in the steepness of the stimulus-response function. However, some cells show two distinct sets of temporal patterns (response-concentration profiles), one profile for some odors and a second profile for other odors. The fact that these profiles are distinct at all suprathreshold stimulus concentrations demonstrates the effect of odorant quality differences that are independent of differences in stimulus strength. Similar observations have been reported in the past but without controls for stimulus intensity. These results suggest the presence of antagonistic circuits influencing a single output cell.

*present address, Dept. Oral Biology, Univ. of Michigan, Ann Arbor, MI 48109. Supported by NS-12400 and a Biomedical Research Support Grant through Emory University. Stimulus Intensity Discrimination by Lobster Olfactory Receptors. B.R. JOHNSON (Cornell University, Ithaca, NY), R. VOIGT, C.L. MERRILL and J. ATEMA (Boston University Marine Program, Marine Biological Lab., Woods Hole, MA).

Lobster prey contains concentrations of chemical stimuli in the high mM range. To stimulate feeding behavior, mixtures of these chemicals must be perceived against seawater backgrounds of uM to nM concentrations of the same and other compounds. Thus, olfactory chemoreceptors in the lateral filament of lobster antennules may have to provide intensity information for different mixture components over at least a six-fold log concentration range. We examined olfactory receptors sensitive to hydroxy-L-proline (Hyp) to determine their sensitivity and their ability to discriminate different concentrations of Hyp over a large concentration range. Single cell stimulus-response (SR) curves were calculated from extracellular recordings. Hyp cells (21) were tested at six 1-log concentration steps beginning with $3 \times 10^{-8} \mathrm{M}$ Hyp. We measured the number of spikes in the first 500 ms of a response. Hyp cells had very shallow SR curves, with rates from 0 to only about 15 spikes/500 ms over a 5 log concentration range of Hyp. Statistical analyses based on mean population response magnitude for each concentration showed differences between all test concentrations except the highest two, $3 \times 10^{-3} M$ and $3 \times 10^{-4} M$ Hyp. When responses were normalized to the best response in each cell, Pearson's correlation coefficient (=0.01) demonstrated that every test concentration evoked a different response pattern across the Hyp cells. We cannot ignore the possibility that in some cases a neural code for intensity based on patterns of activity across sensory receptors may yield better discrimination than codes based strictly on mean response magnitude. Population codes may be especially important for sensory cells with low maximum firing rates functioning over wide ranges of stimulus intensity. Neural patterns for stimulus intensity within narrowly tuned cell populations may combine with patterns of stimulus identity across differently tuned populations to code the quality of natural stimulus occurring mixtures.

Supported by NSF (BNS 8512585) and the Whitehall Foundation

PROLONGED INHIBITION OF ANTERIOR COMMISSURE INPUT TO THE OLFACTORY BULB BY STIMULATION OF THE DIAGONAL BAND. W.T.Nickell and M.T. Shipley (University of Cincinnati College of Medicine.)

The two main olfactory bulbs (MOB) are connected by a large and at least partially ordered commissural system, which terminates upon granule cells in the granule cell layer (gcl). We report here that the strength of this synaptic input is significantly modulated by the centrifugal projection from the nucleus of the diagonal band (DB).

Anaesthetized adult male rats were used. Stimulating electrodes were placed stereotaxically in the ipsilateral DB and in the rostral wing of the contralateral anterior commissure (ACc). Stimulation of ACc results in a negative field potential in gcl, which reflects an excitatory post-synaptic potential (epsp) in the granule cells. We previously described the field potential in MOB caused by DB activation. Stimulation of DB at 10 Hz for several seconds results in a large potentiation of this response. We now report that this same stimulation paradigm results in a powerful and long-lasting inhibition of the ACc response. After stimulation of the DB sufficient to produce potentiation, the ACc response is reduced to 15% of the control value and does not completely recover for more than 20 seconds. Shorter periods of DB stimulation result in faster recovery. During the period of inhibition, a burst of shocks to ACc will cause the response to recover more quickly.

At least two mechanisms could account for the inhibition. (i) A postsynaptic increase in conductance of granule cells (eg. postsynaptic inhibition) would shunt excitatory synaptic currents. Alternatively, (ii) presynaptic inhibition of ACc terminals synapsing onto granule cells would also inhibit the recorded response. The DB projection to the bulb contains a cholinergic component; exogenous application of acetylcholine in hippocampus produces presynaptic inhibition of both afferent inputs and intrinsic inhibitory interneurons. The inhibition of ACc by DB reported here may represent an analogous mechanism produced by synaptic release of acetylcholine. Evidence for cholinergic presynaptic inhibition by stimulation of a cholinergic projection has not been previously reported. The functional significance of this phenomenon remains to be determined, but may represent a mechanism for regulating intrabulbar communication.

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Morphological and Physiological Characterization of Individual Olfactory Interneurons in the Protocerebrum of the Crayfish. CHARLES DERBY and DAVID BLAUSTEIN (Department of Biology, Georgia State University, Atlanta, Georgia 30303)*

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Toward understanding the functional organization of the crustacean olfactory system, we are using intracellular, dye-filled microelectrodes to correlate the structure and function of single, odorant-sensitive interneurons in the brain of crayfish (Procambarus clarkii). We report here on a subset of interneurons that connect the brain proper with the medulla terminalis, a region of the protocerebrum (forebrain) that is located in the eyestalk and which represents a higher-order integrative center. While penetration of the tiny interneurons in the olfactory-globular tract has proven to be extremely difficult, we have successfully studied other larger, odorant-sensitive interneurons that either ascend or descend the eyestalk nerve. Most neurons studied were ascending interneurons, with soma located in the brain proper and axon extending toward the medulla terminalis. Most neurons were also multimodal, responding not just to odorants but also to touch and light. A wide diversity of morphological types of ascending interneurons was found. A neuron might have its soma or major dendritic branches in either the fore-, mid-, or hind-brain. Fewer descending interneurons were found. One type of descending interneuron had its soma in cluster D of the medulla terminalis, its dendrites in the glomeruli centralis of the medulla terminalis, and its axon extending into the brain proper. This study, together with others (Tautz et al., 1986, Naturwiss. 73, 154-156; Arbas et al., 1986, Chem. Senses 11, 578; Blaustein et al., 1986, Chem. Senses 11, 582-583), provides an initial foundation for a functional understanding of the chemospace and the standard of the functional understanding of the chemosensory regions of the crustacean brain.

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Intracellular Recordings of Odor Induced Responses in the Rat Olfactory Bulb. DAVID P. WELLIS and JOHN W. SCOTT (Department of Anatomy and Cell Biology, Emory University School of Medicine Atlanta, GA 30322).

Intracellular recording and marking will be essential for unequivocal identification of the multiplicity of cell types of the olfactory bulb (OB) during studies of odor responses. In addition, intracellular recordings reveal more about the mechanisms shaping these responses (Hamilton and Kauer, Brain Research 338:181, 1985). In spite of this, few intracellular recordings have been used for the characterization of odor responsiveness in mammals.

Intracellular recordings of rat OB cells were made with KCl-Tris-horseradish peroxidase filled microelectrodes, bevelled to resistances of 60-130 MOhms. Cells, seven to date, were held for 15 minutes to over 4 1/2 hours and showed resting membrane potentials of -44mV to -75mV. Odors were presented as in Harrison and Scott (J. Neurophys 56:1571, 1986) following characterization of the cell's antidromic and orthodromic response properties.

All output cells exhibited complex patterns to different odors over a range of concentrations. Odor responses were stable over several hours of recording with repeated odorant concentration series. Spike train matching analysis showed that odor responses could be systematically correlated with odorant concentration differences. One tufted cell evidenced completely different response patterns, for two odorants, being excited by amyl acetate but inhibited by all concentrations of ethyl butyrate. This cell's odor response pattern changed with hyperpolarization, although graded responses over concentration series were still pre-These preliminary data suggest that the amount of inhibitory input may shape a neuron's response to odor; differences in odor responses between cell types may be dictated by differing amounts of granule dendrodendritic inhibition. These studies show that detailed analysis of odor induced responses with intracellular recording can be performed in the mammalian olfactory bulb.

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Evidence of lateral synaptic interactions in olfactory bulb output cell responses to odors.

DONALD A. WILSON and MICHAEL LEON, Department of Psychobiology, University of California, Irvine

Lateral inhibitory circuits are found throughout the nervous system. While the neuroanatomical basis for lateral inhibitory interactions exists in the olfactory bulb of Norway rats, there has been no direct demonstration of lateral inhibition in the responses of olfactory bulb output neurons to odor stimulation. We recorded the extracellular single unit activity of a large number of sequentially recorded mitral/tufted cells in young, urethane anesthetized rats. Responses of these cells to odor stimuli at two different concentrations, as well as the inter-cell distance between them, were recorded. Responses were classified simply as excitatory, suppressive or no response. The probability of recording two cells with excitatory responses to the same odor was then determined for inter-cell distances up to 500um. For cells stimulated with high concentration odors, the probability of two cells 100-200 um apart both being excited by the same odor was significantly lower than that predicted if all cells responded to the odor independently. Cells separated by longer or shorter inter-cell distances did not differ from the predicted value. Responses to the low odor concentration were not dependent on inter-cell distance. These results demonstrate that lateral synaptic interactions within the olfactory bulb influence output cell responses to odor stimulation.

This work was supported by grant BNS 8606786 from NSF to D.A.W. and M.L.

Responses of Mitral/Tufted Cells to Electrical Stimulation in the Olfactory Bulb of the Tiger Salamander. K.A. HAMILTON and J.S. KAUER (Tufts-New England Medical Center, Boston MA)

In order to examine the effect of stimulus intensity on the responses of mitral and/or tufted (M/T) cells in the olfactory bulb of the tiger salamander, we have recorded the responses of 46 cells to electrical stimulation of the olfactory nerve and medial olfactory tract using intracellular electrodes. The responses were assigned to categories that reflected both response pattern (K.A. Hamilton and J.S. Kauer, Brain Research (K.A. Hamilton and J.S. Kauer, Brain Research (I), and the latencies, durations and amplitudes of components of the responses were compared.

The latencies of depolarizing and hyperpolarizing components, and also the duration and peak amplitude of hyperpolarization, differed significantly among responses elicited by low to high I Increases in I, however, did not appear to affect orthodromic and antidromic responses in the same way. For example, in the orthodromic responses, the mean latency, duration and amplitude of hyperpolarization in responses which were elicited by moderate I and contained single, initial spikes differed from the mean values for responses which were elicited by low I and lacked spikes. By contrast, in the antidromic responses, the mean values for responses which were elicited by moderate I and contained spikes did not differ from the mean values for responses which lacked spikes, but did differ from the mean values for responses which lacked spikes, but did differ from the mean values for responses which were elicited by high I and contained single spikes also.

Comparisons such as these provide information about the activation of synaptic connections in the olfactory bulb, which should be useful in evaluating responses to odor stimulation obtained from the same group of cells.

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100 Olfactory Bulb Neurons Develop Rhythmic Activity In Cell Culture. Stephen P. Fracek Jr., Linda E. Czisny and Rollie Schafer (North Texas State University, Denton, TX 76205)

Dissociated olfactory bulb tissue from 14-15 day mouse embyros aare being cultured on multimicroelectrode plates (MMEPs) using the culture system previously used for mouse spinal cord tissue (Gross, G.W. and Lucas, J.H., 1982, J. Electrophysiol. Tech. 9:55-67). Each MMEP consists of a 6X6 array of indium-tin oxide (ITO) electrodes, for a total of 36 recording electrodes, which allow single- and multiunit recording from the culture. Recording from the MEPP takes place in a sterile chamber on a microscope stage and the virtually transparent ITO electrodes allow visual monitoring of the culture during recording. Recording conditions include a constant temperature of 36°C and pH of 7.4. Recordings and optical microscopy done continuously over a 6 hr period show no apparent degradation of the morphological or electrical characteristics of the culture while in the chamber. After approximately two weeks in culture large numbers of neurons develop spontaneous activity, often in the form of bursting spike activity. Strong, rhythmic bursting activity continues in olfactory bulb cultures for two months or longer. Thus the electrophysical activity of cultured olfactory tissue is similar to cultured spinal cord, in that the neurons of both tissues develop coupled, bursting activity in culture. Attempts are now underway to co-culture peripheral olfactory epithelium (containing receptor neurons which detect odorants in the environment) and bulb tissue (containing the central integrative circuits for olfaction). The hope is to create a model neural system in which the physiologically relevant stimuli (odorants) are known.

101 Contrasting Patterns of Serotonergic Innervation of the Main and Accessory Olfactory Bulb In the Adult Rat. M.T. SHIPLEY, J. H. MCLEAN and M. N. LEHMAN (Univ. of Cincinnati).

Serotonergic (5-HT) innervation of cortical structures is a common feature in the CNS. Varying degrees of lamination have been described in neocortex (Kosofsky et al., 1984) and we have reported a particularly striking lamination of the 5-HT input to MOB where serotonergic fibers show strong preferential innervation of the glomerular layer (McLean & Shipley, in press). It was of interest, therefore, to compare this pattern of serotonergic innervation to that in a closely related structure, the accessory olfactory bulb (AOB).

The serotonergic input to the olfactory bulb arises in the dorsal and median raphe nuclei. The innervation of the AOB was accessed by two methods: 1) Sections of the AOB were processed for immunocytochemistry using an antibody specific to 5-HT conjugated to bovine serum albumin to visualize the distribution of serotonergic fibers. (2) The tracer, WGA-HRP, was injected into the dorsal and median raphe and anterograde labelling in the AOB was visualized by TMB histochemistry.

In the MOB, the infraglomerular layers contain moderate innervation by predominantly fine caliber 5-HT fibers. Essentially the same pattern of fine fibers is observed in the infraglomerular layers of AOB. The glomerular layer of MOB is heavily innervated by larger caliber, intensely stained fibers; glomerular fiber densities are 2-3 times greater than in the deeper layers. By contrast, the glomerular layer of the AOB is essentially devoid of 5-HT fibers. The anterograde labelling experiments entirely supported the laminar differences found by immunocytochemical staining.

We have suggested (McLean & Shipley, in press) that the preferential 5-HT innervation of the glomerular layer in MOB provides the chemoanatomical substrate for a potentially powerful rapheserotonergic regulatory action at the first level of synaptic integration in the main olfactory bulb. The absence of this innervation in the AOB suggests that 5-HT actions at the initial level of processing may be less potent in the accessory system. Though frequently cited as anatomically "parallel", MOB and AOB systems may differ significantly in their specific synaptic organization.

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102 Evidence For Olfactory Inputs To The Septum. C. REYHER (University of Berlin, West Germany), J.H. McLEAN and M.T. SHIPLEY (University of Cincinnati)

A unique feature of the olfactory system is its remarkably direct and heavy innervation of telencephalic limbic structures. The main and accessory olfactory bulbs have dense and often reciprocal connections with the hippocampal formation, (entorhinal cortex, hippocampal rudiment) and the amygdala. Direct hypothalamic inputs to the bulb are well established and primary olfactory cortex appears to have reciprocal connections with the hypothalamus. Early neuroanatomists reported olfactory projections to the septal region in several vertebrate species but subsequent tract-tracing experiments have failed to support this view. Here, we report that there is a dense, direct projection to the septal area from a restricted subfield of the anterior olfactory nucleus (AON).

Discrete, iontophoretic injections of WGA-HRP were made in subdivisions of AON. Our injections labeled bulbar and olfactory cortical connections previously described by others. With focal injections in the dorsomedial transition area (AONdm) and dorsal peduncular cortex (DPC) there was dense anterograde labelling in the medial half of the lateral septal nucleus. The labelling directly abutted but did not significantly encroach upon the medial septal nucleus. The projection was confirmed by placing WGA-HRP injections in the septum. A discrete population of neurons were retogradely labelled in AONdm and DPC. The apical dendrites of these neurons are directed towards the molecular layer of AON, a zone which receives a dense terminal projection from the main olfactory bulb.

Classical reports of axons from the olfactory bulb to the septum were based on descriptive methods, and the observations were largely confined to non-mammalian vertebrates. The present findings suggest that an olfacto-septal circuit may exist, but that the projection arises from second order neurons in AON rather than the bulb.

Based on these results, we hypothesize that olfactory bulb mitral/tufted cells may activate a discrete population of dorsomedial AON neurons that project to the septum. This could provide relatively direct olfactory input to septal neurons that regulate hippocampal and hypothalamic circuits. In addition, this MOB-AON circuit may modulate septal neurons known to drive hippocampal rhythm, which is correlated with sniffing.

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The location of neurotransmitters in the olfactory bulb of <u>Xenopus laevis</u> was compared to published reports on the location of the same neurotransmitters in mammals. Tadpoles and young frogs were anesthetized with MS 222 and fixed by immersion or perfusion with 2% paraformaldehyde and 0.15% picric acid or 1% paraformaldehyde and 1.25% glutaraldehyde (GABA, only) in 0.1M phosphate buffer. Cryostat sections (15 um) were processed for immunocytochemistry using immunofluorescence, PAP, or ABC-peroxidase methods. We examined the location of immunoreactive cells and fibers using primary antisera against tyrosine hydroxylase (for catecholamine neurotransmitters), serotonin, and GABA.

Tyrosine hydroxylase-like (TH) immunoreactivity was present within fibers in all layers except the olfactory nerve layer (ONL) and within cell bodies and dendrites located in the glomerular (GL), external plexiform (EPL), and mitral cell (MCL) layers. The largest neurons containing TH immunoreactivity were located in the GL (10um X 10um, mean diameter), while there appeared to be two populations of immunoreactive neurons located in the EPL and MCL (10um X 8.75um and 8.75um X 7.5um, mean diameters). Gamma-amino butyric acid-like (GABA) immunoreactivity was present within neurons of similar size (10um X 7.5um, mean diameters) in the GL and granule cell layer (GRL). Dendrites and axons were difficult to follow away from the cell body, but neuronal processes stained for GABA were present within the glomerular neuropil. Serotonin-like (5-HT) immunoreactivity in the olfactory bulb was only located within nerve fibers. Fine, beaded 5-HT fibers distributed throughout the EPL, MCL, and GRL. Occasional positive fibers projected into the GL, but most fibers stopped at the border between the EPL and GL. Similar patterns of staining were observed in the main and accessory olfactory bulbs for all three antibodies.

In summary, the pattern of immunoreactivities in Xenopus olfactory bulb resembles that of mammals except there are more neurons with TH staining in the EPL and MCL and fewer fibers with 5-HT staining in the GL.

Supported by BRSG funds (#RRO5675) to GDB.

Neurochemical Analysis of the Salamander Olfactory
Bulb. K.A. HAMILTON, R.H. KRBAM, J.S. KAUER. (Tufts-New
England Medical Center, Boston. MA)

The tiger salamander (<u>Ambystoma tigrinum</u>) has been shown to be a useful model system in which to study olfactory function because its anatomy affords advantages for physiological recording and because its ability to discriminate odors can be tested using behavioral methods. Here we report a preliminary analysis of neurochemical properties of the salamander brain, including the olfactory bulbs, using high pressure liquid chromatography with electrochemical detection (HPLC-EC) and immunohistochemistry.

