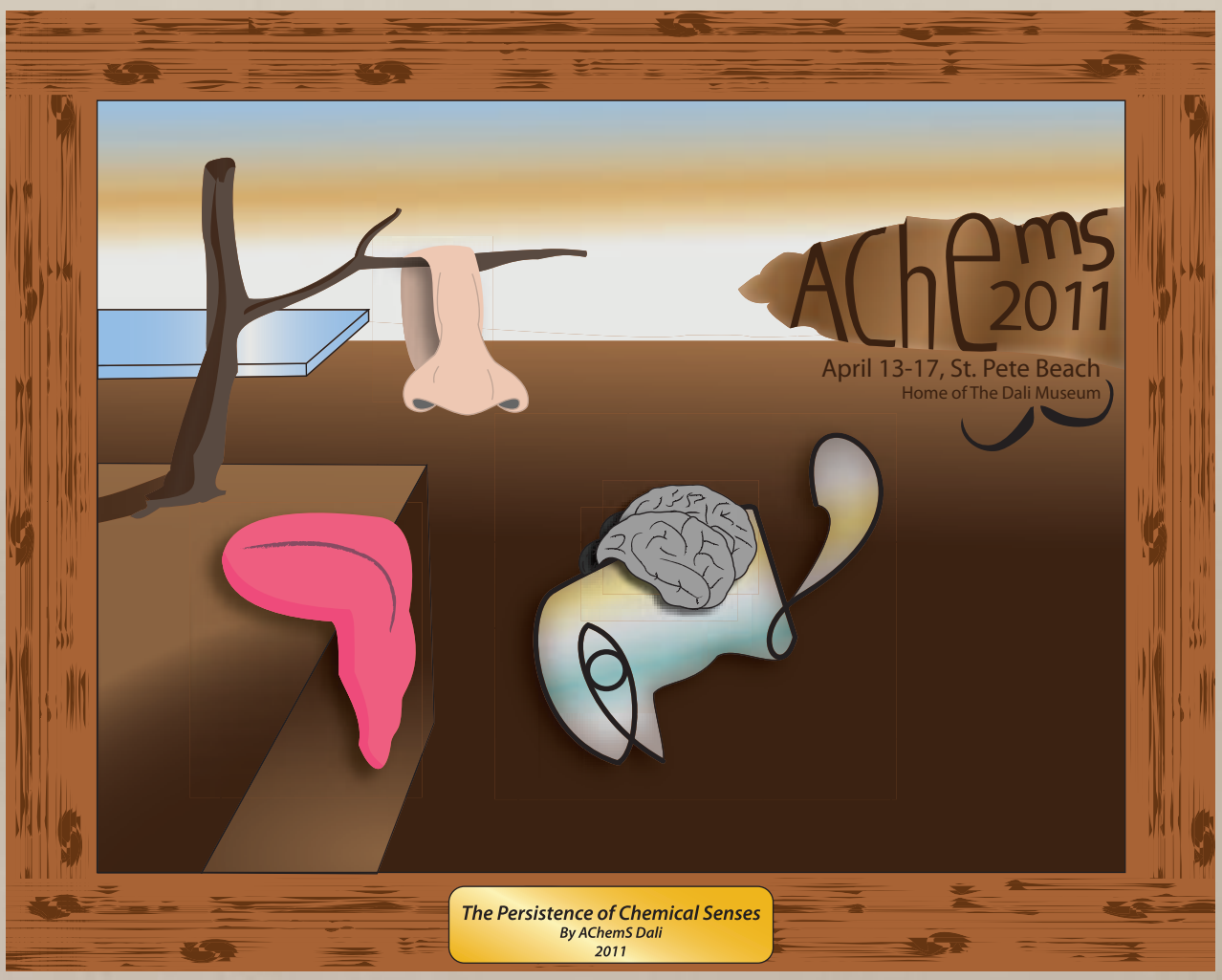
AChemS
Association for Chemoreception Sciences



ACHEMS 2011 ANNUAL MEETING

ABSTRACTS

APRIL 13-17, 2011
TRADEWINDS ISLAND GRAND
ST. PETE BEACH, FLORIDA

refresh**everything**.com

www.iff.com

IFF International Flavors & Fragrances Inc.

we create unique scent & taste experiences people love

AChemS
Association for Chemoreception Sciences

AChemS extends special thanks and appreciation for grant support from:

The National Institute on Deafness and Other Communications Disorders
and the
National Institute on Aging, NIH

The Association for Chemoreception Sciences is also grateful for the generous support of its Corporate Sponsors:

Diamond Level Sponsors

Givaudan

PEPSICO

Platinum Level Sponsor

IFF

Silver Level Sponsors

A taste of the future. AJINOMOTO.