Using HPLC-EC, we have assayed for norepinephrine(NE), dopamine (DA), serotonin (5HT), and epinephrine(EPI) in the olfactory bulbs, telencephalon, and midbrain of 13 animals. The findings are summarized in the following table (values in pg/mg tissue +SEM; number of samples=()):

 NE
 DA
 5HT
 EPI

 olfact.bulb
 65+12(5)
 .131+25(8)
 399+43(6)
 not det.(9)

 telencephalon
 151+15(6)
 170+40(6)
 1383+236(7)
 120+66(6)

 midbrain
 264+24(7)
 494+45(7)
 2090+370(6)
 429+48(5)

The ratios of concentrations among these compounds are similar to those found in the hamster brain.

Using immunohistochemical methods, we have examined the distribution of tyrosine hydroxylase (TH), 5HT, and gammamino butyric acid (GABA). TH-like immunoreactivity was found in some cells in the granule cell layer (GCL) which had dendrites extending into the external plexiform layer (EPL), and in fibers and a few cell bodies in the periglomerular (PG) region. 5HT-like immunoreactivity was found in fibers in the EPL. GABA-like immunoreactivity was found in the GCL, EPL and PG regions, most intensely in PG cell bodies at the olfactory nerve-glomerular border.

These results will be discussed in relation to the

These results will be discussed in relation to the concentrations and distributions of these compounds in the mammal and in relation to intracellular recordings we have obtained from mitral/tufted cells in response to odors.

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Fine structure of the olfactory (aesthetasc) sensilla
of the spiny lobster. ULRIKE GRÜNERT and BARRY W. ACHE (C.
V. Whitney Laboratory, University of Florida, St. Augustine,
FL 32086)*

The first antennae or antennules of the spiny lobster bear tufts of hair sensilla called aesthetascs. These sensilla are thought to be responsible for the "olfactory" sensitivity of the animal (Ache and Derby, TINS 8:356-60, 1985). The aesthetascs are about 20 μm in diameter and 0.8 mm long. The ends of the hairs taper to fine tips, which do not show tip pores. The cuticle of the aesthetascs varies in thickness and consistency over the length of the hair. At the base of the hair it is considerably thicker than in the distal region, where it has a spongy appearance. This region is thought to be the place where chemical stimuli enter the hair. Each aesthetasc is innervated by 318 (x, range 270-370) bipolar neurons, the dendrites of which project as a bundle into the hair shaft. Each dendrite develops two cilia. Within a very short distance each of these cilia branches repetitively and dichotomously, resulting in 8,000 to 10,000 outer dendritic segments per hair, or about 20 outer dendritic segments per neuron. The branches intertwine frequently before running parallel to the tip of the hair. Each hair also possesses inner and outer non-neural (auxiliary) cells. The inner auxiliary cells surround the bundle of dendrites, extending distally to the origin of the ciliary segments. Extensions of these cells project into the bundle of dendrites, separating groups of dendrites into discrete clusters. Outer auxiliary cells wrap the inner ones, but do not extend beyond the base of the hair. This morphological study provides a basis for ongoing biophysical and biochemical investigations of early events in olfaction, the former of which should be particularly interesting since these are among the longest dendritic processes reported for chemoreceptors.

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106 Neighborliness of Cobalt-Filled Frog Olfactory Axons from Epithelium to Olfactory Bulb. M.M. DASTON and G.D. ADAMEK (Dept. Anat. & Cell Biol., Univ. of Cincinnati)

We do not know the relative relationships of groups of olfactory axons from a contiguous origin in the olfactory epithelium to their termination in the olfactory bulb. In particular, we do not know the relative postions of such axons a) as they leave the epithelium, b) within the nerve, or c) at their terminations within the glomeruli of the olfactory bulb. Previous studies from other laboratories only followed portions of the courses of axons. In a previous study using HRP injections in frog olfactory epithelium, we showed that the axons of cells that are neighbors in the epithelium are widely separated within the olfactory nerve. The limitation of HRP in this preparation is that it is transported in discrete particles. It was therefore impossible to trace the complete paths of individual axons to determine how and where they diverged. We have developed a method using cobalt filled cells which allows us to visualize the course of axons from the epithelium to the olfactory bulb within one preparation. Pithed frogs were kept at 4°C throughout the injection. A pipette with a tip diameter of 20µ, filled with 0.6 M cobalt chloride, was placed in the exposed ventral olfactory epithelium and left in place for 24-72 hours. Passive diffusion is sufficient to allow movement of cobalt from the pipette into the tissue. Following the injection, the epithelium, nerve and bulb were removed in one piece and placed in a weak solution of ammonium sulfide for about 20 minutes. This resulted in a black precipitate within the cobalt-filled cells. The injection site was always immediately visible by eyc. Approximately 2000 cells contained precipitate. The tissue was then prepared for light and electron microscopy. With this new application, the relative positions of the axons of a group of cells as they leave the epithelium and enter the olfactory nerve are readily visible with the light microscope. Axons diverge immediately below the basal lamina before they enter the nerve. Within the nerve, axons maintain the spatial relationship established at the beginning. They diverge again at the olfactory bulb. We will place cobalt in various regions of the olfactory epithelium to look for differences in the pattern of projection to the olfactory bulb. The physiological implications of these results are discussed in another abstract.

Supported by NIH NS23523, NS23348 and NSF BNS854025.

O7 Spatial Organization of Receptor Surfaces and Odorant Passageways in the Hamater Nasal Cavity: Morphometric Analyses Based on a Stereotaxic Atlas. ANDREW N. CLANCY, THOMAS A. SCHOENFELD and FOTEOS MACRIDES (Worcester Foundation for Experimental Biology, Shrewsbury, MA 01545)

In macrosmatic mammals, potential studies of the movement of odorant molecules and their spatial and temporal patterns of interaction with mucosal surfaces during inspiration are hampered by our lack of understanding of the spatial organization of the nasal cavity. We approached this problem by creating a stereotaxic atlas of the hamster masal cavity and applying morphometric analyses of the cavity spaces and different types of epithelial surfaces. The organization of nasal spaces and surfaces could then be related to the organization of sensory input to the olfactory bulbs. Also, convolution was measured in the coronal plane throughout the rostro-caudal extent of the cavity. This measure reflects the surface-to-volume ratio and is related to chromatographic resolution along the longitudinal axis. Several findings are salient. The circumferential extent of olfactory epithelium (OE) increases with the convolutions of the olfactory turbinates and peaks just caudal to the septal window. through which negative pressure is applied during inspiration. The maximum circumferential extent of the OE (39 mm) is much greater than its rostro-caudal extent (11 mm), and 75% of its surface area is distributed over a rostro-caudal extent of only 3.6 mm centered at this caudal peak. Spatial resolution of rhinotopically organized central projections is high at this level (of. Schoenfeld et al., this meeting). The respiratory turbinates produce a rostral region of high convolution, organized as longitudinal channels of theoretically high chromatographic resolution. These channels all lead into a region of minimum convolution in the center of the cavity, rostral to the septal window. The septal organ and rostral extensions of the OE are located in this region and collectively provide rhinotopic input to all circumferential quadrants of the main olfactory bulb. These sectors of the sensory epithelium may be optimally positioned for monitoring sorptive effects on odorant distribution.

Supported by NSF grant BNS85-10743.

NOTES

Is the Northern Fulmar's Large Olfactory Bulb Designed for High Sensitivity in Odor Detection?

E. MEISAMI (Physiology Dept. Univ. Illinois, Urbana, IL 61801); B. WENZEL (Physiology Dept. Univ. California, Los Angeles, CA 90024)

recently shown that the olfactory bulbs have (OB) of the Northern Fulmar (a procellariiform bird) is very large, its size and the total number of mitral cells (M-cells) exceeding those in the macrosmatic mammals (1). Further morphometric and quantitative studies of other layers in the Fulmar OB indicate a contrasting though functionally interesting picture: A) The external and internal plexiform layers, associated with the M-cells' and recurrent axon-collaterals, respectively, are thick and wide (well-developed). B) The internal granular layer is massive but its tens of millions of granule cells are loosely organized. unlike the crescent islets formations, seen in the macrosmatic mammals. C) The total number of glomeruli is very large (>30,000) but the glomerular layer is poorly differentiated, the periglomerular and the external tufted cells being clearly sparce. If in the macrosmatic mammals, the highly organized glomerular layer's architecture and the abundant periglomerular and tufted cells these animals in their great capacity for discriminatation at the OB level. therefore suggest that the structural features of the Fulmar OB may represent a neural system which while not suited for optimal odor discrimination, is well adapted for high sensitivity in odor detection. This interpretation is supported by the massive size of the olfactory mucosa (an index of olfactory sensitivity) in this bird (2), and by the field studies on the Fulmar's olfactory guided foraging behavior (3). REFS: 1. Wenzel & Meisami ('87) Ann NY Acad Sci (in press); 2. Bang ('71) Acta Anat 79 (sup):1-76; 3. Hutchison & Wenzel ('80) Condor:82:314-319

109 Neuron Number and Topography in the Human Olfactory Nucleus.

Kunwar P. Bhatnagar (Anatomy Dept, Univ. Louisville, Louisville, KY 40292); Esmail Meisami (Physiology Dept, Univ. Illinois, Urbana, IL 61801)

neurons of the anterior olfactory nucleus (AON) participate in feedback interactions with the olfactory bulb's (OB) relay cells (mitral & tufted). In humans, the (AON) lies in part within the OB's core (stratum album), providing a unique opportunity for the numerical analysis of such feedback interactions. Thus a knowledge of AON topography and mitral: AON cell ratio will be very valuable. Bhatnagar et al (Anat Rec., 218, 1987) estimate the number of mitral cells in the adult human OB to be about 51,000 at age 25. Here we report the topography and number of the AON cells in the adult human female OBs. We found that the AON often comprises three separate islands set apart by 1-3 mm along the OB's length. Each island is 1-2 mm long and consists of clusters of similarly shaped neurons that form an incomplete ring in the OB's core. All large neurons showing a distinct nucleolus were counted in 10um Nissl sections (frontal serial) at 50um intervals. Counts varied between 2-3 thousand cells per island, totalling to a mean of about 8,000 per OB. The 3 islands correspond to the most rostral parts of the mammalian AON (other divisions of the human AON are located in the brain hemisphere). These findings imply: 1) the intrabulbar AON neurons are likely involved mainly in mitral-AON feedback interactions; each island probabely interacts with a separate group of OB relay cells along the OB length. 2) the AON-mitral cell ratio in the human OB is about 1:6. In macrosmatic mammals with large AON, this ratio must be much higher. A correlation between this ratio and the olfactory abilities (discrimination/sensitivity) in various animals may prove interesting.

Adaptation and Mixture Component Suppression in Olfaction.

R.C. GESTELAND and G.D. ADAMEK (Osphresiopolis, Dept. Anat. & Cell Biol., Univ. of Cincinnati)

The perceived intensity of an odor is reduced when it is presented paired with another odor at intensities well above threshold. A maximum number of action potentials is evoked from olfactory receptor neurons by a particular concentration of an odorous stimulus. Lower and higher concentrations evoke fewer action potentials. At low concentrations, repeated presentations of an odor evoke about the same response. A few presentations at high concentrations often lead to adaptation lasting minutes to hours. Different neurons have different thresholds and maximal response concentrations. Thus, an increase in stimulus concentration usually results in an increase in the number of cells which respond. Some stimuli affect only a small fraction of the cell population. Others, e. g., amyl acetate, are capable of activating most neurons. Stimulation with these odors reduces the amplitude of the compound action potential.

In the nerve olfactory axons are closely apposed within Schwann cell invaginations. There is little Schwann cell cytoplasm and Extracellular potassium concentrations are extracellular space. elevated by spike activity in proportion to the spike rate and number of active fibers. Intracellular calcium concentration may also increase. The depolarization caused by changes in membrane ion gradients causes increased axon sodium inactivation. Axon excitability is not restored until appropriate concentrations return. This occurs slowly due to the limited Schwann cell cytoplasmic volume and scarce mitochondria. For most effective transmission of stimulus information, axons from cells which respond to the same stimuli should be spatially separated in the nerve so that their axons are not mutually inhibited by ionic fluxes of neighbors. Either neuron somata of cells with similar odorant sensitivities are not spatially close or the axons of neighboring cells must diverge at the basement membrane so that they are not neighbors in the olfactory nerve. Spike generation may also be affected by inactivation processes. Neuron somata are densely packed with with very thin intervening supporting cell processes, implying low ion homeostatic capacity. Receptor currents lead to potassium efflux which, if not rapidly offset, will raise the threshold for spike generation.

Sodium inactivation in olfactory nerve axons appears to provide a parsimonious explanation for several curious experimental observations related to perceptual and electrophysiological adaptation. Supported by NSF BNS854025 and NIH NS23523 and NS23348.

111 Cross Adaptation of Primary Chemosensory Neurons.
PAOLA F. BORRONI and JELLE ATEMA. (Boston University Marine Program, Marine Biological Laboratory, Woods Hole, MA).

In the lobster, Homarus americanus, olfaction and taste are based on narrowly tuned primary neurons. The response of these receptors to their best compound can be altered by simultaneous exposure to mixtures of other stimulatory and non-stimulatory compounds, resulting in mixture suppression and enhancement (Johnson et al. 1985). In this study we describe the effects of cross-adaptation of lobster NH_4 receptors by single compounds: Glu, Bet and Hyp. Stimulusresponse curves of 40 NH4 receptors were obtained from extracellular recordings. Stimuli (.003, .03,.3,3, and 30mM NH4Cl, and .3mM Glu, Bet and Hyp) were injected into constantly flowing artificial seawater (ASW) or background solutions of cross-adapting compounds in ASW (.003 and .3mM Glu or Bet). Glu and Bet (and to a lesser extent Hyp) are the most common and most effective second-best compounds for NH4 receptors. NH4 receptors with Glu or Bet as secondbest compounds were tested in a Glu or Bet background respectively; cells with no second-best compound were tested in either background, randomly. In the Glu second-best cells, the mean responses to Glu, Bet and Hyp were 75%, 19% and 15%of the response to equimolar NH4Cl, respectively. These cells are not very narrowly tuned (H=.651) and their secondbest compound is an effective stimulus; their responses to NH4 are reduced equally (~31%) by cross-adaptation in both Glu backgrounds. In the Bet second-best cells, the mean responses to Bet, Hyp and Glu were only 8.7%, 2.7% and .7% of the response to equimolar NH4Cl, respectively. These cells are very narrowly tuned (H=.277) and their second-best compound is not a very effective stimulus; their responses to NH4 are not affected by cross-adaptation with a .003mM Bet background, but they are reduced (~37%) by a .3mM Bet background. The response of NH, receptors with no second best compounds was not affected by cross-adaptation with either background at either concentration. These results suggest that the effects of cross adaptation on lobster NH4 receptors depend on the tuning breadth of the receptors and on the stimulatory efficacy of the cross-adapting compound.

Supported by the Whitehall Foundation and NSF (BNS 8512585).

Inosine-5'-monophosphate and inosine enhance some sweet tastes. SUSAN S. SCHIFFMAN, CAMILLA GRAHAM, and ZOE WARWICK (Department of Psychiatry, Duke Medical Center, Durham, N.C. 27710)

5' ribonucleotides have a distinct taste that is called umami in Japanese (see Umami: A Basic Taste, Y. Kawamura & M. R. Kare, eds., Marcel Dekker, Inc., New York, 1987). Those nucleotides first recognized to have an umami taste were those having a purine nucleus with a hydroxy group in the 6- position and a ribose moiety esterified in the 5'position with phosphoric acid; an example is inosine-5'monophosphate (IMP). In addition to the unique taste
quality, mixtures of 5' ribonucleotides and monosodium glutamate have been found to be synergistic when mixed together. In this study, the perceived intensity of a range of tastes was examined after the tongue was preadapted to 10⁻³M IMP for 4 minutes. Statistically significant enhancement was found for two sweeteners: sucrose (.2M) and aspartame (.002M). No statistically significant increases were found for higher concentrations of sucrose or aspartame. No enhancement for the sweeteners sodium saccharin, calcium cyclamate, fructose, stevioside, glucose, or neohesperidin dihydrochalcone were found at any concentrations. No increases with IMP were found for NaCl, HCl, KCl, CaCl₂, citric acid, or quinine HCl. Adaptation of the tongue to 10⁻³M inosine was also found to enhance low concentrations of sucrose and aspartame.

NOTES

 $\frac{\text{An Analogy to the Release}}{\text{(S. C. Johnson }\epsilon} \frac{\text{Cocurs}}{\text{Son, Inc.)}} \frac{\text{In Odor}}{\text{In Color}} \frac{\text{From Mixture Suppression}}{\text{Mixtures.}} \frac{\text{HARRY T. LAWLESS}}{\text{HARRY T. LAWLESS}}$

In perceptually analyzable two-component odor mixtures, the perceived intensity of each component is reduced, relative to its intensity in equally concentrated unmixed stimuli. This is an example of odor counteraction or masking. When the intensity of one component is reduced through adaptation, several lines of evidence would predict that the other component should be released from masking, and increase in perceived intensity. Investigation of two-component mixtures of vanillin and cinnamaldehyde under various conditions of adaptation showed such a "release from odor masking" effect. After adaptation to vanillin, the cinnamon component of the mixture increased in odor strength relative to its intensity in the mixture (after adaptation to solvent). A similar increase was observed for vanillin in the mixture, after adaptation to cinnamaldehyde. This effect is consistent with a central mechanism for odor masking or counteraction, as suggested by previous studies of masking in dichorhinic odor mixtures.

1 1 4 Experience Fecilitates Olfactory Discrimination and Mixture Component Analysis by Humans Michael D. Rabin (Connecticut Chemosensory Clinical Research Center, University of Connecticut Health Center, Farmington, CT 06032).

An implicit assumption of psychophysical testing is that basic sensory measures such as detection or discrimination are free of the contaminating influence of higher order perceptual variables. A common assumption in olfaction has been that these psychophysical measures reflect the inherent operating characteristics of the lowest levels of the olfactory system. However, these assumptions appear to contradict phenomena which imply a more complex perceptual analysis. For example, how is it that a flavor panel can eventually come to discriminate between extremely fine nuances in product composition? Or, what endows a perfumer with the ability to treat a mixture analytically, whereas a layperson may perceive the same mixture holistically? Standard discrimination and mixture perception procedures have ignored cognitive variables that may influence performance. Two experiments were performed to investigate the role of learning in olfactory discrimination. Experiment 1 investigated the effect of training on subsequent discrimination performance. The purpose of this experiment was to discover whether training participants to label simple test adorants prior to discrimination testing would improve subsequent performance. Training participants to label the target odorants did improve discrimination performance relative to other control groups. The results also suggested that label training is a more accurate method for odor training than adjective list profiling. Experiment 2 was designed to relate what odor knowledge a perticipant brought into the experiment to eventual performance on a discrimination involving mixtures. Six targets were selected for each individual participant to represent three familiar and three unfamiliar odors. In this discrimination each target odorant was paired either with itself or with a transformation of itself. Transformations were mixtures consisting of target odorant plus a contaminant varying along one of two dimensions: familiarity or pleasantness. Both target familiarity and contaminant familiarity or pleasantness influenced the ability to discriminate between an odorant and its transformations. Familiar targets and familiar or unpleasant contaminants made discrimination easier. These data support the idea that olfactory discrimination and mixture perception can be mediated by cognitive variables that standard psychophysical assessment of olfactory ability ignore. Individual differences owing to differing olfactory experience may account for this.

Variation in Odor Detection Thresholds Determined by Charm Analysis. A. B. MARIN, T. E. ACREE & J. BARNARD (Cornell University, Geneva)

Variation in odor detection threshold for four groups of individuals cross-classified by sex and age, was determined using six standard aroma compounds: ethyl butyrate, ethyl-2methyl butanoate, ethyl hexanoate, 1,8-cineole, l-menthol, and l-carvone. These compounds were selected for study because they are all naturally occurring food constituents, and specific anosmia has been reported for some of them (Amoore, Chemical Senses & Flavour, 4, 153-161, 1979). Also, specific and general loss of odor sensitivity with age has been reported (Murphy, J. Gerontology, 38, 217-222, 1983; Stevens & Cain, Chemi-cal Senses, 10, 517-529, 1985). In this study, odor detection thresholds were determined using charm analysis.

The charm technique presents pure aroma compounds in a stream of cool, humidified air for subjects to smell. During a stream of cool, humidified air for subjects to smell. During the procedure, the subject sits at a video terminal and records the instant an odor is detected, the quality of the odor perceived, and the instant the smell disappears. Repeated analyses at several serial dilutions of a stimulus yield a measure of odor intensity called charm. Some details are given in Acree et al. (AChemS 9, 1987).

Results of this experiment show that charm analysis is a reliable and reproducible method of determining gas-phase odor detection thresholds; variability of replicated measurements within individual subjects was consistently less than variability between subjects. A significant age effect was

than variability between subjects. A significant age effect was found for detection thresholds of three of the aroma compounds, but there was no sex effect for any. A positive interaction of age and Kóvats retention index of the aroma standards was observed; differences in odor sensitivity between older and younger subjects increased at higher retention indices.

Adaptation Characteristics of NST Taste
Neuron Responses to Salts in Fetal and Postnatal
Sheep. M. B. VOGT and C. M. MISTRETTA (Dept. Oral
Biol., School Dent., and Center for Nursing
Research, Univ. Michigan, Ann Arbor, MI 48109)

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We have described developmental differences in salt responses and lingual receptive field sizes for taste neurons in the nucleus of the solitary tract (NST) of fetal and postnatal sheep. We now report differences in adaptation characteristics for NST neurons in fetuses and lambs, and for neurons that respond with highest frequency to NaCl versus NH₄Cl. Responses to 0.5M NH₄Cl, NaCl, and KCl were recorded from 14 neurons in fetal sheep about 130 days gestation (term=147 days) and 30 in lambs 40-50 days postnatal. Adaptation of the neural response was quantified by calculating the linear slope of response frequency (impulses/sec) over time for the 2nd through 7th seconds of the response. In lambs, rrough /th seconds or the response. In lamos, 'NaCl-best' neurons produced responses with positive slopes during stimulation with NH₄Cl or KCl [mean (SEM): NH₄Cl, 1.09 (0.32); KCl, 0.44 (0.24)], whereas 'NH₄Cl-best' neurons produced responses with negative slopes [NH₄Cl, -0.82 (0.29); KCl, -0.95 (0.44)]. For both 'NaCl-best' and 'NH₄Cl-best' neurons, the slope of the response to NaCl was flatter and opposite in sign to that produced by the other salts ['NaCl-best' to that produced by the other salts ['NaCl-best' -0.29 (0.32); 'NH₄Cl-best', fetuses, both 'NaCl-best' (0.31)]. In 'NH4C1-best' 0.22 fetuses, and neurons produced negative response slopes for each salt and fetal slopes declined more steeply than negative response slopes in lambs. This presumably reflects the general inability of neurons to produce a sustained . Our studies of the NST demonstrate differences in neuron type and development for response frequencies, adaptation characteristics, and receptive field sizes. (Supported by N.S.F. Grant BNS 8311497.)

Environmental Effects Upon the Developing Gustatory 117 System: Towards a "Sensitive Period". P.R. PRZEKOP JR. & D.L. HILL (University of Virginia)

The development of taste in rat is a postnatal phenomenon in which sodium, initially a poor stimulus, becomes very effective. Development appears to involve the addition of an amiloride-sensitive sodium component to the receptor membrane which is absent in the immature rat. During development, peripheral taste responses are susceptible to environmental manipulation. By instituting a sodium restricted diet (.03%) at 3 days post-conception, chorda tympani responses to sodium stimuli are lower than normal. In adult rats sodium deprivation has no effect upon multifiber chorda tympani responses. Therefore, a specific time period exists in which the taste system can be altered. The present study examines this time period by instituting sodium deprivation during different periods of both pre- and postnatal development and comparing multifiber chorda tympani responses. Concentration series (0.05-0.5M) of NaCl, NH4Cl, KCl, and sodium acetate were flowed over the anterior tongue. Results suggest that the degree of response attenuation is determined by the time at which sodium deprivation is imposed upon the animal. Therefore deprivation is not an all or none phenomenon in altering sodium responses. Rather, there appears to exist a continuum with given endpoints corresponding to periods of development when deprivation has maximal through no effect. Thus, the taste system remains plastic for an extended period of early development; however, the degree of plasticity changes during this period. Although these effects are reversible at the peripheral level, they may not be reversible in the central nervous system.

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Smelling through the Nasopharynx: Effect of Aging. JOSEPH C. STEVENS and WILLIAM S. CAIN (John B. Pierce Foundation Laboratory and Yale University).

Groups of 20 elderly subjects (67-83 yrs) and 20 young subjects (18-24) made magnitude estimations of NaCl and ethyl butyrate solutions, sampled orally, first with the nose unpinched, then with the nose pinched shut to prevent the airborne (volatile) molecules from reaching the olfactory receptors through the nasopharynx. In the concentrations studied, ethyl butyrate had weak taste and strong odor. On the basis of earlier work, which has shown widespread weakening of perceived intensity among elderly persons, it was hypothesized that pinching the nose would depress the perceived intensity of ethyl butyrate more in the young than in the elderly subjects. In order to discount individual propensities to use large or small numbers, the NaCl estimations were used to normalize the ethyl butyrate estimations. It then became clear that the average young person perceived the overall intensity of ethyl butyrate solutions to be far stronger with the nose unpinched than pinched. In contrast, for the elderly it made little difference whether the nose was pinched or unpinched. Even the unnormalized estimates of ethyl butyrate prove essentially the same point: at each concentration the difference between the two magnitude estimates for the pinched and unpinched conditions was on the average much larger for the young than for the elderly. Also the young (but not the elderly) typically expressed surprise at how much larger were the overall intensities of ethyl butyrate seemed when the nose was unpinched. Olfactory losses inferred in this study are of comparable magnitude to those measured in earlier studies, in which the odorants were sniffed via the nostrils.

This research was supported by NIH Grant No. AG04287. The authors thank Amy Pauw for extensive technical assistance.

119 Olfactory Nerve Transection In The Aging Hamster: Neurogenesis and Axon Reconnection Of The Olfactory Bulb.
RICHARD M. COSTANZO and EDWARD E. MORRISON (Medical College of Virginia, Richmond, VA 23298).

The aging mammalian brain has been shown to possess unsuspected neural plasticity. Recently, we have demonstrated that the olfactory neuroepithelium in old adult hamsters (>1 yr.) maintains the capacity for continued neurogenesis. Following nerve transection degenerating receptors are replaced by new cells that develop into mature olfactory neurons. In the present study, we examined the growth and development of newly formed olfactory axons and their capacity to reestablish new connections with the olfactory bulb in the aged hamster. Unilateral nerve transection was performed on 30 hamsters and morphological observatons were made at 0-120days recovery. Following nerve transection there was a retrograde degeneration of olfactory neurons. At day 4 degenerating axon fascicles were observed within the lamina propria and olfactory bulb. At 15 days we observed axons from developing neurons entering the lamina propria and growing centrally toward the olfactory bulb. By 60-120 days recovery the number of mature olfactory neurons had increased significantly and mitotic figures could still be observed adjacent to the basal lamina. Axon fascicles reached the olfactory bulb and established normal and ectopic glomeruli. These data demonstrate for the first time, that the capacity for neurogenesis and the ability to reestablish axonal connection with the olfactory bulb persist in the old adult animal. Such findings may have important implications for new areas of research involving repair and replacement of damaged pathways in the adult central nervous system.

Supported by NIH Grant 16741 to RMC and $\,$ VCU Grants-In-Aid to EEM.

NOTES

120 Hedonic Reactions to Odors in Three-Year-Old Children.
HILARY J. SCHMIDT and GARY K. BEAUCHAMP (Monell Chemical Senses Center)

Previous studies of young children have suggested that adult-like hedonic responses to odors are not evident until between 4 and 7 years of age. The present experiment used a new method to investigate 3-year-olds' (n = 19) hedonic reactions to 9 odorants, and to contrast their responses to those of adults (n = 17). In a forcedchoice categorization procedure presented as a game, subjects gave smells that they liked to one puppet, and smells that they didn't like to another puppet. A pretest evaluated children's comprehension of this task, and those who failed were excluded from the analyses. Odorants were equated for perceived intensity and included: C-16aldehyde, amyl acetate, eugenol, phenyl-ethyl-methyl-ethylcarbinol, methyl salicilate, 1-carvone, butyric acid, pyridine and androstenone. In addition, 4 blanks (mineral oil, the diluent for the odorant stimuli) were included in the stimulus set. Analyses of the proportions of subjects in each age group who rated each odorant as pleasant revealed: 1) systematic and essentially similar preferences and aversions in children and adults (log-linear Pearson chi square = 111.1, p < .01); 2) a high degree of overall agreement amongst children and adults on the rank ordering of stimulus desirability (Kendall's tau = .53, p < .02); and 3) that children and adults differed on their ratings of some odorants (log-linear Pearson chi square = 26.7, p < .01). Post-hoc analyses showed that children were less likely than adults to rate androstenone as pleasant (12% vs. 42%, chi square = 4.4, p < .05), but were more likely than adults to rate pyridine as pleasant (29% vs. 0%, chi square = 5.6, p < .05). Given that approximately 50% of adults are anosmic to androstenone, this pattern may indicate that a greater proportion of children than adults are sensitive to this odorant. It further suggests that not all odor aversions in children can be attributed to trigeminal stimulation.

In sum, it appears that young children exhibit odor likes and dislikes that are much like adults' preferences. Negative findings in earlier research may have been attributable to the selection of stimuli, and/or to methods which may have been inappropriate to the cognitive and behavioral limitations of the young child.

Supported in part by NIH NS22014.

121 NE modulation of one-trial olfactory conditioning and olfactory bulb neural responding to an attractive odor.
REGINA SULLIVAN and MICHAEL LEON (University of California - Irvine)

In Experiment 1, 6 day old pups were trained in a one-trial olfactory classical conditioning experiment. Experimental pups were conditioned to prefer peppermint odor by pairing that odor with stroking (to mimic reinforcing maternal stimulation). Control pups received only the odor, only the stroking, or neither stimuli. The next day, pups were given a two-odor choice test or 2-DG with exposure to only the odor. Pups that received simultaneous odor and stimulation were the only pups which exhibited an attraction to the conditioned odor. Furthermore, such pups had greater focal 2-DG uptake in specific olfactory bulb glomeruli than pups in all other treatment groups. These results suggest that a single, brief conditioning trial in young rats modifies both behavioral and neural responses to subsequent presentations of that stimulus. The mechanism underlying the neurobehavioral response to one-trial learning on day 6 differs, however, from that underlying olfactory preference training from days 1-18.

We the found that norepinephrine (NE) is involved in the development of the neural and behavioral response to olfactory preference training on day 6. Odor paired with either stroking or an NE agonist (isoproterenol) was sufficient to produce both an odor preference and enhanced uptake of 2-DG. Furthermore, an NE antagonist (propranolol) blocked both the behavioral preference and enhanced 2-DG uptake which followed odor preference training. These results suggest that NE is involved in modulating the conditioned behavioral and neural responses to attractive odors.

Supported by HD06818 from NICHD to RS and NS21484 from NINCDS and MH00371 from NIMH to ML.

Relation of MHC-Related Mating Preferences to Postnatal Chemosensory Imprinting. KUNIO YAMAZAKI, GARY K.
BEAUCHAMP, DONNA KUPNIEWSKI, CATHI STAHLBAUM (Monell Chemical Senses Center), JUDY BARD, LEWIS THOMAS and EDWARD A. BOYSE (Memorial Sloan-Kettering Cancer Center)

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Previous studies of mating preferences of inbred strains of mice differing only at the major histocompatibility complex (MHC) of genes, a set of genes which are known to confer olfactory individuality as well as to control important aspects of immune function, often demonstrated a tendency for matings between males and females of different MHC types. This mating bias would tend to favor heterozygosity at MHC loci which could have important selective advantages. Other earlier data suggested that some form of imprinting may influence mating preferences according to MHC type. In light of the importance of linking imprinting with genetically-determined chemosensory individuality, we report here an investigation of mating preference according to MHC type in males and females reared in association with foster parents of differing MHC types. Pairs of F_3 homozygous mouse panels obtained from typed F_2 segregants (C57BL/6 crossed with the congenic strain C57BL/6-H-2 $^{\rm K}$) were established. Entire litters were fostered on postnatal Day 0 (at birth) to mothers and fathers which were either of the same MHC type as the biological parents or to the other MHC type. Litters were reared until weaning in the company of both parents. At weaning, usually at 21 days of age, each litter was segregated according to sex. In mating tests with fostered (test) males, a pair of normally-reared MHC-congenic females, both in estrus, was introduced into the cage of an isolated adult test male. The trio was then observed until copulation occurred with one of the two females. In tests with fostered (test) females, the test female, in estrus, was introduced to a pair of normally-reared MHC congenic males in the test cage. The results with males implicated early parental exposure as a factor modulating expression of mating preferences based on MHC type. In contrast, tests with females revealed (a) a mating preference based on MHC type, but (b) no effects of differential fostering. Supported by NIH 9 RO1-GMCA-32096.

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Relative Contribution of Different Genetic Regions to
Urinary Odors Distinguishing Inbred Strains of Mice. HEATHER J.
DUNCAN, GARY K. BEAUCHAMP and KUNIO YAMAZAKI (Monell Chemical Senses Center)

That individuals possess unique odor profiles has been documented in several species. It is also known that genetic relatedness (and hence, similarity) can be conveyed by olfactory cues. However, no specific genetic locus has been identified as controlling individual odor differences. Studies with inbred strains of mice have identified the major histocompatibility complex (MHC), a linked set of genes known to be involved in control of immune function, as one area of the genome which contributes to strain differences in odor. Mice which carry different alleles in this area, but which are genetically identical elsewhere, have distinctive odors, identifiable in the urine by other mice, rats or humans. As one approach to determining the relative importance of the MHC to the urine odor "profile", compared with the combined effects of a multitude of other genetic differences (background) which differentiate mouse strains, four rats were trained to discriminate between the urine odors of two strains of mice, differing both at the MHC and other areas of the genome. During generalization testing, subjects were presented with various mixtures of urine from the training (S+) strain which had been adulterated with urine from mice identical at the MHC but with a different background, or urine from mice with the same background but with different MHC alleles. As the percentage of either adulterant increased, the tendency for the rat to respond to the urine sample decreased. For three of the rats, the decrease in responding was greater when the adulterant came from mice with different backgrounds. These results suggest that urinary odorants are determined by both the MHC genes and a multitude of other genes located throughout the genome, and the combined effects of the non-MHC genes may make a larger contribution to strainspecific odors. In addition to the particular issue of genetic control of olfactory individuality in mice, this novel dilution technique may prove useful for studying other questions about perception of odor mixtures by rats.

124 The Influence of Olfactory Bulbectomy on the Mate Choice of Female Mice. CATHI C. STAHLBAUM (Monell Chemical Senses Center and Cornell University)

The major histocompatibility complex (MHC) of the mouse has previously been demonstrated to be involved in mate preference. In general, experiments have shown that matings between males and females of different MHC types (congenic) are more frequent than those between similar MHC types (syngenic). Because it is not clear whether this bias is due to male or female mate preference alone or to an interaction of the two the study reported here investigates female mate preference between tethered MHC congenic males (B6 and B6-H-2^K). Sexually inexperienced estrus females of these (). Sexually inexperienced estrus females of these congenic strains were permitted access to a pair of males tethered at either end of a test arena designed to allow the female the choice of interaction with each male individually or to remain out of contact with both males. Interactions were videotaped for at least 12 hours after copulation and subsequently evaluated. B6 females preferred to mate with syngenic B6 males rather than congenic B6-H-2^k males. On the other hand, B6-H-2^k females expressed no preference for either type of male. In order to determine if the mate preference of the B6 females was based upon olfactory cues each of four groups of females were subjected to one of the following treatments before being tested in the above manner: (1) bilateral olfactory bulbectomy (BBX), (2) unilateral olfactory bulbectomy, (3) sham olfactory bulbectomy (SBX) or (4) no surgery (C). Unlike the other groups, whose mating preferences were unaffected by the treatments, the BBX females failed to mate at all in the test situation. However, these same BBX females did subsequently mate with untethered males in their home cages. These results suggest that BBX females exhibit a general decrease in sexual receptivity.

25 Conditioned Discrimination of an Airborne Odorant by Garter Snakes, Thamnophis sirtalis sirtalis. DAVID BEGUN, MIMI HALPERN (SUNY Health Science Center at Brooklyn).

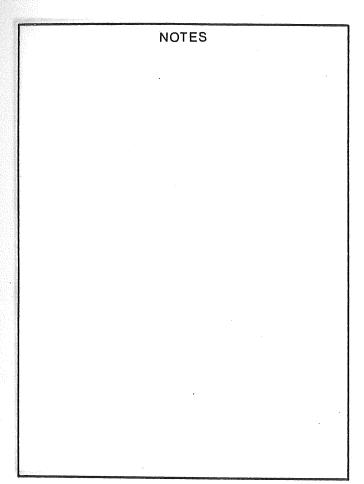
The nasal chemical senses of garter snakes constitute the most important components of the sensory systems used by them in intraspecific communication and prey recognition. it is known that garter snakes can respond differentially to airborne odorants (Burghardt, 1977; 1979; Halpern, J. et al., 1985; Halpern & Kubie, 1983), only one previous study (Plough & Kubie, 1980) has demonstrated that snakes can learn a task using airborne odors as discriminative stimuli. In the present study seven adult male snakes (Thamnophis sirtalis sirtalis) were trained, using a correction procedure, to traverse a two-choice maze using the presence or absence of amyl acetate (AA) odor as the conditioned stimulus. The snakes were pretested for odor vs non-odor preference and were trained to go to the initially non-preferred stimulus. Two of the seven snakes showed a strong initial preference for the side without AA, otherwise no significant preferences were noted. Five of the seven snakes achieved a predetermined criterion (2 training sessions above the .05 confidence level) within 85 trials. Snakes (N=4) trained to go away from AA reached criterion performance in fewer trials (M=41.2 trials) than the snake trained to go toward AA (70 trials). The two snakes failing to reach criterion within 85 trials were both training to go toward AA.