Firmenich

kao

Other Sponsors

chromocell
ACCELERATING SCIENCE

REDPOINT BIO

A special thank you to **Ghislaine Polak** and the late **Ernest Polak** for supporting the **Polak Young Investigator Awards** and the **Junior Scientist Travel Awards**.

The Association for Chemoreception Sciences thanks our Corporate Members for their support:

A taste of the future. AJINOMOTO.

IFF

TAKASAGO

AChemS
Association for Chemoreception Sciences

2011 Annual Meeting Exhibitors

Noldus Information Technology, Inc.
Noldus Information Technology (www.noldus.com) offers systems for the recording, tracking, coding, and analysis of behavior and simultaneously recorded data. Our solutions include EthoVision XT, The Observer XT, and DanioVision. Please visit our booth for a free demonstration and information on how our solutions can assist you in your research.
Company representative: Wilant van Giessen

Osmic Enterprises, Inc.
Osmic Enterprises, Inc. produces and distributes the OLFACT™ Test Battery, a series of computerized tests to assess olfactory function. Tests include a threshold test, and identification test, a discrimination test, and an odor memory test. Stimuli are generated via a miniature olfactometer, with administration of the tests and recording of responses under computer control.
Company representatives: Lloyd Hastings and Bruce Johnson

Oxford University Press
Oxford Journals, part of Oxford University Press, publishes Chemical Senses, which is sponsored by AChemS, ECRO and JASTS.
Company representative: Jennifer Boyd

Tucker-Davis Technologies
TDT provides integrated hardware/software solutions for sensory neuroscience, such as evoked responses and single unit recording recordings. Stop by our exhibit to preview the latest additions to the System 3 platform, including our next generation multi I/O processor, real-time data streamer, video tracker, and integrated data acquisition with olfactometer.
Company representatives: Victor Rush and Nafi Yasar

AChemS
Association for Chemoreception Sciences

2011 Awardees

33rd Annual Givaudan Lectureship - Givaudan Corporation
Karel Svoboda, PhD, Howard Hughes Medical Institute,
Janelia Farm Research Campus

17th Annual Ajinomoto Award to Promising Young Researcher in the Field of Gustation
John Hayes, PhD, Pennsylvania State University

International Flavors and Fragrances Award for Outstanding Research on the Molecular Basis of Taste
Scott Herness, PhD, Ohio State University College of Dentistry

20th Annual Moskowitz Jacobs Award for Research in Psychophysics of Human Taste and Smell
M. Yanina Pepino de Gruev, PhD, Washington University, St. Louis

Max Mozell Award for Outstanding Achievement in the Chemical Senses
Thomas Finger, PhD, University of Colorado, Denver, School of Medicine

AChemS Young Investigator Award for Research in Olfaction
Nathalie Mandairon, PhD, Lyon University, France

Don Tucker Memorial Award (2010 Awardee)
Mavis Irwin, University of Utah

The Polak awards are funded by the Elsje Werner-Polak Memorial Fund in memory of our niece gassed by the Nazis in 1944 at age 7: Ghislaine Polak and the late Ernest Polak

Polak Young Investigator Award Recipients
Vikas Bhandawat, Duke University
Gabriela Krasteva, Justus-Liebig-University/Institute of Anatomy and Cell Biology
Victor Luna, NIH
Tadahiro Ohkuri, Kyushu University
Alison Ventura, Monell Chemical Senses Center
Jan Weiss, University of Saarland School of Medicine

We are pleased to announce that six 2011 Polak Junior Scientists Travel Fund Awards were given for this year's meeting.
The purpose of this award is to provide funds to cover travel and meeting expenses for junior investigators who would not otherwise be able to attend because of financial constraints.