Supported by NINCDS grant NS11713

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26 A Possibly New Axon-Bearing Spiny Neuron in the Olfactory Bulb of Developing Rats. E. Meisami, (Physiology Dept. Univ. Illinois, Urbana, IL 61801)

a rapid Golgi study of the developing rat olfactory bulb an interesting axon-bearing spiny neuron type was encountered that may not have been described before. The cell body resembles the mitral shape and is located in or near the mitral cell layer. A branched axon pierces into the internal granular layer. A prodigious dendritic bush bearing numerous spines radiates into the external plexiform layer, not unlike the basal dendrites of the mitral cells. The cell is found also in the newborn's bulbs showing similar axonic and dendritic morphology but smaller in size and spine number. Following measurements were obtained from several newborn and weanling rats: The cell body's vertical and horizontal diameters are 18 and 10 um in the weanling and 12 and 12 um in the newborn, respectively. The diameter of the dendritic field is about 500 um in the weanling and 100 u in the newborn. The total number of spines on the dendrites is about 1200 in the weanling and 100 in the newborn; spine morphology is similar to those of the granule cells. In both the newborn and weanling about 10 spines is found on the cell body. Thus these neurons appear to be somewhat smaller than the mitral cells but much larger than the granule cells found in the mitral cell layer. This neuron type cannot be a mitral subtype because of the absence of apical dendrite and the presence of highly spiny horizontal dendrites. It may either be a giant axon-bearing dendrites. It may either be a giant axon-bearing granule cells or a subtype of the short-axon cells. The function of this cell is unclear but its precocious morphology in the newborn rats that exhibit limited but sufficient olfactory ability for nipple location may provide some clues.



127 A Longitudinal Study of Sucrose Intake in the Fischer-344 Rat. JAMES C. SMITH, LAURA WILSON and DANA MERRYDAY, (The Florida State University).

Supported by a Grant from NIA, 5 RO1 AG04932

Observations were made on the effects of aging on the taste of sucrose in the Fischer-344 rat. Ninety-six weanling rats were divided into six groups receiving 1.0, 0.5, 0.25, 0.125, or 0.0625 Molar Sucrose. The sixth group received no sucrose. For 4 days each week the rats had access to ground Purina Chow, a water bottle and the bottle containing their particular sucrose concentration. Measurements of ingestion were made daily and body weights were recorded weekly. Recordings for the first 80 weeks are reported here. Increases in body weight were significantly different across the six groups by week 15. These increases are directly related to the concentration of sucrose available. Total caloric intake is higher for the groups receiving the higher concentrations of sucrose. Caloric intake from the sucrose increases across the groups in a linear manner with the increase in concentration of the sucrose. Caloric intake from the Purina Chow decreases in a similar fashion. In a comparison of weeks 15 and 70, it can be seen that sucrose intake for the 0.0625, 0.125 and 0.250 groups was significantly higher for the 15week measurements. When ingestion is corrected for body weight, the 15 week sucrose intakes are higher for all sucrose concentrations. Mean food and fluid intakes are remarkably stable across the 80 weeks of testing. However, variability in the water group is near zero, but quite high in the sucrose groups. At approximately six month intervals, a sample of six rats from each group were placed in special cages equipped with sensors at the ingestion sites, allowing for comparison of patterns of ingestion as the rats grew older.

<u>Salt Appetite in Hamsters is Linked to Escape from Retention Effects of DOCA.</u> RUDY A. BERNARD, TIMOTHY W. PRIEHS and KAREN MOONEY (Dept. of Physiology, Michigan State University, East Lansing MI 48824).

Sodium replete rats and hamsters develop a large salt appetite in response to deoxycorticosterone acetate (DOCA). This effect is considered a behavioral counterpart of the sodium conserving role of the mineral corticoid hormones and as support for the theory that salt appetite is regulated by the renin-angiotensin-aldosterone (RAA) system. Most of the procedures employed in the study of salt appetite, such as adrenalectomy, dietary sodium restriction, I.P. dialysis, parotid gland fistulation, formalin injection, administration of furosemide and other diuretics, tend to reduce plasma sodium and consequently achieve their effect by activating the RAA system. The resulting increase of sodium intake is a classic example of regulatory behavior.

DOCA-stimulated salt appetite, however, cannot be readily explained by the same mechanism, for it is initiated by positive rather than negative sodium balance, which suppresses the activity of the RAA system, and the resulting salt appetite does not promote sodium homeostasis. We propose instead that the stimulation of salt appetite by DOCA is related to the phenomenon of renal escape; in which the initial sodium retention produced by DOCA is followed within a few days by a return to normal sodium balance in spite of continued hormone administration. Renal escape is now believed to be due to the release of endogenous natriuretic factor(s) through the stimulating effect of plasma volume expansion. It is our hypothesis that DOCA-stimulated salt appetite is due to activation of the same endogenous natriuretic system.

We undertook the experiments reported here after Chimoskey et al. (1984) reported that the atria of BIO 14.6 hamsters were deficient in natriuretic factor. We found that these hamsters failed to escape from the salt and fluid retention effects of DOCA, gaining 56% of their initial body weight in 10 days, whereas the controls gained only 5%. During this time the myopath hamsters did not increase their salt and water intake significantly, whereas the controls increased their sodium intake by over 80% and their water intake by over 250%. It is clear from these results that stimulation of salt appetite occurs only in the animals that can escape retention by activating a natriuretic mechanism. The failure to escape from the high endogenous levels of mineral corticoids produced by sodium deficiency is explained by the absence of volume expansion in this experimental situation.

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Comparison of Aspartame and 4-Chloroaspartame Taste Characteristics in Mice and Monkeys. CHARLES N. STEWART, MARCUS W. THOMSEN and JOSEPH M. DALTON (Franklin & Marshall College, Lancaster PA 17604)*

While humans readily accept aspartame as a sucrose substitute, a number of studies show that rodent species fail to display a preference for the substance in solution or show only a mild preference at high concentrations. In addition rats and hamsters fail to generalize a conditioned flavor aversion from aspartame to sucrose suggesting that they do not classify it as "sweet". In the studies reported here, we show that Mus musculis generalizes a conditioned flavor aversion from aspartame to quinine. Also, this species shows an even greater generalization to quinine from chloroaspartame which has chlorine on the 4 position of phenylalanine. Our results parallel those obtained from studies with gerbils which show a greater electrophysiological response to 6-chlorotryptophan than to tryptophan, a substance which is "coded" as sweet by this species. In a parallel study with <u>Maccaca mulatta</u> we find that this species displayed a strong preference for aspartame. This preference was reduced when the halogenated compound was used. The 4-chloroaspartame was prepared in four steps in overall yield of 39% by coupling N-(thiocarboxy)-Laspartic anhydride with 4-chlorophenyl alanine methyl ester using a method similar to the one developed by Vinick and Jung (1982). The product had a melting point of 148-152° C. Infrared and nuclear magnetic resonance spectral analyses were consistent with the product's structure.

Ref. Fredric J. Vinick and Stanley Jung, Tetrahedron Letters, Vol. 23, No. 13, pp. 1315-1318, 1982.

*This research was supported by the Grants Committee of Franklin & Marshall College. The aspartame was a gift from The G. D. Searle & Co.

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Sucrose Detection Thresholds Before and After Peripheral Gustatory Nerve Transection in Rats. A. C. SPECTOR (Univ. of Penn.), G. SCHWARTZ (Monell Chem. Senses Ctr.), & H. J. GRILL (Univ. of Penn.).

Animal psychophysical research has not focused on the integrity of the taste system in taste processing. We developed a taste-testing apparatus for this purpose. Thirty drinking spouts are mounted to a motor-driven wheel. Rats have access to one spout at a time through a small opening in the chamber. Seven 17 G tubes are positioned directly above this "access" spout. Each tube is connected to 7 miniature solenoid valves, which, in turn, connect to 7 fluid resevoirs. Small volumes of taste stimuli are deposited into the "access" drinking spout and immediate behavioral responses (e.g., spout contact) are measured by a microcomputer. Contamination from previously delivered tastants is eliminated by rotating the wheel which provides a "fresh" spout. This apparatus was used to measure sucrose detection thresholds in a shock avoidance paradigm. Two water-deprived rats were trained to maintain spout contact for intermittent water reinforcement (70 ul); taste trials (70 ul) were delivered on a variable interval schedule. Taste trials, as well as reinforcement trials, were 5 sec in duration; spout contact was measured. If the rat had any contact with the spout during the latter 3 sec of the taste trial, it received a mild footshock. Lack of consistent spout contact during a water trial resulted in a 15 sec time-out. The sucrose concentrations used were: 0.3 M, 0.1 M, 0.03 M, 0.01 M, 0.005 M, and 0.001 M. Rats reliably detected 0.01 M, whereas neither could discriminate 0.001 M from water as determined by several dependent measures. Marginal degrees of discrimination were observed with 0.005 M sucrose. Preliminary results demonstrate that bilateral chorda tympani section had little effect on sucrose sensitivity. Results from these same rats following bilateral glossopharyngeal transection will also be presented. The successful use of this paradigm to reliably measure detection thresholds in intact and nerve-sectioned rats demonstrates its utility in elucidating the neural basis of taste processing. Future work will focus on the role of other portions of the gustatory system with respect to taste sensitivity and quality discrimination.

Taste Quality Distinctions Between Maltose and Sucrose in Rats: Issues Involving the Generalization of Conditioned Taste Aversions. ALAN C. SPECTOR (Univ. of Penn) and Harvey J. GRILL (Univ. of Penn.).

Recently, generalization of conditioned taste aversion (CTA) has been used to determine gustatory similarity among chemicals. We address the critical question of which psychophysical dimension (intensity vs. quality) contributes to the generalization. For example, it has been hypothesized that maltose and sucrose may produce qualitatively distinguishable taste sensations for the rat as opposed to evoking one taste sensation (e.g., "sweetness") differing in intensity. To overcome this interpretive weakness, we used small volumes of taste stimuli (100 ul) to test for both intrachemical and interchemical generalizations of a CTA when either 0.1 M sucrose or 0.1 M maltose served as the conditioned stimulus (CS). In Experiment 1, six rats were given a CTA to 0.1 M sucrose by pairing 15 min access of the CS in the home cage with a 3.0 mEq/kg dose of LiCl on two occasions. Six control rats had water paired with LiCl. Rats had been previously trained to maintain spout contact for intermittent water reinforcement (100 ul) in a speciallydesigned apparatus. All rats were tested for their avoidance of 100 ul samples of sucrose and maltose of the following concentrations: 0.03 M, 0.1 M, & 0.3 M. Rats received $1\bar{0}$ trials (5/session) of each stimulus quasi-randomly presented in 2 sessions. Results indicated that all sucrose concentrations were avoided (in Exp. group), but only the 0.3 M concentration of maltose was avoided. The lowest sucrose concentration was significantly less avoided than the higher concentrations. In addition, the intrachemical generalization gradient broadened with extinction, whereas the interchemical generalization steepened. In Experiment 2, different group of rats were treated identically except that O.1 M maltose served as the CS. With minor exceptions, the results of Experiment 2 paralleled, in principle, the results of Experiment 1. In conclusion, qualitative differences between maltose and sucrose explain the outcomes of these experiments better than differences in the relative "sweetness" of these sugars.

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Taste and Bulimia. ADAM DREWNOWSKI (University of Michigan School of Public Health), FRANCE BELLISLE (College de France, Paris, France), PIERRE AIMEZ and BRIGITTE REMY (Hotel-Dieu, Paris, France).

Binge-eating episodes in bulimia involve the consumption of sweet, calorie-dense foods -- such as ice cream, cookies, or other desserts. It may be that this choice of binge foods is reflected in altered profiles of taste responsiveness. Sensory perceptions and preferences for sweetness and fat were examined in 16 normal-weight women with a diagnosis of DSM IIIR bulimia (mean age 26.7; mean wt 56.7 kg), who were commencing psychotherapy at Hotel-Dieu Hospital in Paris. Sixteen normal-weight females (age 28.9; wt 55.8 kg) of comparable age and socioeconomic status served as controls. The taste stimuli were 15 semi-liquid mixtures of desserttype soft white cheese ("fromage blanc") containing 0, 20, or 40% fat as solids wt/wt, and sweetened with 1, 5, 10, 20, or 40% sucrose (w/w) in a 3x5 factorial design. The samples were presented chilled to 5°C and in a randomized order. The subjects used 9-point category scales to rate the perceived sweetness, fatness, and creaminess of the stimuli. and assigned an overall hedonic preference rating to each sample. Taste responsiveness profiles were modelled using the Response Surface Method (RSM). On the average, bulimic patients and normal controls did not differ in their estimates of sweetness intensity. However, the two groups did show distinct profiles of taste preference. The optimal stimulus sweetness level was 15% for bulimic patients and only 9% for normal controls, while optimal fat levels were lower for bulimic patients relative to controls. The present data are consistent with previous reports that patients with eating disorders crave sweetness but often dislike fat-containing foods.

Perceptual and Salivary Responses to Oral Chemical Irritants Among Frequent and Infrequent Consumers of Hot Spices. BEVERLY J. COWART (Monell Chemical Senses Center, Philadelphia, Pa.).

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Previous studies (e.g., Lawless et al., Chem. Senses, 1985, 10, 579-589) have shown that frequent consumers of chili peppers assign lower intensity ratings to the "burn" produced by a given concentration of capsaicin than do infrequent consumers, although there are suggestions this difference may primarily reflect differences in the perceptual processing of irritant sensations rather than sensory desensitization resulting from frequent pepper exposure. The present study further explored this phenomenon by assessing salivary and perceptual responses to a range of suprathreshold concentrations of two chemical irritants in groups of frequent (n=12) and infrequent (n=12) consumers. The irritants and their concentrations were: synthetic capsaicin (CAP: 0.5, 1.0, 2.0 and 4.0 ppm) and piperine (PIP: 12.5, 25.0, 50.0 and 100.0 ppm). Subjects received each series in separate sessions and rated perceived irritation using. magnitude estimation. Measures of salivary flow were obtained before and after presentation of each stimulus, and the change in flow rate was determined. Results indicate that burn ratings of frequent consumers were significantly lower than those of infrequent consumers across concentrations of both irritants; no group differences in slope were evident. However, the difference between groups in mean ratings tended to be greater in their responses to CAP than in responses to PIP. No significant difference between groups in mean salivary responses to oral irritation was observed although means for infrequent consumers consistently fell above those of frequent consumers. Again, there was some tendency for mean group differences to be greater in response to CAP than to PIP. These data further indicate that reductions in perceived irritant intensity among frequent spice consumers do not reflect substantial peripheral desensitization. Nonetheless, they suggest that marginal desensitization may occur and that differences among individuals in responses to oral chemical irritants may to some extent be irritant specific.

*Supported by NIH Grant #NS-20616

 $\frac{\text{Effect of Cooling on the Sweetness of Natural and Artificial}}{\frac{\text{Sweeteners.}}{\text{(Monell Chemical Senses Center).}} \frac{\text{Effect of Cooling on the Sweetness of Natural and Artificial}}{\text{BARRY G. GREEN}}$

Previously we reported that the sweetness of sucrose and the bitterness of caffeine were reduced in intensity when the tongue was cooled to 20°C, whereas the sourness of citric acid and the saltiness of sodium chloride were unaffected. In contrast, cooling only the solution had slight but insignificant effects on sweetness and bitterness. Because the critical variable for obtaining a cooling effect was the temperature of the tongue, it was hypothesized that variations in temperature sensitivity across substances were more likely due to differences in receptor mechanisms than to differences in the molecular properties of the solutions. We therefore selected four additional sweet-tasting substances to learn if cooling the tongue and/or the solution would affect the perception of all sweet stimuli similarly. Magnitude estimates of sweetness were obtained for five concentrations (in quarter log unit steps) of fructose (0.095-0.95 M), glucose (0.18-1.8 M), saccharin (0.002-0.003 M) and aspartame (0.0004-0.004 M) in each of four conditions: (1) tongue and solution at 20°; (2) tongue at 20° and solution at 36°; (3) tongue at 36° and solution at 20°; (4) tongue and solution at 36°C. The temperature of the tongue was manipulated by having the subject rinse repeatedly with cold (ca. 5°C) or warm (ca. 38°C) water (tongue temperature was monitored with a micro-thermocouple). Preliminary results indicate fructose and glucose have temperature sensitivities similar to that of sucrose, whereas the sweetness of saccharin appears to be unaffected by cooling. The sweetness of aspartame is affected in a complex manner that may be indicative of an interaction between the temperatures of the tongue and the solution. Thus, although the results are more readily explained by a multiple- than by a single-receptor theory of "sweet" chemoreception, the data for aspartame imply that thermallyinduced changes in the molecular properties of the stimulus may also affect perceived sweetness.

Supported by funds from the Dairy Research Foundation and the National Institutes of Health (NS 20577).

135 <u>Spatial Summation of Chemical Irritation on the Tongue.</u>
BARRY G. GREEN (Monell Chemical Senses Center)

Two experiments were carried out to discover the extent to which the amount of lingual skin exposed to a chemical irritant (ethanol) affects the perceived intensity of irritation. Experiment 1 investigated the effect of doubling and quadrupling the area of a single stimulus on both the perceived intensity and the latency of onset of irritation. The stimuli, which were rectangles of filter paper (0.5-2.0 cm²) saturated with solutions of ethanol and water (35-85%), were placed across the midline of the tongue 3 cm posterior to the tip. Magnitude estimates of perceived irritation did not increase significantly with stimulus size, and the latency to the onset of irritation was shorter only for the largest stimulus. A second experiment was run to learn if the same result would be obtained when stimulus size was increased by adding a second stimulus rather than by increasing the size of a single stimulus. Subjects judged the perceived intensity of ethanol irritation produced by two pieces of filter paper placed across the midline of the tongue (separated by 2 cm) when either one or both of the papers was saturated with ethanol (45-85%). Under those circumstances, doubling stimulus area produced an approximate 50% increase in perceived intensity together with a significant reduction in the latency to sensation onset. The implication of these data is that spatial summation of chemical irritation may occur only when irritation is induced at more than one lingual locus. Experiments are continuing in order to investigate the spatial characteristics of summation in greater detail.

Supported by grants NS 20616 and ES 04356 from the National Institutes of Health.

SVEET AND BITTER PERCEPTION IN PTC TASTERS AND NON-TASTERS: A MULTI-HEASURE APPROACH. Aliki Akontidou, Ross Carter & Robert A. Frank (Dept. of Psychology, Univ. of Cincinnati).