AChemS Travel Fellowships for Diversity Recipients
Funded by a generous grant from the National Institute on Deafness and Other Communication Disorders and the National Institute on Aging, NIH
Madelyn Baez, Brandeis University
Mavis Irwin, University of Utah
Valerie Mendez-Gallardo, University of Iowa
Carolina Penalva-Arana, Indiana University
Aleida Silva-Garcia, Philadelphia College of Osteopathic Medicine
Tamika Wilson, Drexel University

AChemS Student Housing and Travel Award Recipients
Funded by the Polak Foundation: Ghislaine Polak and the late Ernest Polak
Jessica Armstrong
Matthew Biehl
Alison Boxwell
Julie Boyle
Elizabeth Brown
Trevor Cessna
Rui Chang
Jennifer Chen
Kepu Chen
Chandra Cherukuri
Annika Cichy
Priya Date
Yi Dong
Sara Dudgeon
David Dunston
Amanda Elson
Cathryn Anne Griffiths
Constanze Hartmann
Pauline Joussain
Richard Keith
Florence Kermen
Amanda Klein
Franziska Krone
Kurt Krosnowski
Peter Lai
Michael La Sala
Matthias Luebbert
Kristin McCombs
Lingbin Meng
Susanna Mitro
Midori Ogawa
Arthi Padmanabhan
Matthew Phillips
Andrea Ponting
Janet Prince
Michelle Rebello
Paige Richards
Kyle Roddick
Konstantin Rybalsky
Stefanie Sandgruber
Valentin Schriever
Sara Shanahan
Madison Shoaf
Amber Walker-Bolton
Yuanyuan Wang
Jennifer Whitesell

AChemS
Association for Chemoreception Sciences

Committees

AChemS Executive Committee 2010-2011

President	Don Wilson, PhD	Nathan Kline Institute & NYU School of Medicine
Past President	Scott Herness, PhD	Ohio State University
Senior Advisor	Peter Brunjes, PhD	University of Virginia
President Elect	Timothy McClintock, PhD	University of Kentucky
Secretary	Dana Small, PhD	JB Pierce Laboratory/Yale University
Membership Chair	Steven Munger, PhD	University of Maryland School of Medicine
Program Chair	Matt Wachowiak, PhD	University of Utah
Treasurer	Carol Christensen, PhD	Monell Chemical Senses Center
Program Chair Elect	Minghong Ma, PhD	University of Pennsylania
Sr. Councilor	Helen Treloar, PhD	Yale University
Jr. Councilor	Kevin Daly, PhD	West Virginia University

AChemS Program Committee 2010-2011

Matt Wachowiak (Chair, 2011)
Harriet Baker
Linda Barlow
Tom Bozza
Paul Breslin
Jean-Francois Cloutier
Diego Rodriguez Gil
Don Katz
Sue Kinnamon
Chris Lemon
Minghong Ma
Frank Margolis
Robert Margolskee
Hiro Matsunami
Steve Munger
Catherine Rouby
Susan Travers

MEETING EVALUATION

The meeting evaluation is available online this year. Please visit www.achems.org to give us your feedback on the meeting. Your input helps AChemS' leadership continue to offer quality annual meetings and member services.

#.5 **GIVAUDAN LECTURE**

The neural networks underlying haptic object localization
Karel Svoboda
Janelia Farm Research Campus, Howard Hughes Medical Institute, Ashburn, VA

The cerebral cortex is the largest part of the mammalian brain and plays roles in most flexible behaviors. Neocortical circuits are remarkably similar across functional areas and species. Our goal is to understand the principles that organize neocortical circuits and to decipher how they process information and guide behavior.

We focus on the neural circuits that underlie whisker-dependent somatosensation. Mice and other rodents use their whiskers to recognize and localize objects. Quantitative psychophysical methods allow us to track the motor strategies and sensory inputs used by mice in whisker-based object localization. At the same time we record from and manipulate specific neuronal populations within mapped neural circuits to discover causal relationships between neural activity and behavior. I will present recent data on how neurons in the somatosensory and motor cortex code for object location and how these representations are transformed during learning.

#1 **SYMPOSIUM - OPTOGENETICS: USING LIGHT TO STUDY SMELL**

Optogenetics: Using light to study smell
Venkatesh N Murthy[1], Justus V Verhagen[2,3]
[1]Harvard University, Dept of Molecular & Cellular Biology and Center for Brain Science Cambridge, MA, USA, [2]The John B. Pierce Laboratory New Haven, CT, USA, [3]Yale Medical School, Dept of Neurobiology New Haven, CT, USA

Progress in understanding the circuitry, coding and behavior in chemosensory systems has been catalyzed by the burgeoning field of optogenetics. Optogenetics involves methods to monitor and control neural activity using light-sensitive proteins. The expression of light sensitive proteins such as channelrhodopsins in targeted neuronal populations in a variety of species has allowed systematic and remote activation of neural elements with micron-scale spatial, and millisecond-scale temporal precision. In this symposium, investigators at the leading edge of this field will present their latest work exploiting optogenetics. Topics will cover multiple species and multiple levels of organization, including synaptic transmission, neural circuit analysis, coding and information processing, neurogenesis and plasticity, and behavior. Investigators will also discuss potential pitfalls, technical and biological limitations and will share practical knowledge. A plenary discussion ends the symposium, which is targeted at a wide audience.

#2 **SYMPOSIUM - OPTOGENETICS: USING LIGHT TO STUDY SMELL**

Optogenetic Dissection of Local Circuits in Sensory Cortex
Jessica A. Cardin
Yale University/Dept of Neurobiology New Haven, CT, USA

The recent development of optical methods for controlling the activity of specific populations of neurons holds tremendous promise for understanding the roles of neural subtypes in local circuits, a major goal of systems neuroscience. Optogenetic tools, in combination with electrophysiology, allow identification and manipulation of specific groups of genetically identified neurons in active neural networks *in vivo* in a spatially and temporally precise manner. Cre-dependent expression of the light-activated nonspecific cation channel Channelrhodopsin (ChR2), the light-activated chloride pump Halorhodopsin (eNpHR), and other optogenetic tools can result in a highly selective population of neurons whose activity is regulated by brief light pulses. This specific expression pattern provides rigorous extracellular identification of neurons via a physiological tag. Furthermore, fine temporal control permits the activation and suppression of specific cells at precise moments during sensory stimulation, allowing online dissection of local sensory circuits. This integrated approach provides unprecedented access to both synaptic interactions and network operations under functionally and behaviorally relevant conditions. We have used these combined techniques to explore the roles of specific classes of inhibitory and excitatory neurons in sensory processing in the primary somatosensory (S1) and visual (V1) cortices in vivo. Using bidirectional optogenetic control of specific classes of GABAergic interneurons, we find that each interneuron class makes distinct contributions to sensory processing and engages different resonant properties of the local circuit.
Acknowledgements: Supported by the NIH/NEI, the Whitehall Foundation, the Klingenstein Foundation, and NARSAD.

#3 **SYMPOSIUM - OPTOGENETICS: USING LIGHT TO STUDY SMELL**

Temporal processing of activity patterns downstream of the olfactory bulb: an optogenetic analysis in zebrafish
Rainer W Friedrich, Francisca Blumhagen, Peixin Zhu, Jennifer Shum, Yan-Ping Zhang Schaerer
Friedrich Miescher Institute for Biomedical Research Basel, Switzerland

In insects, oscillatory synchronization of projection neurons in the antennal lobe has important effects on the read-out of activity patterns in the mushroom body. To examine the impact of temporal patterning on the processing of activity patterns in a vertebrate, we optically manipulated mitral cell activity in zebrafish expressing channelrhodopsin-2 in the olfactory bulb and analyzed responses in telencephalic area Dp, the homolog of olfactory cortex. Spatio-temporal activity patterns with varying degrees of oscillatory synchrony were imposed onto mitral cells using a digital micromirror device. The carrier frequency of the oscillation was 20 Hz, corresponding to the frequency of odor-evoked oscillations. Whole-cell recordings confirmed that the membrane potential dynamics and firing patterns of mitral cells followed the temporal modulation of optical stimulation. In Dp neurons, oscillatory mitral cell input was reflected in subthreshold membrane potential fluctuations. However, these fluctuations

were small and did not contribute substantially to the large depolarization required to trigger action potentials. Dp neuron firing was controlled primarily by a balance of excitation and inhibition that changed on a slow time scale over the course of a response. Firing probability of Dp neurons did not coincide with the time when mitral cell firing was maximal, but occurred at later response phases when mitral cell activity patterns tend to be decorrelated. Similar observations were made in response to odor stimulation. These results indicate that Dp is not specifically optimized to detect oscillatory synchrony in mitral cell activity patterns. Rather, temporal filtering by neuronal circuits in Dp appears to favor the read-out of activity patterns that are maximally informative about odor identity. Acknowledgements: Novartis Research Foundation, Swiss National Fonds, Whitaker Foundation, Max-Planck-Society, German Research Foundation