This experiment explored sweet and bitter perception differences in (PTC) tasters and non-tasters using three measures; absolute thresholds, category ratings and intensity ratings over time. Eighteen tasters and non-tasters of PTC (total n = 36) were selected based on their thresholds and category ratings for PTC. In the next phase of the experiment, absolute thresholds were determined for sucrose, sodium saccharin, nechesperidin dihydrochalcone and quinine sulfate. A two alternative, forced choice procedure was used which required subjects to discriminate the test solution from distilled water. Once thresholds had been determined, the subjects made intensity ratings for suprathreshold concentrations of the test substances using a 21 point category scale. Intensity ratings for PTC and distilled water were also obtained. In the final phase of the experiment, the subjects rated the intensity of the stimuli over time. This was accomplished by having them continuously rate the intensity of a stimulus by moving a joystick whose output was monitored by a microcomputer.

Non-tasters demonstrated consistently higher thresholds for all the substances that were tested, but the difference is thresholds reached significance in only one case (sucrose, p<.05). Non-tasters gave significantly higher intensity ratings to the suprathreshold stimuli in the category rating phase of the experiment (p<.01). This seemingly paradoxical result may be explained as either an anchoring effect induced by the presence of PTC stimuli in this task, or an attempt by non-tasters to compensate for a sensory deficit by over-estimating the intensity of the stimuli. Analyses of the time/intensity data revealed that tasters and non-tasters did not differ in terms of total rating time, maximum rating, stimulus rise time or adaptation time of any of the stimuli (except PTC). This pattern of results suggests that (1) the measures obtained from the time/intensity tests are less sensitive to the anchoring/compensatory effects observed with category ratings, and (2) gustatory perception differences between PTC tasters and non-tasters are subtle and task-dependent.

This research was supported by a University of Cincinnati Research Council grant to Robert A. Frank.

137 A PRELIMINARY STUDY OF CHOCOLATE PERCEPTION IN PTC TASTERS AND NON-TASTERS. Debra L. Korchmar, Aliki Akontidou, & Robert A. Frank (Dept. of Psychology, Univ. of Cincinnati).

Most of the previous research assessing perceptual differences in PTC tasters and non-tasters has used aqueous solutions. The potential commercial implications of these differences merits investigation of differential sensitivity to and/or preference for food items. Chocolate is a particularly relevant food substance because it contains both bitter and sweet components. We were interested in investigating whether there are differences in the perception of chocolate in tasters and non-tasters, and whether these differences predict preference for different types of chocolate. The subjects (N=40) filled out a questionaire about how much they like chocolate and what types of chocolate candy they usually eat. They then sampled Netle's milk and semi-awest chocolate morsels and made overall hedonic and intensity ratings using 21-point category scales. The two groups were not different in their hedonic or intensity ratings for milk chocolate, or in their intensity ratings for bittersweet chocolate. However, as predicted, the hedonic ratings for bittersweet chocolate. However, as predicted, the hedonic ratings for bittersweet chocolate. However, as predicted, the hedonic ratings for bittersweet chocolate. However, as predicted, the hedonic ratings for bittersweet chocolate. However, as predicted, the hedonic ratings for bittersweet chocolate. However, as predicted, the hedonic ratings for bittersweet chocolate. However, as predicted, the hedonic ratings for bittersweet chocolate. However, as predicted, the hedonic ratings for bittersweet chocolate. However, as predicted, the hedonic ratings for bittersweet chocolates. However, as predicted, the hedonic ratings for bittersweet chocolates. However, as predicted, the hedonic ratings for bittersweet chocolates. However, as predicted, the hedonic ratings for bittersweet chocolates. However, as predicted, the hedonic ratings for bittersweet chocolates. However, as predicted, the hedonic ratings for bittersweet chocolates. However, as predicted, the hedonic ratings for bitters

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Effects of Context on Sweetness Evaluation: Generalization

Between Different Flavors. Teresa Anne Vollmecke & Sharon
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Recent experience, or context, exerts a potent influence on current sensory/perceptual evaluations (Helson, 1964; Parducci, 1965). Experience with one beverage (sweetened Kool-Aid) influences subsequent sweetness evaluations for the same beverage (Riskey et.al., 1979). The current study reports that a sweetness context with one flavor influences perceived sweetness of a second flavor. Four groups of 15 subjects (two flavors x 2 contexts) were exposed to a set of sucrose beverages using a sip and rinse procedure. Beverages were evaluated for sweetness and pleasantness with a visual analog scale. Each subject received 34 samples of one flavor (High or Low Context) followed by the same 8 sucrose test samples of the second flavor. The context consisted of 8 sucrose concentrations ranging from .06 to 1.6 M where half the subjects received more low concentrations (Low Contexts) and half received more high concentrations (High Contexts). A robust effect of context on sweetness was found in the main session with Low Context subjects giving higher sweetness evaluations than High Context subjects. Group pleasantness as a function of concentration level demonstrated the normal U shaped curve for the main session with the Low Context group evaluating the lower concentrations as more pleasant. A comparison of sweetness and pleasantness values in the test session showed significant differences between High and Low Contexts. Subjects with a prior Low Context experience evaluated the same concentration as sweeter than subjects with a prior High Context experience. These results indicate that the effects of context extend beyond a particular flavor. The extent of this generalization remains to be determined.

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Percep. & Psychophysics 26(3):171-176

The Relationship between Carbohydrate and Simple Sugar Intake and Measures of Sweet Taste Preference in Free-living Adults. RICK BELL (New York University), MABEL M. CHAN (New York University)

The relationship between carbohydrate intake and simple sugar intake, and sweet taste preference was studied. Seven-day diet records of twelve normal weight, non-smoking, healthy subjects, ages twentyseven to fifty-five, were analyzed for carbohydrate, glucose, fructose, and sucrose content. Intake data were compared to responses of four measures of sweet taste preference: a category scale and visual analog scale, which measured preferred aspartame level in oatmeal; an adjustment task, which measured optimal concentration of sucrose in water; and a questionnaire, which measured the daily frequency of sweet food consumption. Pearson product-moment correlations show that each individual taste measure, except for the questionnaire, could predict only sucrose intake (all p < .01); but results of a discriminant analysis indicate that the combined sweet taste preference measure responses could be correlated with the combined intake of sucrose, glucose, and fructose (p < .03).

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Rate Alpha-amylase Secretion. DEBORAH A. FROEHLICH 1
(Department of Food Science and Technology, University of California, Davis, CA 95616 U.S.A.), ROSE MARIE PANGBORN (Department of Food Science and Technology, University of California, Davis, CA 95616 U.S.A.)

Unilateral parotid saliva was collected from ten subjects following oral stimulation with water (as baseline), chewing of parafilm (1 chew/sec and 2 chews/sec), mastication (1 chew/sec) of 3.0 g celery, 1.5 g and 3.0 g bread, and holding of 1.5 g bread in the mouth. Mastication of a bread and celery food resulted in higher flow rates than did chewing a parafilm. Although limited to two foods at two weights, the results suggest a possible dependence of flow rate on both the type and weight/size of the stimulus. Salivary flow induction was independent of chewing rate. Mastication did increase the secretion rate but not the concentration of protein and alphaamylase. The ratio of alpha-amylase to protein secreted remained relatively constant with various types of stimulation, as indicated by similar values for specific activity of alpha-amylase. The rate of alpha-amylase secretion was influenced by mastication in accord with digestive function, in that more secretion was induced by food than by nonfood stimuli, more by bread than by celery, and more by larger than smaller pieces of bread. However, these increases might be related more to the physical characteristics than the composition of the stimuli, e.g., force of chewing, dryness, and surface area. In general, an increase in stimulated flow rate led to an increase in Na+ concentration, while K+, Ca++, and Mg++ concentrations remained relatively constant.

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Structure, Taste and Solution Properties of

5-Membered Rings

SYED SHAMIL & GORDON G BIRCH
(University of Reading)

Although 5-membered ring skeletons are devoid of taste properties, substitution around the ring creates taste or olfactory potential. Hydrophobic substituents may create specific olfactory properties but polar substituents are likely to create taste effects. The criterion is the effect of the substituent on water structure which can be monitored by measurements of solution properties. Apparent specific volumes of 5-membered ring structures allow the effects of different types of ring substituent to be quantified. The tastes of these simple cyclic structures are then largely predictable.

AFRC is thanked for a grant in aid of this research. A travel grant in support of this work is acknowledged from ECRO.

Sensory Effects of Fat Content in Food. DAVID J. MELA (Monell Chemical Senses Center, Philadelphia, PA).

There is little information on the sensory aspects of food lipids and their interactions with other food components. Excessive fat consumption has been implicated in the etiologies of a number of chronic diseases, and decreased fat intakes have been recommended for western populations. Thus, there is a need to understand how the sensory qualities of foods are affected by differences in their lipid content. The present studies analyzed the relationships between lipid content and perceptions of oiliness and flavor in real food.

A dry, corn-based snack food product was prepared with 10 to 50% (by weight) added exterior oil (hydrogenated soybean oil). Samples were either plain, or had a constant level of added salt or vanilla flavor. Paired comparisons with plain samples demonstrated that a difference in fat content of at least 10 gm/100 gm was necessary for consistent discrimination of differences in oiliness. The perception of oiliness was unaffected by the addition of salt or flavoring.

Perceived intensity of oiliness, saltiness, and vanilla flavor were assessed using a magnitude estimation procedure. The power function exponent relating oiliness to fat content in all three types of samples was approximately .5-.7, and was unchanged when subjects were tested wearing gloves and nose clips in dim red light. This suggests that perceptions of oiliness are largely derived from textural effects sensed in the oral cavity. The perceived intensity of both saltiness and vanilla flavor increased with increasing fat content in these samples.

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Bitter Taste Sensitivity to Caffeine in Users and Non-Users.
DAVID J. MELA, RICHARD D. MATTES, SHUYA TANIMURA,
MICHAEL ADAMS, CAROL CHRISTENSEN (Monell Chemical Senses
Center, 3500 Market St. Philadelphia, PA 19104).

Interactions between gustatory function and dietary behavior remain incompletely understood. The primary aim of this project is to evaluate dietary effects on taste; more specifically, the influence of caffeine consumption on taste sensitivity to caffeine and other bitter compounds.

Twenty-four adult subjects were divided into 3 groups depending upon their caffeine consumption: non-users, moderate users (200-300 mg/d), and heavy users (more than 500 mg/d). Detection thresholds and time-intensity data were obtained twice, one week apart. Before each test, stimulated saliva was collected for the determination of caffeine concentration by HPLC. Detection thresholds were determined by ascending triangle tests. Subjects generated time-intensity (T/I) curves on a strip-chart recorder for 5 caffeine solutions (0.79, 1.99, 5.0, 12.5 and 30.5 X 10⁻³ M) presented randomly in each session. Peak height and area under the T/I curve were computed. Group differences in gustatory responses were assessed by Kruskal-Wallis ANOVA and post-hoc Mann-Whitney U-tests.

Detection thresholds among the non, medium and heavy caffeine users were $5x10^{-4}$, $2.61x10^{-3}$ and $1.31x10^{-3}$ M respectively. Caffeine users were significantly less sensitive to the compound than nonusers (p < .02). No group differences in T/I data were observed. Analyses of salivary caffeine levels are currently underway and will be correlated with threshold data to ascertain whether group differences may be related to adaptation effects.

Supported by NSF Grant #BNS-8418953

144 <u>Chemosensory Magnitude Matching.</u> V.K. STONE, L.E. MARKS, J.C. STEVENS, and L.M. BARTOSHUK (John B. Pierce Foundation Laboratory)

In magnitude matching, stimuli of different modalities are alternated within a single experimental session and subjects try to judge the suprathreshold sensation magnitudes on a common scale. To test the stability of derived cross-modal matches, subjects judged in different sessions different sets of NaCl concentrations (0.032-0.32 M in one session and 0.18-1.8 M in the other) while the set of intensity levels of low-frequency white noise remained constant (40-96 dB). Derived magnitude matches showed considerable lability ("contextual bias"), both when responses were made by magnitude estimation and by graphic rating. Olfactory functioning was then compared in young and elderly. When both groups made magnitude estimates of identical concentrations of 1-butanol (0.08-6.25%) and NaCl (0.032- 0.32 M), the odor function of the elderly was depressed 23% relative to that of the young. This difference between old and young is potentially confounded, however, by "contextual bias". In the second session, therefore, butanol concentrations were decreased for the young (0.03-2.78%) and increased for the elderly (0.31-100%). Magnitude matches obtained with these revised stimulus sets yielded more equivalent perceptual levels in the old and young, and, moreover, revealed a larger depression in the odor function of the elderly (greater than 50%). Despite the pervasive effects of context, with appropriate precautions magnitude matching can prove a valuable tool in the assessment of chemosensory functioning.

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145 EFFECTS OF DIFFERENT RESPONSE TASKS ON DISCRIMINATION OF CHEMOSENSORY STIMULI OF VARYING COMPLEXITY. M. Albright (T. J. Lipton, Inc., Englewood Cliffs, N.J.)

Four response tasks were compared. The tasks differed in levels of measurement: 1) interval (line marking), or 2) ordinal (ranking); and levels of analysis: analytical (specific descriptive attributes), and 2) integrated (proximity to a reference). Twenty-four panelists completed evaluations of a set of stimuli using each of the four tasks. The stimuli were water solutions of three levels of sucrose, two levels of a tastant (caffeine), and two levels of an olfactant (orange essence). Variations in the kinds of responses between tasks, individual panelist response patterns, and discrimination sensitivity of the tasks are described. Practical implications for experiments is discussed.

Chronic Electrophysiological Recordings of Vomeronasal Pump Activation in Awake Animals. MICHAEL MEREDITH (Dept. Biol. Sci., Florida State University, Tallahassee FL)

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The vomeronasal sensory epithelium is sequestered inside the vomeronasal organ (VNO) capsule. Chemical stimuli can be delivered by a vasomotor pumping mechanism. To investigate the circumstances of pump operation, electrical changes in the VNO of awake hamsters responding to putative VNO (and other) stimuli were recorded. A low voltage (40 mv) AC carrier signal was passed between electrodes implanted into each end of the VNO capsule and recorded through a phase lock amplifier (PLA). MA signals are proportional to the resistance and capacitance between the electrodes and extraneous electrical noise is largely eliminated. The animals in these experiments would sleep, eat and mate while connected to the recording circuit-via a head plug and flexible cable. They showed no response when the the carrier voltage was switched on and presumably could not detect it. When another animal was introduced into the cage of a resting implanted male, the relatively flat baseline was replaced by a series of transitory displacements in the direction of decreased electrical resistance, having a period of about 1-3 sec (1-0.3 Hz). These oscillations generally summated to produce a large displacement, especially if the animal was resting or asleep at the beginning of the test. The oscillations were similar to those produced from similarly implanted electrodes when the autonomic supply to the VNO was stimulated repetitively in anesthetised animals. The signals are interpreted as due to repeated vasomotor movements which summate to produce a net change in vasomotor tone. In alert animals, the oscillations could appear without summation. The response was not restricted to reproductive contexts. Male, unreceptive-female and receptive-female intruders all elicited "pump" signals as did other novel stimuli such as the transfer of the implanted animal to a fresh cage. These signals did not appear if electrode wires were implanted under the palatal mucosa but outside the VNO capsules. The signals were not synchronized with heartbeat or respiration, were not associated with chewing, sniffing or movements of the head or body and they did not occur when the animal was anesthetized. Supported by NSF grant BNS 8412141

147 Nasal Trigeminal Chemoreception: Response to Nicotine.
WAYNE L. SILVER and DIANNE B. WALKER. Department of
Biology, Wake Forest University, Winston- Salem, NC 27109.

Trigeminal receptors in the nasal cavity respond to a variety of chemical stimuli. Although some of these stimuli appear to be non-irritating, trigeminal chemoreception is often considered a part of the common chemical sense whose primary function is to elicit protective reflexes when stimulated with irritating compounds. Compared to olfaction, however, there is still relatively little quantitative information available about the kinds and concentrations of chemical compounds which are effective trigeminal stimuli. In the present experiment we examined the effectiveness of nicotine as a trigeminal stimulus. Multiunit activity was obtained from the ethmoid branch of the rat trigeminal nerve. Nicotine was delivered in the vapor phase via a microprocessor controlled, air-dilution olfactometer. Dilution ratios were controlled by electronic mass flow controllers. Concentration-response curves and thresholds (defined as the concentration which first elicited a response discernible from baseline) were obtained from six rats. The response to nicotine differed from responses to other compounds in that it only gradually returned to baseline. Threshold was approximately 4 ppm. Response magnitude increased with increasing stimulus concentration up to about 23 ppm and did not increase further, even at vapor saturation (108 ppm). These results demonstrate that nicotine is an effective trigeminal stimulus.

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Trigeminal and Topographical Characteristics of Odors. M.R.GARCIA-MEDINA, J.E.COMETTO-MUNIZ & A.M. CALVINO.(Laboratorio de Investigaciones Sensoriales, CONICET - Esc. Sal. Públ., Fac. Medicina, UBA, C.C.53, 1453-Buenos Aires, Argentina).

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The aim of the present work is to determine if normal subjects can discriminate among trigeminal characteristics of odors and also to verify if different odors are perceived in different areas of the respiratory tract, from the tip of the nose to the chest (as some everyday experiences suggest). Subjects'task was to describe a set of 30 different odors in terms of their trigeminal or pungent characteristics (burn, sting, tickle, cool, warmth, and irritation) and to localize the area in which each odor was perceived. There were twelve sessions in each of which subjects evaluated ten odors. Each odor was then evaluated four times per subject. Twenty subjects participated (ten males and ten females). Results were analized in terms of frecuency of mentions of each trigeminal characteristic and each area. The results are the following:1)subjects can discriminate between different trigeminal characteristics; 2)odors form a continuum in relation to their trigeminal char-acteristics from those with more intense to those with less intense trigeminal components; 3) odors with more intense trigeminal components (e.g.ethanol, acetic acid, menthol) are perceived more frecuently in the back of the nose, throat and sometimes, respiratory airways in the chest, whereas odors with less intense trigeminal component (e.g. cherry, cocoa,) are perceived in the first portions of the nose; 4) females report trigeminal components in odors that are nontrigeminal for males 5) when presented with the same group of odors females make more mentions of trigeminal characteristics than men and perceive the odors backwards in relation to males.

Behavioral assessment of olfactory and trigeminal responsiveness of starlings (Sturnus vulgaris) to 9
anthranilates. J. RUSSELL MASON (USDA/APHIS/ADC, Denver Wildlife Research Center, c/o Monell Chemical Senses Center) and LARRY CLARK (Monell Chemical Senses Center, 3500 Market Street, Philadelphia, PA 19104).

Irritating substances and the systems mediating irritation may differ fundamentally between birds and mammals. For example, dimethyl anthranilate, a flavoring preferred by at least some mammals, is aversive and apparently irritating to birds. We have begun investigations designed to elucidate relationships between the structure of anthranilate molecules and repellency. Also, we are conducting experiments to identify sensory systems mediating anthranilate detection. Here, we report a series of behavioral experiments that explored the responsiveness of Starlings to dimethyl anthranilate and 8 homologues (ethyl; isobutyl; isobutyl N,N dimethyl; isobutyl methyl; linalyl; methyl; phenyl ethyl; phenyl methyl). Repellency was positively correlated with steric and/or electrical obscuration of the amine function of anthranilate molecules. Tests with intact birds and birds given bilateral olfactory or naso-trigeminal nerve cuts demonstrated that both olfaction and nasotrigeminal chemoreception were important for detection and discrimination. Because combined olfactory and naso-trigeminal nerve cuts did not completely eliminate responding, we infer that gustation and/or oral trigeminal chemoreception may be important. Because behavioral experiments on taste, per se, are difficult to perform, we have begun a series of electrophysiological investigations of that sensory system to assess relative responding to the anthranilate series. Partial correlation analyses will be used to compare olfactory, trigeminal and gustatory electrophysiological responses: (a) to the mean repellency of the anthranilates, and (b) to steric and/or electrical obscuration of the amine function of the anthranilates.