#4 **SYMPOSIUM - OPTOGENETICS: USING LIGHT TO STUDY SMELL**

Smelling Time: Perception of Sniff Phase in Mouse Olfaction

Thomas Bozza[1,2], Matt Smear[1,2], Roman Shusterman[1], Dmitry Rinberg[1]
[1]Janelia Farm Research Campus Ashburn, VA, USA, [2]Dept. of Neurobiology and Physiology, Northwestern University Evanston, IL, USA

Odor responses are spatially and temporally patterned. In mammals, a particularly conspicuous form of temporal patterning is timing of activity with respect to the sniff cycle. Interestingly, neurons often respond to odor stimuli not only by changing firing rate, but also by changing the phase of the sniff cycle in which they fire suggesting that the timing of neuronal activity in the sniff cycle may encode information. However, it is unclear whether sniff phase information is perceptually available to animals. We are using a combination of optogenetics and psychophysics in mice to ask whether timing of neuronal activity with respect to sniff phase by itself can contribute to odor perception. We have generated mice in which channelrhodopsin2 (ChR2) is expressed in all olfactory sensory neurons (OSNs), or in OSNs that express specific odorant receptors. In these mice, light directed at the olfactory epithelium or glomeruli drives firing of mitral/tufted cells in the olfactory bulb. ChR2-expressing mice rapidly learn to report detection of light stimuli, indicating that they can smell the light, even through a single glomerulus. In addition, mice can readily learn to discriminate identical light pulses delivered at different phases of the sniff cycle. The temporal precision of this discrimination is remarkable—light stimuli occurring at inhalation onset can be distinguished from stimuli occurring 10 ms after inhalation onset. This sniff phase perception does not generalize to auditory or somatosensory stimuli. We conclude that mice can discriminate spatially fixed glomerular input patterns solely based on timing relative to the sniff cycle. This optogenetic approach provides unprecedented control of olfactory input to further examine how defined changes in neuronal activity affect odor perception. Acknowledgements: HHMI Visiting Scientist Program and NIDCD 1R21DC010911

#5 **SYMPOSIUM - OPTOGENETICS: USING LIGHT TO STUDY SMELL**

Discrimination of static and dynamic optical patterns presented to the olfactory bulb of transgenic mice expressing channelrhodopsin in mitral cells

David C. Willhite[1,2], Thomas S. McTavish[2], Gordon M. Shepherd[2], Justus V. Verhagen[1,2]
[1]The John B. Pierce Laboratory New Haven, CT, USA, [2]Yale School of Medicine, Dept. Neurobiology New Haven, CT, USA

Bulbar responses to odorants are known to be dynamic, in that glomerular responses evolve over time. Until recently, no stimulus method was available that could systematically vary static or dynamic patterns. Activation of neurons expressing channelrhodopsin with light stimulation allows arbitrarily complex patterns and combinations to be examined which were not possible using electrical methods. Further, the stimulated neuron populations can be defined by the promoter(s) which express the light-activated channel. We integrated a digital micromirror device (DMD) into a microscope to present patterns to the dorsal olfactory bulb surface of mice expressing ChR2 under the Thy1 promoter (Arenkiel et al., Neuron 2007). Using this system, we addressed the question of whether temporal patterning has biological significance, i.e. whether a spatio-temporal code is employed in the mouse OB or a purely spatial code. Using a go no-go water reward paradigm, we show that mice are able to discriminate arbitrary static patterns presented continuously during the trial. We further evaluate the degree of discriminability of various static and dynamic activation patterns in mice. Acknowledgements: This work is supported by NIH/NIDCD grants RO1DC009994, RO1DC00997701, and RO1DC0086.

#6 **SYMPOSIUM - OPTOGENETICS: USING LIGHT TO STUDY SMELL**

Shining light on adult neurogenesis in the mouse olfactory bulb

Adi Mizrahi
The Hebrew University of Jerusalem, Department of Neurobiology Jerusalem, Israel