NOTES

The terminal nerve in the bird CELESTE 150 R. WIRSIG and SCOTT F. BASINGER (Department of Baylor Ophthalmology, of Medicine. College Houston, Texas 77030).

There has been some question as to whether birds possess a terminal nerve (TN). In the present study, we provide positive evidence that the domestic chicken (Gallus gallus domesticus), at least during embryonic stages of development, does possess a TN. In fish and amphibians, FMRFamide-like immunoreactive (FMRFamide-ir) (FMRFamide-ir) material and acetylcholinesterase (AChE) are markers for the TN, so we explored the possibility that these markers would label the chick TN. Embryos between 13 and 19 days of development were perfused with Zamboni's fixative and heads were sectioned sagittally or horizontally (20 um) on a cryostat. Tissue was processed with immunocytochemical and/or histochemical procedures. Fusiform and multipolar cell bodies and their processes belonging to the TN contained FMRF-amide-ir material, AChE or both substances. These labelled neurons are found in ganglia or are dispersed along the medial aspect of the olfactory bulbs, within the medial portion of the olfactory and ophthalmic branch of the trigeminal nerves, and within plexuses in the lamina propria of the nasal septum. The TN enters the forebrain by sending fiber bundles through the medial portion of the olfactory bulb. The termination area of the main branch of the TN, which is found within the olfactory nerves, is within the lamina propria of the dorsal nasal septum at the junction between the olfactory and respiratory epithelium. It is not yet clear onto which structures the TN fibers actually end. Ultrastructural studies are in progress to examine the relationship between TN neurons and other nasal structures.

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The Relationship Between Nasal Anatomy and Human
Olfaction. Donald A. Leopold, David E. Hornung, Maxwell M.
Mozell, Steven L. Youngentob and George R. Petro (SUNY
Health Science Center at Syracuse)

The relationship between nasal cavity anatomy and olfactory ability was evaluated by correlating 30 measurements from nasal cavity CT scans with the results of an established clinical measure of olfactory function (Odorant Confusion Matrix, or OCM). Although all combinations of linear and logarithmic mathematical models were evaluated, the model that best described the experimental data utilized the logarithms of the anatomical measures and the logistic transform of the OCM percent correct score. In this model, two nasal cavity regions were found to be important factors in accounting for the olfactory test results. The most important region was located between the nasal septum and the middle turbinate, between 10 and 15 mm beneath the cribriform plate. As this space increased in volume, the olfactory ability improved. This region just above the inferior margin of the middle turbinate may serve to control the airflow to the more superior nasal regions. The second region was 5 mm high and located just beneath and anterior to the cribriform plate. It's volume was negatively correlated with the olfactory ability, and presumably affects flow characteristics in the region just anterior to the cribriform region. The results of this study support the hypothesis that changes in specific areas in the nasal cavity have a special impact on olfactory function.

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Spatial Organization of Receptor Surfaces and Odorant Passageways in the Hamster Nasal Cavity: Relationship to the Spatial Organization of Central Projections. THOMAS A. SCHOENFELD, ANDREW N. CLANCY and FOTEOS MACRIDES (Worcester Foundation for Experimental Biology, Shrewsbury, MA 01545)

Neuroanatomical studies in the hamster have revealed a rhinotopic organization of peripheral input to the main olfactory bulb (MOB) that is complemented by a topograpically organized intrabulbar associational projection system which interconnects opposing points on the medial and lateral sides within the MOB. The spatial resolution of the peripheral input appears to be finer circumferentially (with respect to the coronal plane) than longitudinally (along the rostro-caudal axis). Morphometric analyses of the nasal cavity (of. Clancy et al., this meeting) show that the maximum circumferential extent of the olfactory epithelium (OE) is much greater than its longitudinal extent. Equivalent analyses of the glomerular layer in the MOB show a similar ratio of maximum circumferential-to-longitudinal extents. This compression of the longitudinal axis for both the peripheral and central receptor sheets in part explains the apparent imprecision of rostro-caudal topography. However, the longitudinal resolution of the peripheral input is more course than that of the intrabulbar associational system, and their circumferential resolutions are comparably fine. The intrabulbar system thus can be conceptualized as series of high resolution reciprocal connections between successive points along opposing medial and lateral strips of peripheral innervation. The OE shows a 400% increase in its circumferential extent over a longitudinal distance of 2 mm in the region that overlies the septal window, and then a comparably rapid decline caudally. These changes are produced by the convolutions of the olfactory turbinates. The second endoturbinate extends 2 mm rostral to the septal window, and appears to divide the cavity caudally into medial and lateral channels lined by OE with non-overlapping projections to the medial or lateral side of the MOB. The associational system may then sharpen spatio-temporal differences in excitation along medial and lateral strips of peripheral input.

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Imposed and Inherent Mucosal Activity Patterns: Their Composite Representation of Olfactory Stimuli. M. MOZELL, P. SHEEHE, D. HORNUNG, P. KENT, S. YOUNGENTOB, S. MURPHY. (SUNY Health Science Center at Syracuse).

Both regional differences in mucosal sensitivity and a gas chromatographic-like process along the mucosal sheet have been separately proposed in two sets of earlier studies to produce different odorant-dependent activity patterns across the olfactory mucosa. This investigation evaluated, in one study, whether and to what degree these two mechanisms contribute to the generation of these activity patterns. Summated multiunit discharges were simultaneously recorded from lateral (LN) and medial (MN) sites on the bullfrog's olfactory nerve to sample the mucosal activity occurring near the internal and external nares, respectively. Artificially produced sniffs of four odorants (benzaldehyde, butanol, geraniol, and octane) were drawn through the frog's olfactory sac in both forward (H $_1$) and reverse (H $_2$) hale directions. By combining the four resulting measurements, LN $_{\rm H}$ 1, LN $_{\rm H}$ 2, MN $_{\rm H}$ 1 and MN $_{\rm H}$ 2, in different mathematical expressions, indexes reflecting the relative effects of the chromatographic process, regional sensitivity and hale direction could be calculated. Most importantly, the chromatographic process and the regional sensitivity differences both contributed significantly to the mucosal activity patterns, although their relative contributions varied among the four odorants. The more strongly an odorant was sorbed by the mucosa, the greater was the relative effect of the chromatographic process; the weaker the sorption, the greater the relative effect of regional sensitivity. Likewise, the greater an odorant's sorption, the greater was the effect of hale direction. Other stimulus variables (sniff volume, sniff duration, and the number of molecules within the sniff) had marked effects upon the overall size of the response, with both volume and duration having opposite effects for odorants strongly and weakly sorbed by the mucosa. How-ever, aside from the effects upon the absolute response magnitudes, these variables had only a minor influence, if any, upon the mucosal activity patterns.
(Supported by NIH Grant NS03904)

154 Regulation of Odorant Information Transmission Occurs within the Olfactory Nerve, G.D. ADAMEK (Osphresiopolis, Dept. Anat. & Cell Biol., Univ. of Cincinnati)

The anatomical arrangement of the olfactory nerve is unique among scnsory nerves. Its structure is such that it could regulate transmission of odorant information. In vertebrates it is comprised of about 10⁷ unmyelinated axons of uniform diameter of about 0.2 µm. Hundreds to thousands of these axons lie in groups surrounded only by thin sheaths of Schwann cell processes. Axons can be stimulated antidromically by electrical current from an electrode positioned in the olfactory bulb and orthodromically by odorants presented to the receptor epithelium. The activity of these axons can be measured with an extracellular electrode in the epithelium! At low stimulus currents, responses of single cells can be studied. Single cells respond to one electrical shock with a a single spike with a characteristic latency. At frequencies of 0.2, 0.5, and 1 Hz, the cell will respond to every shock or most of them with unchanged latency. Stimulation rates of 2, 5 and 10 Hz result in an increase in the latency, and the cell ultimately becomes inexcitable. During this process the number of failures until firing again commences is directly related to the stimulating frequency and the extent of recovery of the cell as manifested by decreases in latency shift of the evoked spike. At higher stimulating currents, the potential changes of other, more distant, activated cells are recorded as well and the compound action potential (CAP) appears. With continuous strong stimulation, the amplitude of the CAP decreases and there is an increase in latency. Stimulation of a multitude of olfactory neurons produces conduction failure within the olfactory nerve. This also occurs when the nerve is electrically stimulated during concomitant odorant stimulation. The amplitude of the electrically evoked CAP is greatly reduced during stimulation with a high concentration of amyl acetate. The CAP subsequently recovers. Amyl acetate activates a large fraction of the olfactory neuron population and leads to conduction failure of the axons. Therefore, orthodromic odor stimulation can lead to diminished olfactory bulb input due to axon conduction failure. I think that the safety factor for action potential propagation in the olfactory nerve is so low that this becomes the dominant factor controlling adaptation and odor-mixture suppression at stimulus intensities much above threshold.

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155 Tracer Studies of Microvillar Cells Suggest A Second Morphologically Distinct Class of Sensory Neuron Exists in Mammalian Olfactory Epithelia. J. CARTER ROWLEY III, DAVID T. MORAN, AND BRUCE W. JAFEK (University of Colorado School of Medicine).

The olfactory epithelia of a number of mammalian species contain large, electron-lucent cells equipped with a small tuft of short microvilli at their apical pole. microvillar cells, which bear close resemblance to "brush cells" described elsewhere in the respiratory tract, do not resemble the microvillar olfactory receptors described in fish, amphibians, and vertebrate vomeronasal organs. They do, however, have a fine structure similar to that of a variety of bipolar sensory neurons described in other sensory systems. The present study was initiated to answer the question: do the microvillar cells in the mammalian olfactory epithelium have axons that pass through the olfactory nerve? Adult rats of both sexes were placed under general anaesthesia, a small incision was made in the skull, and a 2% aqueous solution of horseradish proxidese (HRP) was injected into the olfactory bulb of the brain. The incision was sealed with bone wax. 24 hours following HRP injection, the animals were re-anesthetized and perfused with fixative. The olfactory epithelium was dissected free, incubated for 18 hours in 0.1% 3,3' diaminobenzidine (DAB), reacted for 3 hours with DAB in the presence of hydrogen peroxide, post-fixed with osmium tetroxide, and prepared for transmission electron microscopy. Examination of sections revealed that two types of cells were backfilled with HRP: ciliated olfactory receptors and microvillar cells. The observation that microvillar cells become backfilled with HRP following injection of that tracer into the olfactory bulb strongly suggests they have axons that project to the olfactory bulb through the olfactory nerve. This finding is consistent with the hypothesis that microvillar cells represent a second morphologically distinct class of sensory neuron in the rat olfactory epithelium. (Supported by NIH Program Project Grant NS20486).

NOTES

156 Tight-Junctions in Developing Rat Olfactory Epithelia.
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At E14 the olfactory epithelium of rat embryos began to differentiate. Early E14 embryos had an undifferentiated epithelium depicting tight-junctional strands in regions only where three cells joined (tricellular tight-junctions). At that stage strands could not be seen in regions where two cells met (bicellular tight-junctions) but fracture plane irregularities which resemble outlines of strands-to-beformed were present. Areas which will eventually give rise to the bicellular strands displayed lower densities of intramembranous particles than areas lateral and apical from those, suggesting that tight-junctions are able to separate membrane domains in the absence of strands. The transformation of the above tight-junctional apppearance into the familiar one with bicellular as well as tricellular strands took place within one developmental day-E14-and accompanied the transformation of an undifferentiated into a differentiate ing olfactory epithelium surface. The following observations were made on differentiating epithelium surfaces. Numbers of strands tended to be lowest in E14, and did not differ for other age groups investigated (E16, E17, E18, E19 and adult). This is the case for sensory and supporting cells. Throughout development supporting cell-supporting cell junctions had lower numbers of strands than supporting cell-sensory cell and sensory cell-sensory cell junctions. Strand densities, i.e., the number of strands per um reflecting the packing of the strands, tended to increase in a vertical plane (strands par allel to the epithelium surface) and increase and then decrease in a horizontal plane as a function of development. This too is the case for sensory as well as for supporting cells. In conclusion: It seems that during development the packing rather than the number of strands of tight-junctions changes.

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157 Sustentacular Cell Ultrastructure: Evidence for Secretion and Absorption in the Salamander B. ZIELINSKI, M.L. GETCHELL AND T.V. GETCHELL (Dept. of Anatomy and Cell Biology, Wayne State University School of Medicine, Detroit, MI 48201)

Previous light microscopic studies have shown that sustentacular cells consist of an elongate cell body, a central stalk and its basilar expansion (BE) located at the junction of the epithelium with the lamina propria (LP) (Rafols and Getchell, Anat. Rec., 206:86-101, 1983). Transmission electron microscopic examination demonstrated distinct characteristics for these regions. The supranuclear region contained vesicles enclosing secretory material, endoplasmic reticulum, mitochondria and a Golgi complex. Conventional exocytosis from the apical surface was observed. The stalk passed through layers of olfactory receptor cell nuclei and basal cells. It contained mitochondria, intermediate-sized filaments, membranous whorls and multi-vesicular bodies. Fine granular extracellular material was located intercellularly. Stalks interdigitated suprajacent to the basement membrane in areas in apposition to blood vessels in the LP. The BE had lateral folds that formed a complex network of extracellular channels and surrounded receptor axons. interfaced with the basement membrane that consisted of laminae lucida, densa and fibroreticularis. The fine granular material of the lamina densa was present between BE. Basal surfaces adjacent to blood vessels and Bowman's glands of the LP were folded with the basement membrane closely following the contours. BE contained mitochondria, intermediate-sized filaments and micropinocytotic vesicles. The results indicate that: 1. the apical region of the cell body synthesizes and secretes material into the mucociliary complex; 2. intercellular spaces contain material resembling the basement membrane and 3. the BE has characteristics of transporting cells that include basolsteral membrane folds and abundant mitochondria.

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Localization of NGF-like, VIP-like and Substance P-like Immunoreactivity in the Olfactory Mucosae of the Salamander, Bullfrog and Grass Frog. MARILYN L. GETCHELL, THOMAS E. FINGER and THOMAS V. GETCHELL (Department of Anatomy & Cell Biology, Wayne State University School of Medicine, Detroit, MI 48201 and Department of Cellular & Structural Biology, University of Colorado Medical School, Denver, CO 80262, USA

NGF stimulates and directs the growth of sensory and sympathetic axons. The neuropeptides VIP and substance P (SP) occur and play modulatory roles in nasal mucosa [DeLong and TV Getchell (1987) Chem. Senses 12]. Initial immunohistochemical experiments were performed to detect and localize these antigens in olfactory mucosae. Standard procedures for 2-step immunofluorescence were followed. For 3-step fluorescence procedures with biotinylated secondary antibody and FITC-labeled avidin, sections were pretreated with avidin and biotin to eliminate non-specific binding. Control sections, treated with non-immune serum, exhibited no specific immunoresctivity (ir). NGF-like ir was localized in ducts and acinar cells of Bowman's glands (BG) near the basement membrane, and in cells in the basal olfactory epithelium (OE). VIP-like ir was observed in fibers extending to the OE surface, around BG and blood vessels and in the lamina propria (LP). SP-like ir was found in varicose fibers extending to the OE surface, around BG acini in the LP and intraepithelial blood vessels in immature OE, and in fibers in the LP. SP-ir fibers reaching the epithelial surface were far more numerous in frogs than salamander. Neither peptide was observed in cross-sections of bundles of olfactory axons. These results indicate that NGF is present in the region of the OE where proliferative activity of stem cells of olfactory neurons has been demonstrated, and suggests that the OE and BG receive "extrinsic" innervation by fibers containing VIP-like and SP-like neuropeptides.

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Morphology of Chorda Tympani Fiber Receptive Fields and Proposed Neural Rearrangements during Development. C. M. MISTRETTA (Dentistry and Nursing, Univ. of Michigan, Ann Arbor), S. GURKAN and R. M. BRADLEY (Dentistry, Univ. of Michigan)

Average number of fungiform papillae in receptive fields of single chorda tympani fibers decreases during development in sheep, and an increasing proportion of small receptive fields that are highly responsive to NaCl is acquired. To learn whether there are developmental differences in number of taste buds per receptive field, we examined 58 histologically reconstructed fields that had been mapped electrophysiologically in fetal, perinatal and postnatal sheep. We also counted fibers in the chorda tympani nerve. There is a significant, developmental increase and subsequent decrease in number of taste buds in receptive fields (means: fetal = 46 taste buds, perinatal = 93, lamb = 57). These differences cannot be attributed to alterations in numbers of fungiform papillae, because total number of papillae on the tongue remains constant. Average number of taste buds per papilla, however, increases and then decreases, and the increase in perinatal animals is accompanied by the appearance of large, multipored taste buds. Number of chorda tympani nerve fibers also apparently increases up to perinatal stages and then decreases postnatally. We propose that in perinatal animals there is a period of perinatal animals there is a period of hyperinnervation of existing receptive fields by new chorda fibers or branches that induce de novo taste bud formation and/or division of existing buds. Later in development, we suggest that there is an elimination of some innervation. Taste are not static during rather undergo substantial receptive fields development. but changes in morphology and neural organization. (Supported by N.S.F. Grant BNS 8311497.)

Some Effects upon Fungiform and Foliate Taste Buds of Condensing the Innervation by the Chorda Tympani nerve.

D.R. RIDDLE and B. OAKLEY (University of Michigan, Ann Arbor, MI 48109).

The chorda tympani (CT) fibers in the mongolian gerbil, Meriones unguiculatus, can be condensed by suturing the parent proximal stump of the chorda-lingual nerve to the most posterior of its three distal branches near the tongue. The higher initial density of regenerated CT taste axons in the middle and posterior part of the fungiform taste bud field did not result in new fungiform taste buds. More than two thirds of the fungiform taste buds were absent from the tip of the tongue. This indicates that taste axons were constrained by their route of entry into the tongue and did not spread to the tip of the tongue to reinnervate their original sites. Each foliate papilla in a normal animal contains an average of 133±21 (N=12) taste buds in 6-12 slits (9.2±1.1) located on the lateral margins of the tongue. Adult foliate taste buds are maintained by axons of the CT and glossopharyngeal (IX) nerves. The IX nerve alone will maintain all foliate taste buds (136±27, N=4), while the CT supports 26±12 (N=4). With the IX nerve present, the regenerated CT fibers caused 167±22 (N=12) foliate taste buds to form. This is a significant increase of about 30 foliate taste buds above normal (p.005). When the IX nerve was removed several months after the initial surgery, only 19±7 (N=3) taste buds continued to be supported by the condensed CT nerve. The IX nerve appears to contribute to the maintenance of the 30+ additional taste buds, because in its absence the condensed CT maintains no more taste buds than a normal CT nerve (19±7 vs. 26±12). This is the first demonstration that adult taste papillae retain a latent morphogenetic capacity to respond to hyperinnervation with an increased number of taste buds.