The mammalian olfactory bulb (OB) receives continuous supply of adult-born interneurons throughout adulthood. These unique neurons have been postulated to provide the OB with a heightened capacity for neuronal plasticity. However, the contribution of adult-born neurons to sensory coding remains completely unknown. Difficulties in studying adult neurogeneis arise for many reasons but one reason is that they are sparsely intermingled with other (non adult-born) resident neurons. "Light" and "genetics" are therefore potentially good candidates to target, manipulate and "read" from this specific neuronal population. Moreover, the superficial location of the rodent's OB makes it an experimentally convenient location for combining optogenetics with imaging. I will present and discuss our recent efforts to study adult-born neurons with "optogenetic" probes. In order to read sensory response profiles from these unique neurons, we developed a novel preparation to enable high resolution two-photon time lapse imaging experiments through a chronic cranial window for many months in the intact animal. To test the potential of this preparation we transduced adult-born neurons by lentivirus injections into the stem cell niche and

imaged them in the OB. Over-expression of the optogenetic calcium sensor "GCapm3.0" provides first insights into the odor responses of these neurons. Over-expression of the optogenetic silencer "NpHR3.0" allows us to inhibit these neurons efficiently and specifically *in vivo* with high precision. Thus, our experiments provide a comprehensive toolbox to study the physiology of this unique neuronal population as it replenishes the bulbar network.

#7 **SYMPOSIUM - OPTOGENETICS: USING LIGHT TO STUDY SMELL**

Optogenetic analysis of olfactory cortical circuits
Venkatesh N Murthy, Akari Hagiwara, Sumon K Pal, Foivos Markopoulos
Harvard University, Dept of Molecular & Cellular Biology and Center for Brain Science Cambridge, MA, USA

Primary olfactory cortical areas receive direct input from the olfactory bulb, and are involved in odor perception and learning. In addition to direct inputs from the olfactory bulb, there are abundant associational inputs to cortical neurons, including local recurrent connections and projections from other olfactory cortical regions. Disentangling the contributions of different association inputs to cortical function has not been easy, in part, because of the difficulty in separately activating them. We studied the functional properties of olfactory cortical feedforward and feedback connections by selectively labeling different classes of axons using viral expression of channelrhodopsin-2 and activation of synapses using light. These experiments revealed target- and pathway-specific heterogeneity in the synaptic properties of different olfactory association connections. A separate set of experiments combining calcium imaging with electrophysiology indicated that recurrent excitatory connections within the anterior piriform cortex are functionally sparse and weak, but are denser in the posterior piriform cortex. Finally, optogenetic studies of feedback connections from olfactory cortex to the olfactory bulb revealed functional connections not described previously. In sum, our experiments shed light on information processing by olfactory cortical circuits. Acknowledgements: Astellas Foundation for Research on Metabolic Disorders, Swiss National Science Foundation, an Anonymous Foundation, and Harvard University.

#9 **PRESIDENTIAL SYMPOSIUM: OLFACTION IN TRANSLATION**

Control of olfactory bulb circuitry by cortical feedback pathways
Ben W. Strowbridge
Case Western Reserve Univ., Neurosciences Dept. Cleveland, OH, USA

Most GABAergic inhibition onto olfactory bulb mitral cells arises from dendrodendritic microcircuits. Through these spatially-localized reciprocal synapses with the distal dendrites of granule cells, mitral cell discharges are shaped by widespread glomerular input patterns. Surprisingly, the ability of dendrodendritic reciprocal synapses to mediate recurrent and lateral inhibition appears to be tonically attenuated by Mg^{2+} ions normally present in the CSF that block critical NMDA receptors. Using 2-photon-guided microstimulation in rodent olfactory bulb brain slices, we found that cortical feedback projections often target spines along the proximal dendrites of granule cells and can relieve the tonic NMDAR blockade at distal dendrodendritic synapses. We also found different forms of long-term plasticity at proximal and distal excitatory inputs to granule cells. The convergence of two types of excitatory inputs onto GABAergic granule cells provides a novel mechanism for piriform cortex to regulate the degree of inter-glomerular processing within the olfactory bulb. Acknowledgements: Supported by NIH grant DC04285

#10 **PRESIDENTIAL SYMPOSIUM: OLFACTION IN TRANSLATION**

Using the chemical senses to investigate behavioral control in Drosophila
Scott Waddell
Department of Neurobiology, University of Massachusetts Medical School, Worcester, MA, USA