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161 Basal Cells and Synaptic Connectivity in Taste Buds of Necturus R. J. DELAY and S. D. ROPER (Department of Anatomy. Colorado State University, Ft. Collins, CO 80523 and Rocky Mountain Taste and Smell Center, University of Colorado Health Science Center, Denver, CO 80262)

Taste buds in the mudpuppy, Necturus maculosus were examined with electron microscopy. Dark and light taste cells extend from the base of the taste bud to the apical surface (taste pore). Located at the base of the bud is a third cell type, the basal cell. The nucleus comprises most of the basal cell. This cell does not extend to the apical surface. Small spine-like processes project from basal cells into the surrounding cytoplasm of adjacent dark and light taste cells. The spine-like processes are packed with actin-like filaments and are approximately 0.25 µm in diameter and 2-3 µm in length. The cytoplasm of basal cells contain numerous vesicles, both clear and dense-cored. Morphologically, basal cells appear quite similar to Merkel cells, a type of mechanoreceptor found in the skin of many species, including amphibians.

Unmyelinated nerve processes enter the taste bud at the base and course mainly through the lower third of the bud, entwining with basal cells and processes of dark and light taste cells. Synapses were observed between taste cells (including basal cells) and nerve processes and between taste cells themselves. The majority (~70%) of the synapses found in the mudpuppy taste bud involve the basal cell. These observations suggest that some degree of signal processing or integration involving basal cells may occur in taste buds, as suggested by Reutter in fish (I.S.O.T. IX, in press). Alternately, basal cells might serve some kind of mechanoreceptor function comparable to Merkel cells.

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NOTES

Synabsin-like Imminoreactivity of Nerve Fibers in and around Lingual Taste Buds of the Rat. THOMAS E. FINGER, MARY WOMBLE (Univ. of Colorado Med. Sch.), VAR L. ST. JEOR, JOHN C. KINNAMON (Univ. of Colorado, Boulder), AND TETSUFUMI UEDA (Univ. of Michigan).

While attempting to locate synaptic foci in taste buds, we found that synapsin antibodies appear to react with nerve fibers associated with taste buds. Rats were perfusion-fixed with a mixed aldehyde solution. Sections were treated with 1% normal goat serum, then exposed to a 1:300 dilution of polyclonal synapsin antiserum. Control sections were incubated under similar conditions, except normal rabbit serum was substituted for the synapsin antiserum. The sections then were prepared according to standard avidin-biotin-peroxidase methods. The sections were flat embedded in plastic and examined with a light microscope. Appropriate areas were removed and sliced for high voltage or conventional electron microscopy. Control sections exhibited no specific immunoreactivity.

Sections exposed to synapsin antisera revealed dense immunoreactivity (IR) of relatively coarse nerve fibers (approximately 1-2 µm in diameter) entering the fungiform and circumvallate taste buds. Finer (<1 µm) varicose IR fibers were also found between taste buds in the lingual epithelium. The intragemmal fibers entered the base of taste buds to ramify profusely within each bud. Most fibers within the taste buds reached only halfway up in the bud, to about 30 µm below the epithelial surface, however, some IR fibers terminated within 10 µm of the surface. Ultrastructural analysis revealed that IR material was distributed throughout the cytoplasm but was especially densely associated with filamentous organelles tentatively identified as microtubules. HVEM has shown that at least some of the IR nerve processes are postsynaptic to the taste receptor cells in the taste bud. Thus, intragemmal sensory fibers are IR for synapsin. Synapsin-like IR is not, however, limited to intragemmal fibers. Fine, perigemmal fibers also appear to be IR to synapsin antisera.

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163 Persistence of Taste Buds in Denervated WHITEHEAD (Dept. Oral Biology, Ohio State Univ., Columbus, OH 43210), MARION E. FRANK, THOMAS P. HETTINGER, LEIN-TUAN HOU and HYUN-DUCK NAH (Dept. Biostructure and Function, Univ. of Conn., Farmington, CT 06032)*

For more than a century, taste buds have been reported to disappear entirely from adult mammals after denervation. In contrast, other sensory endorgans (e.g. Pacinian corpuscles, Merkel cells) persist in an atrophic state after denervation. Electron microscopy was utilized to evaluate the long term fate of fungiform buds in the hamster and to reevaluate whether they are unique in requiring innervation for their existence. Electrophysiological recording from nerve sheaths distal to sites of nerve resection on the experimental side, and from the contralateral nerves, evaluated possible reinnervation. Taste buds persisted in an atrophic state for as long as days after chorda tympani denervation or 50 days after combined chorda-lingual resection. These buds were smaller, contained fewer cells than normal, and exhibited decreased staining of the taste pore region. However, the cells of denervated buds were elongated apically, had ultrastructural features of light and dark cells, and were readily distinguished from non-taste, keratinized, epithelial cells. Thus, although taste bud structure depends on innervation there is no absolute neural requirement for taste bud survival. Buds with normal structures and functions reappear rapidly upon reinnervation (Cheal and Oakley, 1977), and buds with crossed reinnervation impart their sensibilities to the foreign nerve (Nejad and Beidler, 1987). These findings and ours suggest that buds do not regenerate de novo, but derive from specialized cells that survive denervation.

*Supported by NIH Grant NS16993.

Polypeptide (VIP) in Hamster Taste Cells. M. SCOTT HERNESS (The Rockefeller University, New York, NY 10021)

Vasoactive intestinal polypeptide (VIP) is a 28 amino acid residual peptide orginally isolated from porcine duodenum in 1970 which has potent hypotensive, vasodilatory, as well as secretory properties. Distribution of VIP has been extended from gut to include brain, peripheral nerves, and nasal mucosa. Using an immunocytochemical (ICC) procedure, evidence is presented here that VIP is also contained within mammalian taste cells.

The posterior region of Bouin's fixed paraffin-embedded hamster tongue was sectioned at 8 μm and processed for immunocytochemical localization of VIP. The sections were deparaffinized, rehydrated, and incubated in primary rabbit antiserum to VIP (A-VIP; Cambridge Research Laboratories) at concentrations of 1:1000, 1:2000, and 1:5000 in a closed moist chamber for 72 hours at $^{1/2}$ C. Biotinylated secondary antibody and an avidin-biotin-horseradish peroxidase complex (Vectastain "ABC" technique; Vector Laboratories) were applied, followed by reaction in 3,3'-diaminobenzidine tetrahydrochloride (DAB; Sigma) with H₂O₂. Positive immunoreactivity was seen as dark brown granular reaction product.

Reaction product to VIP antiserum was observed in taste buds of the foliate papilla at all antiserum dilutions. Surrounding epithelium was not stained but some basal cells at the epidermal - dermal interface appeared to be labeled. Some but not all taste buds in fungiform papilla showed positive immunoreactivity. Reaction product was localized in the cytoplasm and the nuclei were clear. It did not appear to be localized to any polar region of the cell, as for example adenylate cyclase and phosphodiesterase have been localized to the apical region only. Possible localization within circumvallate papillae is in progress. In addition to taste buds, reaction product was observed surrounding blood vessels within the tongue musculature and occasional cells within filiform papilla.

This work was supported by BRSG S07 RR07065 awarded by NIH.

165 Gustatory information is processed in multiple facio-spinal pathways in the catfish brainstem. JAGMEET S. KANWAL and THOMAS E. FINGER (Univ. of Colorado Medical School).

Facial gustatory information is the primary determinant of food search and pick up in the channel catfish, Ictalurus punctatus. Details of the neural circuitry and mechanisms involved in gustatory mediated motor output are, however, unknown.

In catfish, swimming and reflex turns associated with food search and pick up are coordinated by the spinal cord. In order to delineate the pathways by which gustatory input can reach this final common motor system, we injected horseradish peroxidase (HRP Type VI) into the spinal cord and facial lobe (FL) of the channel catfish. Spinal injections of HRP retrogradely labeled clusters of medium sized (20 um) neurons in the ventromedial reticular formation (RF) as well as somatotopically arranged giant (60-100um) neurons in the lateral lobule of the facial lobe, i.e. that portion which receives gustatory input from the flank region. Injections in the lateral lobule of the FL retrogradely labeled bipolar neurons in the dorsomedial region of the medullary RF; a few axonal projections to the ventromedial region of the RF were also observed. In contrast, injections in the medial lobule of the FL (which receives gustatory input from the head region) revealed dense axonal projections to ventromedial portions of the RF. In double-label studies, labeled axons from the medial lobule were seen to contact cell bodies of RF neurons that project to the spinal cord. Electrophysiological studies of these reticular neurons reveal large gusto-tactual receptive fields in contrast to the discrete receptive fields of neurons in the FL. Thus, gustatory information from both the medial and lateral lobules of the FL converges onto spinally projecting neurons of the medial RF, while giant neurons in the lateral lobule project directly to the spinal cord.

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Tyrosine Hydroxylase-Like Immunoreactivity in the Gustatory Zone of the Nucleus of the Solitary Tract in the Hamster. BARRY J. DAVIS and SUSAN MOORE (University of Alabama at Birmingham)

Our original groupings of the morphologically distinct neurons within the gustatory zone of the nucleus of the solitary tract (NST) could not accommodate the variety of neuronal types encountered in Golgi impregnated material. We embarked on a series of immunocytochemical studies in an attempt to associate specific chemical markers with certain classes of NST neurons so that our grouping could be more comprehensive and include functional attributes. TH-like immunoreactivity somata were observed throughout the gustatory NST. At the light microscopic level, these somata appeared Golgi-like and primary dendrites of varying orientations were clearly visible. The features of these immunoreactive neurons were quantified at the EM level. The range of somal areas for neurons having invaginated nuclei was $53-336~\mathrm{um}^2$ and the mean somal area was $124~\mathrm{um}^2$ (8.6 x 15.4 um). These values agree with our previous EM morphometric studies and indicate that all members of the so-called X1 and X3 classes show TH-like immunoreactivity. Neurons possessing non-invaginated nuclei had a range of somal areas of $51-166~\rm um^2$ and their mean somal area was $102~\rm um^2$ (9.2 x 13.9 um). Again, these somal areas span the range of sizes for members of the X2 and X4 classes. We conclude that a variety of morphologically distinct NST neurons express TH-like immunoreactivity. Although the gustatory NST appeared heavily immunoreactive at the light microscopic level, at the EM level only about 10% (N=1388) of the neurons encountered were immunoreactive. Therefore, many other NST neurons remain to be associated with some specific chemical marker. Modest DBH-like and no PMNT-like immuoreactivity was seen in the gustatory NST and our tentative conclusion is that many (but not all) of neurons that demonstrate TH-like immunoreactivity are dopaminergic.

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167 Neonatal Olfactory Deprivation Results in a Dramatic Reduction of Tyrosine Hydroxylase Levels in Adult Rat Main Olfactory Bulb. HARRIET BAKER (Cornell University Med. Coll.)

Peripheral afferent denervation of the main olfactory bulb produced either chemically or surgically results in large decreases in all parameters reflecting the dopaminergic phenotype of intrinsic juxtaglomerular neurons. The role of afferent neuronal activity in maintenance of phenotype is unknown. Unilateral neonatal cauterization of the nares prevents odorant access to the receptor epithelium and presumably afferent neuronal stimulation. These experiments sought to determine if lack of neuronal stimulation during postnatal development of juxtaglomerular cells also produced a reduction in the levels of tyrosine hydroxylase (TH), the rate-limiting enzyme in dopamine biosynthesis. Unilateral nares closure (right side) was produced by cauterization in 3 day-old rat pups. TH immunoreactivity and activity were assessed 1-2 months post-lesion. TH immunoreactivity was reduced markedly in the main olfactory bulb ipsilateral to the lesion. In the contralateral bulb, TH-staining was similar to that observed in control rats. In contrast to animals with surgical or chemical deafferentation, the glomeruli retained a relatively normal organization. Staining for olfactory marker protein demonstrated that olfactory afferent fibers were present indicating that receptor afferents did not degenerate. TH activity was measured in both control and deprived rats to quantitate the changes in dopaminergic function. In control animals (n = 12) right and left olfactory bulbs displayed similar TH activity (right, 3.74 ± 0.34; left, 4.34 ± 0.35 nmoles dopa/bulb/15 min.). Olfactory deprivation (n = 6) produced a dramatic decrease in TH activity in the ipsilateral olfactory bulb (right, 0.79 ± 0.18 ; left, 2.96 ± 0.50). Interestingly, the bulb contralateral to the nares closure had significantly less TH activity than the olfactory bulbs of control animals. These experiments demonstrate that odor deprivation produces decreases in TH activity and immunoreactivity similar to those observed following direct peripheral afferent denervation. These data suggest that afferent neuronal activity is necessary to maintain the dopaminergic phenotype either directly or through the release of trophic agents linked to synaptic transmission.

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NOTES

168 Immunofluorescence Studies of Embryonic Development of the Rat Olfactory Bulb.

V.McM.CARR, A. I. FARBMAN (Dept. Neurobiology & Physiology, Northwestern Uni., Evanston, IL), and J. I. MORGAN (Roche Institute of Molecular Biology, Nutley, NJ)

We have previously reported (Chem Senses 11:596, '86) studies using Mab's to study the devopmental expression of molecules in the rat olfactory epithelium. The present report is an extension of those studies using the same Mab's to examine development of the rat olfactory bulb. Three Mab's react with membrane molecules in axons, dendrites, and cell bodies (Neu 4, Neu 5, Neu 9) of olfactory receptor cells and with supporting cells (Neu 9). One reacts most intensely with components of the luminal edge of the epithelium (1 A-6).

Bulbar reactivity to all four Mab's is first apparent at E14 and is both pericellular and fibrous in nature. All four also bind pericellularly to as yet unidentified cells in the olfactory nerve and ONL. Through E17 fluorescent patterns show little OB organization beyond a subdivision into a radially organized inner region and an outer more randomly organized region. These fluorescent patterns change dramatically at E18, the time when initial receptor cell synapses are made in the OB (Farbman, unpub.). At E18 the EPL, MCL, GCL, and ONL are reactive with all four Mab's. All but the MCL show pericellular reactivity. With continued development the GL becomes distinguishable, especally with Neu 4 and Neu 9. The IPL appears by E22 as a region of bright amorphous fluorescence with all Mab's but Neu 5. With each Mab the MCL init-ally appears as a region of radial fluorescent fibers; the mitral cells themselves show no reactivity. By E22, however, some reactive cells can be seen. Neu 4 and Neu 9 also react with mitral cell fibers directed more superficially and deeply. Supported by NIH grant NS 23348.

169 Postnatal Development of Noradrenergic Afferents to the Olfactory Bulb. J.H. MCLEAN and M.T. SHIPLEY (Univ. of Cincinnati).

Noradrenergic (NA) fibers innervate most forebrain areas early in development. This pattern of development has led to the suggestion that NA fibers have a trophic role or modulatory action on developing circuits. The olfactory bulb is an excellent model for the ontogeny of cortical circuits and neurotransmitters because of its relative simplicity. Here, we have examined the developmental expression of NA fibers in the main (MOB) and accessory olfactory bulb (AOB) of rats and compared this system to the development of another monoamine in the bulb, serotonin (5-HT).

Sections of the bulb at several postnatal ages were immunocytochemically stained to demonstrate the presence of NA by using an antibody to dopamine-B-hydroxylase (DBH). There is a gradual and steady increase in the density of NA fibers in the MOB during ontogeny. The laminar pattern appears to develop at a uniform rate in all innervated layers. The fibers are most prevalent in the granule and internal plexiform layers and are evenly, although less densely, distributed in the external plexiform layer. These results are comparable to the developmental pattern of 5-HT immunoreactive fibers in MOB (McLean and Shipley, in press) which seem to have a similar developmental time course. A distinct difference between 5-HT and NA innervation of MOB is that 5-HT fibers heavily innervate glomeruli, while at all stages of development NA fibers sparsely innervate glomeruli or avoid them altogether. NA fibers heavily innervate the AOB by postnatal day one while serotonergic innervation is sparse. Neither neurotransmitter system innervates the AOB glomeruli at any developmental stage.

These results show that during early postnatal development, NA fibers proliferate steadily in the infraglomerular layers of both MOB and AOB. Neuron proliferation, migration, differentiation, and/or synaptogenesis occur much later in the olfactory bulb than in the neocortex. Thus, although most NA fibers appear to enter the MOB and AOB postnatally, they could still influence major developmental events in these cortical structures.

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170 Unilateral Odor Deprivation: Changes in Olfactory Bulb Cell Number and Density. L. L. FRAZIER and P. C. BRUNJES (University of Virginia)

Unilateral external naris closure on postnatal Day 1 results in a 25% reduction in the volume of the olfactory bulb 30 days later (Brain Res. Rev., 11, 1986, 1). The smaller size of deprived bulbs may result from changes in either the number of neurons and/or glia, their size, or both. In order to determine how experience affects bulb development, changes in cell density and number were quantified at a number of early postnatal ages in both deprived and sham-operated control animals. Rat pups underwent either naris closure or sham surgery on postnatal Day 1 and were reared to Days 2, 5, 10, 20 or 30. Bulbs were removed and embedded in glycol methacrylate for thin sectioning. Cell densities were determined for all cell types and all laminae. Cell density was translated into cell number per bulb using estimates of laminar volumes made from frozen sections. Growth curves were the same in deprived and control conditions until Day 20, when deprivation resulted in significant decreases in cell density and number in the internal plexiform and granule cell layers; and in light and dark granule cells and glia. Tufted and mitral cell nuclear area also exhibited a significant decrease after Day 20. These results suggest that deprivation may alter relay neuron function, which in turn may affect the survival of the granule cells which innervate them. Most of the observed changes did not occur until after Day 20, suggesting they represent a culmination of a series of experience-induced changes within the maturing bulb. We are presently investigating how changes in density and number are occurring. It is possible that changes in cell number are the result of altered proliferation or cell death patterns. In order to assess the first option, both occluded and sham-operated pups were injected with tritiated thymidine at the ages noted above and sacrificed 24 hr later. Bulbs were removed and processed by standard autoradiographic techniques. No differences in numbers of labelled subependymal cells have been encountered at Days 2, 5, or 10, suggesting no early differences in proliferation rates.

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171 <u>Unilateral Odor Deprivation and Olfactory Bulb Development in the Precocial Mouse Acomys cahirinus.</u>
P. C. BRUNJES (University of Virginia).

Single naris closure on Day 1 results in a 25% decrease in the size of the ipsilateral olfactory bulb when laboratory rat or mouse pups are examined 30 days later. Deprivation appears to have its greatest effects during periods of rapid bulb maturation: rats occluded on Day 10 and reared to Day 40 also reveal a 25% decrease in bulb size while those occluded from Day 20-50 exhibit very little change (Brain Res. Bull. 14, 1985, 233). This hypothesis was examined by testing the effects of deprivation on a species exhibiting truncated early bulb growth. Acomys cahirinus (the "spiny mouse") is born fully furred and with coordinated motor capabilities after 38 days of gestation. Rapid postnatal bulb growth occurs, with 90% of adult size reached by Day 10 (Dev. Brain Res. 8, 1983, 335). If plasticity is limited to accelerated periods of growth, Acomys should exhibit truncated lability. However, deprivation in Acomys actually results in more drastic consequences than in either the lab rat or mouse. Occlusion on postnatal Days 1, 5 or 10 followed by a 30 day survival period resulted in a 40% decrease in the size of the ipsilateral bulb. Deprivation from Days 30-60, which has no effect in the rat, resulted in a 20% decrease in bulb size in Acomys, indicating a much longer period of susceptibility to occlusion. A 6% decrease in receptor numbers was found in Acomys deprived from Days 1-30 (with no effect on basal or supporting cells), while rats occluded at the same time exhibit a 10-12% decline. Thus, the large changes seen in the Acomys bulb do not result from gross changes within the mucosa. The results suggest a complicated relationship between growth and plasticity, and that state at birth is perhaps not a reliable indicator of early malleability.

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172 Changes in Olfactory Sensitivity to Androstenone with Age Experience. C.J. WYSOCKI, G.K. BEAUCHAMP, H.J. SCHMIDT, and K.M. DORRIES (Monell Chemical Senses Center)

Approximately 50% of normal human adults have a specific anosmia to the odor of androstenone, a naturally occuring odorous steroid, found in perspiration, pork products and some vegetables. Previous research has shown that variation in sensitivity to this compound has a significant genetic component. However, this genetic trait appears not to be immutable.

In a study of 215 subjects, age 9 to 50, we observed that, while androstenone thresholds did not differ across age for females, males appeared to show considerable variation with age. To simplify the data analysis, individuals were divided into three age groups on a post-hoc basis. Under these conditions, males in the youngest age group had thresholds significantly lower than the two older groups. Furthermore, the percentage of males who were anosmic to the odor, reporting no smell at the highest concentrations, increased significantly with increasing age over the three groups. Females showed no difference in androstenone detection over the three age groups. We found no significant effects for age or sex in the same subjects tested with pyridine.

Changes in androstenone thresholds within subjects were demonstrated in a second study. Androstenone-anosmic male subjects were exposed to the compound daily over a 6 week period and thresholds were determined weekly. In 8 of 13 subjects tested to date, we observed the emergence of a clear, persistent detection threshold. Subjects also began to describe the odor, using characteristic terms. For 3 subjects we saw no change; they continued to be anosmic. The remaining 2 subjects detected the odor in some sessions, but not consistently thoughout the study. Thresholds for amyl acetate, to which subjects were also exposed, remained stable.

These studies suggest that while the ability to perceive the odor of androstenone is determined in part by genetic mechanisms. sensitivity to it may change with age and experience.

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Monoaminergic Activity in Olfactory Bulb and Cerebellum During Olfactory and Motor Learning Tasks. R.G. MAIR, C. SLADE (University of New Hampshire), and P.J. LANGLAIS (Harvard Med).

We studied the turnover of catecholamines (CA) in olfactory bulb and cerebellum of 48 9 week old Long-Evans rats by inhibiting tyrosine hydroxylase (TOH) activity (with a-methyl-ptyrosine [AMPT]) for 60 minutes and measuring concentrations of norepinephrine (NE) and dopamine (DA). The turnover of serotonin (5-HT) was also measured by determining the concentration of 5-HT and its metabolite 5-HIAA. Twelve rats (group 0) were trained to dig up scented sucrose pellets in a bed of pine shavings during 6 30 minute sessions. Twelve (group M) were trained in 6 30 minute sessions to remove a ball from a tube to retrieve an equivalent amount of odorless sucrose. Twenty-four (groups C1 and C2) were fed an equivalent amount of odorless sucrose in their home cages in 6 daily sessions. Thirty minutes prior to their sixth training sessions, animals received 1 ml i.p. injections of AMPT (200 mg/ Kg) (groups O, M, C2) or isotonic saline (group C1). After training (60 minutes after injections) animals were sacrificed by decapitation and their brains removed and frozen immediately at -80 C.

AMPT treatment did not suppress the rate of behavioral responding. Chemical analyses demonstrated increased turnover of NE (but not DA) in groups O and M. NE turnover was slightly higher in group O than M in both olfactory bulb and cerebellum, thus there was no evidence of behavioral-anatomic specificity. In olfactory bulb, NE turnover in group O was estimated as 35 femtograms/ ul tissue/ second, a rate comparable to rates of ionophoretic release of NE reported to suppress mitral cell activity. The concentration of 5-HIAA (but not 5-HT) was reduced within the olfactory bulb of group O, and thus there was evidence that 5-HT activity may be decreased selectively within the bulb during this task.

NOTES

The Bitter-Sweet Taste of Alcohol: Aversion 174 Generalization to Various Sugar-Quinine Mixtures in the Rat. STEPHEN W. KIEFER, GLORIA J. LAWRENCE, KARRY L. SCHWEIGER (Kansas State University)*

Rats generalize alcohol aversions to specific combinations of tastants: sucrose + quinine and-sucrose + acid solutions (DiLorenzo et al., 1986, Alcohol). Presently, two experiments were conducted to examine the "sweet" taste quality of alcohol in the rat. Rats were trained to avoid a 5% ethanol solution and then tested for aversion generalization. Four test solutions were combinations of .0001 M quinine hydrochloride and each of the following: .1 M sucrose, .75 M glucose, .3 M fructose, and .0001 M sac-charin (Experiment 1). The same saccharide solutions were combined with .01 M hydrochloric acid and used as test solutions in Experiment 2. Results showed that rats exhibited significant generalization to the sweet-bitter solutions in Experiment 1; trained rats consumed significantly less quinine plus sucrose, glucose, and fructose, than the untrained rats, p<.05; the saccharin + quinine approached significance, p<.10. In Experiment 2, no significant generalization to any of the sweet + sour solutions was found. The results suggest that alcohol does have a general sweet (and bitter) quality, one that is not specific to sucrose.

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Preference for NaCl Solutions in an Experimental Model of Liver Disease: The Bile Duct Ligated Rat.

RHONDA OETTING DEEMS and MARK I. FRIEDMAN (Monell Chemical Senses Center, Phila., PA).

Recent behavioral and physiological evidence suggests a role for the liver in salt preference and appetite. Patients with liver disease have been reported to have increased detection and recognition thresholds for NaCl which, in acute liver disease, improve with recovery. To more fully investigate the taste for salt in liver disease an animal model was used to assess changes in taste preference for NaCl with development of liver pathology. Acute bile duct ligation in rats produced by surgically ligating and severing the common bile duct, produces pathology which is comparable to human liver diseases. Taste preferences for NaCl were assessed by 24 hr two-bottle (NaCl and distilled H2O) and 20 min single bottle taste tests in both bile duct ligated (BDL) and sham animals. In Group 1, beginning 2 weeks post-ligation, animals were presented with ascending concentrations of NaCl (6.13, 25, 100, 200, 300 & 400 mM) on consecutive days. Preferences for NaCl were determined by amount of NaCl consumed divided by total intake. In Group animals were tested with short term 20 min single bottle taste tests. Group 3 was continually presented with a concentration of 200 mM NaCl for 2 weeks pre-ligation and 3 weeks post-ligation. Daily intake was monitored and preferences for NaCl were determined. Statistical analyses using 2-way ANOVAs indicated that BDL rats have higher preferences for NaCl than sham controls. In Group 1, BDL preferences for 200 and 300 mM NaCl were significantly greater than controls (F=2.97, p<.05). Likewise for Group2, intake (ml/g body weight) of BDL rats was greater at 100, 200 and 300 mM concentrations (F=5.92, p<.05). For Group 3, BDL rats showed significant absolute changes from baseline as compared to sham animals (F=2.13, p<.01). The data indicate that the BDL rat provides a useful model to study changes in taste preferences in experimental liver disease. Additionally, the results emphasize the involvement of the liver in modulating the preference for salt.

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Anterior vs. Posterior Tonque: Teste and Hedonic Responses Differ.
ROBERT J. HYDE (San Jose State University, San Jose, CA 95192).

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Sandick and Cardello (1981) have shown that the sweetness of sucrose is judged more intense from circumvallate papillae than from anterior tongue; however, studies relating tongue loci and hedonic responses seem to be lacking. Presently, 48 college women judged teste intensities and degree of liking on 100 mm lines (labeled "No Taste" or "Dislike Extremely" at one end and "Extremely Intense" or "Like Extremely" at the other) for anterior compared to posterior tongue. For chocolate milk (CM), lemonade (L), and Pepsi (P), all at room temperature (21°C), subjects responded under the following conditions: (a) while immersing the front of the tongue into 10-ml of testent in a 30-ml cup and (b) while swallowing. For a portion of vanilla ice cream (1) served on a small plastic spoon (-4 °C) and for a bolus of bittersweet chocolate (C), made from rapidly chewing a small square 25 times, subjects responded while immersing the tip of the tongue into the bolus held in the front of the mouth and again while swallowing. Subjects rinsed orally with water between stimuli. Judgements of sweetness and degree of liking for posterior tongue (and posterior oral cavity) significantly exceeded those for enterior tongue (matched /-test and ANOYA, p<0.001 for all stimuli except CM, p<0.01). For both loci, hedonic responses correlated significantly with sweetness (anterior: r = 0.911, 3 df, p<0.05; posterior: excluding P, r = 0.999, 2 df, p<0.001). Also for posterior compared to anterior: CM (stronger chocolate flavor, p<0.001), L (less salty, p<0.05; equal sourness), P (less stinging sensation, p<0.001), I (stronger vanilla flavor, p<0.001), and C (less bitterness, n.s.). These results agree with Sandick and Cardello (1981) for sweet and salty. Without mixture suppression from sweetness, sour and bitter might also have been judged more intense from posterior tongue, as reported earlier. By transmitting perceived sensations at different intensities, the tongue loci seem to be uniquely organized for enhancing any novel properties of gustatory stimuli. As such, the tongue might possess its own arousal and reward system; anterior tongue relays strong sensations of stinging, bitterness, and sourness into the CNS, whereas posterior tongue and oral cavity convey stronger sensations of sweetness, flavor, and pleasure.

177 Multidisciplinary Measures of Taste
Hedonics. J.E.STEINER & A.STEINBERGER.; Department
of Oral Biology, "HADASSAH" Faculty of Dental
Med.; The Hebrew University, Jerusalem
(Israel).P.O.B.1172 *)

hedonics may induce EEG-arousal Taste negonites heart-rate(HR) changes, both in man heart-rate(HR) For humans the physiological with measures may be expected to hedonic estimates. 40 yo correlate hedonic estimates. 40 young healthy male volunteers were therefore exposed to randomized intraoral stimuli. These were: tap-water; a clear vegetable broth; the broth seasoned with 2 concentrations of MSG (0.5;0.75%); ageuous solutions of sucrose (12%) and of citric acid (0.5%). Stimuli were infused from syringes via tubings the mouth. EEG and ECG were recorded at rest, during stimulation and for follow up. 90 swallowing, subjects were asked secs.after to:a)identify stimulus,b)to rate hedonics and intensity each on a 100 mm.analog scale.Than subjects rinsed their mouth. After a pause, procedure was continued.[Entire session=. 40 min.] Results showed: that all intraoral stimuli induce water Increments due to or acceleration did differ, while sour not caused sucrose significantly higher HR acceleration, than did water or sucrose. 3 types of soup induced a HR increment ranking between values to water or sucrose and that to acid. EEG-arousal was found to be sensitive than HR in reflecting hedonics. Some subjects responded with marked arousal, others to a degree. "Responders "displayed lesser stronger reactions to aversive samples than to pleasant ones. HR changes correlate considerably hedonic estimates Still, further testing is with establish to correlation between physiological and psychophysical measures.

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*)Study sponsored by Internat.Glut.Tech.Comm.(IGTC)

<u>Infant</u> Sucking Responses to Sweet Solutions GRINKER J and BOSE K (University of Michigan)

It is generally accepted (see Beauchamp and Moran, 1982; Birch, 1979) that while infants are responsive to sweet tastes immediately postpartum, without exposure or experience this heightened response disappears. However, all previous studies have been based on intake measures. It is possible in naturalistic experiments that the infants' willingness to consume sweetened solutions interacts with maternal efforts to feed them. We report on a longitudinal study of infants of white middle or upper-middle class women. Sucking responses to small quantities (2ml) of sucrose solutions ranging from 1/16 to 1/2 M were elicited immediately postpartum. All infants showed increased number of sucks, decreased rate and bout number and there were no differences between infants of obese and nonobese mothers Grinker and Bose, 1986). Maternal obesity was based on pregnancy weights (greater than 120% of ideal weight/height ratios (Metropolitan Life Insurance Co, 1959) and reconfirmed by anthropometrics at the time of delivery. Our recent data at retest (age 3 months) suggests that not only do infants maintain an enhanced response to sucrose compared with water but also that infants of obese mothers respond more highly than infants of normal weight mothers. All mothers denied feeding sweetened solutions to their infants and were concerned about the development of obesity. Furthermore, taste test of liquid dairy products sweetened with sucrose suggest differences between obese and nonobese mothers. These data suggest that taste preference and intake are not identical in older infants and that if infants of obese mothers continue to show heightened response to sweet solutions a potential mechanism for the development of increased fatness could exist.

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79 Factors Responsible for Changes in Salt Taste Preference
Following Alterations in Dietary Sodium Intake in Human Adults.
GARY K. BEAUCHAMP, MARY BERTINO (Monell Chemical Senses
Center) and KARL ENGELMAN (Clinical Research Center, Hospital
of the University of Pennsylvania)

Variations in dietary exposure to salt alter salt taste preferences. In previous studies, we reported that individuals placed on self-maintained lowered-sodium diets subsequently preferred as optimal lower levels of salt in food relative to predict preference. These results have been replicated in several studies in other laboratories as well as in our own. We suggested that mechanisms underlying this change in preference could include physiological and/or psychological accommodation to the lowered-sodium diets. Two recent studies have implicated the latter. First, we found that when individuals increased their salt consumption by either (a) adding extra salt to their food; or (b) consuming the same amount of added salt as tablets, thereby not tasting it, only the former group exhibited changes in taste preference: the optimum amount of salt in food increased. Thus, tasting of the added salt appeared crucial. Second, we have recently completed a complimentary study on the effects on lowered-sodium diets combined with the availability of ad libitium salt on sodium consumption and salt taste preferences. Eleven students consumed all their meals and snacks in a Clinical Research Center for 13 weeks. During the first 3 weeks and the last week, the diet contained approximately 135 mEq Na which was reduced to approximately 70 mEq during weeks 4-13. A preweighed salt shaker was available for use throughout the entire period. Evaluations of salt taste preference and intensity scaling were conducted at regular intervals. Subjects compensated only slightly for the reduction in dietary sodium, making up less than 20% of the decrement with increased salt shaker use. This resulted in a substantial overall reduction in sodium consumption. In spite of this, no changes in taste preference or perception were found. We suggest that since the salt shaker placed salt on rather than in the food, subjects retained enough sensory experience with salty tastes to preclude changes in preference. In conjunction with the work described above, it is hypothesized that changes in taste perception following changes in dietary sodium are due to the amount of experience with salty tastes and not directly to the change in the absolute amount of sodium ingested.

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Heightened Salt Taste Sensitivity in Amiloride-Treated Normotensive Adults. RICHARD D. MATTES (Monell Center), CAROL M. CHRISTENSEN (Monell Center), KARL ENGELMAN (Hosp. of the Univ. of Penna.)

Accumulating evidence indicates that part of the salt taste transduction mechanism involves passage of sodium ions through amiloride-sensitive channels in taste cell membranes. Amiloride is used clinically as a potassium sparing diuretic. The extent to which amiloride, used under this context, is secreted in saliva, modifies salivary flow and composition, alters taste function and/or influences sodium intake was a primary aim of this study.

NOTES

Seventy-three normotensive adults were randomly assigned to receive 5 mg BID of amiloride (n=24), 50 mg/d of Hydrochlorothiazide (HCTZ) (n=24), or a placebo (n=25) in a double-blind cross-over study. Subjects were administered an appropriate placebo for weeks 1 and 2 active drug (except for controls) for weeks 3-10 and placebo again for 3 weeks. Test sessions conducted at weeks 1,2,4,6,10 and 13 involved the following in the stated sequence: Resting and then stimulated saliva collection (both analyzed for flow rate, Na, K and, when appropriate, amiloride), measurement of taste recognition thresholds for NaCl, sucrose and citric acid via a forced-choice staircase procedure, and concurrent assessment of suprathreshold sensitivity and preferred concentration by magnitude estimation and line scale ratings respectively for aqueous NaCl, crackers with added NaCl and aqueous citric acid. Two 24-hour urines collected prior to test sessions were analyzed for Na, K and creatinine and served as estimates of sodium intake.

Relative to baseline, a specific decrease in NaCl threshold was observed among patients treated with amiloride (p.05, .01, .05 at weeks 4, 6 and 10 respectively). This group also had significantly lower salivary Na levels (p.05). Amiloride was was not detected in saliva at levels as low as 10^{-6} M. HCTZ-treated and placebo subjects displayed no consistent changes in salivary or gustatory function, but relative to baseline, the former excreted (consumed) significantly more Na (p.05, .01, .05 at weeks 4,6 and 10 respectively). There was no subjective awareness of changes in salt preference or diet.

In summary, chronic exposure to therapeutic doses of amiloride heightens salt taste sensitivity without influencing sodium intake, whereas the reverse applies to HCTZ use.

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181 Taste Intensity Tracking of Aqueous Square Wave Trains. STEVEN KELLING (Dept. of Psychology and Field of Physiology) and BRUCE HALPERN (Dept. of Psychology and Section of Neurobiology and Behavior) (Cornell University, Ithaca NY 14853).

GENERAL PROCEDURE Subjects (4) tracked total taste intensity of 2 mm NaSac with a single axis joystick, sampled every 100 msec by a computer. At stimulus onset a trace appeared on a monitor and moved from left to right. If the joystick was moved, the trace swept vertically. Taste intensity was judged in proportion to 2000 msec continuous flow standards. The standard was represented by a line on the screen located at 75% of maximum height. The Kelling and Halpern closed flow liquid delivery apparatus (Chem. Senses: 1986) was used at a flow rate of 10 ml/sec for 4 sessions. The same number (2) of stimulus trials and control trials (distilled water) were run at 200, 600, 1000, and 2000 msec duration 5 Hz square wave trains (Kelling and Halpern, ISOT IX, in press). Distilled water preceded for 10 sec, and followed for 15 sec, each trial. RESULTS Onset of taste intensity tracking was \$\geq 1000\$ msec after stimulus onset. Maximum intensity increased with increased duration: 26% of the standard for 200 msec trains, 43% for 600 msec trains, 60% for 1000 msec trains, and 83% for 2000 msec trains. The time the intensity was above baseline increased with train duration: 900 msec at 200 msec trains. The time the intensity was above baseline increased with train duration: 900 msec at 2000 msec trains. The time the intensity slopes were best fit by logarithmic functions (r2 = .757 to .988). Stimulus errors were \$\leq 6\%; control errors were \leq 6\%; control error

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