A goal of neuroscience is to understand how the brain orchestrates appropriate behavioral responses. We use the precision afforded by genetics in the fruit fly and the chemical senses to study the neural processes underlying the formation, consolidation and expression of appetitive memory. Consolidated memory is most efficiently formed by training flies with odor exposure and nutritionally valuable sugars. Palatable taste and nutrient quality apparently provide parallel reinforcing signals. Furthermore, learning and the display of memory performance are most robust in hungry flies permitting an investigation of mechanisms of state-dependence. We found that the internal state of hunger is integrated with appetitive memory expression through a hierarchical modulatory mechanism. Stimulating neurons that express Neuropeptide F (dNPF), an ortholog of mammalian NPY, mimicks food-deprivation and promotes memory performance in satiated flies. dNPF works through a subset of dopaminergic neurons that innervate a distinct region of the mushroom bodies. Blocking these dopaminergic neurons releases memory performance in satiated flies whereas stimulation suppresses memory performance in hungry flies. The mechanisms constraining learning to hungry flies are apparently different. I will present our latest understanding of state-dependent learning and action selection. This research is supported by NIH MH09883 and MH081982.

#11 **PRESIDENTIAL SYMPOSIUM: OLFACTION IN TRANSLATION**

Olfaction Research and the Fragrance Industry
Charles S. Sell
Givaudan, Ashford, United Kingdom

The last two decades have seen enormous progress in our understanding of olfaction. Thanks to the pioneering work of Linda Buck and Richard Axel, we now have access to the entire spectrum of human olfactory receptors and are beginning to map their selectivities for odorant molecules. Crystal structures of GPCRs are appearing and so we are now learning more about how odorants bind to and activate their cognate receptors. Through developments in imaging techniques we are beginning to study how olfactory signals are transmitted and interpreted in the

brain. Over the same time period, the fragrance industry has seen increasing constraints in terms of safety, environment, regulation, economics and raw material issues and consumers are becoming more demanding. In this talk, I will discuss some of the issues currently facing the industry and how research into olfaction might address these and also open up new opportunities.
Acknowledgements: None

#12 PRESIDENTIAL SYMPOSIUM: OLFACTION IN TRANSLATION

Using Olfaction to Investigate Alzheimer's Disease

Claire Murphy[1,2]
[1]San Diego State University San Diego, CA, USA,
[2]University of California San Diego, CA, USA

Alzheimer's disease (AD) is a devastating neurodegenerative disorder that afflicts 4.5 million Americans, is rising at an alarming rate as the population ages, presents a significant public health issue and has profound economic consequences for the individual and for society. The disease erodes memory and cognition and ultimately the personality and sense of self, leaving its victims unable to care for themselves. Efforts to develop prevention strategies and effective pharmacological interventions are keen, but, at present AD is incurable. As interventions become available, determining who is at risk for the disease, the point of disease onset, disease progression, and the effectiveness of interventions will be critical to prevent significant neurological compromise. Neuropathological changes in AD begin in entorhinal and transentorhinal areas and in the olfactory bulb, regions critical for processing olfactory information, and significantly, neuropathology in these areas precedes clinical symptoms. Entorhinal cortex and perirhinal cortex, CA1 area of hippocampus, amygdala, and the association cortices show the heaviest pathological burden as the disease progresses, suggesting further involvement of areas key to olfactory processing. We and others have used the unique vulnerability of olfaction to investigate Alzheimer's disease. This presentation will briefly summarize key neuropsychological, event-related potential, and neuroimaging findings and present new data from ERP and fMRI research focused on the olfactory system and on brain areas that show connectivity with olfactory regions. Understanding the relationship between olfactory system dysfunction and dementia may facilitate identification of biomarkers and targets for intervention to prevent, delay or arrest disease progression.
Acknowledgements: Supported by NIH grants AG04085 from the National Institute on Aging and DC02064 from the National Institute on Deafness and other Communicative Disorders.
I gratefully acknowledge the SDSU Lifespan Human Senses Laboratory, UCSD Alzheimer's Disease Research Center (P50 AG05131), and The Scripps Research Institute, Ethan Solomon, Lori Haase, Erin Green, Joel Kowalewski, and Drs. Charlie D. Morgan, John Polich, Robert Katzman, and Leon Thal.

#13 PLATFORM PRESENTATIONS: OLFACTION

Smelling sulfur: An odorant receptor for divalent sulfur compounds employs copper ion as a cofactor

Hanyi Zhuang[1,2], *Xufang Duan*[1,2], *Jian Zhang*[1], *Zhimin Huang*[1], *Zhen Li*[1], *Yi Pan*[1], *Qiuyi Chi*[4], *Siji Thomas*[3], *Shao-Zhong Zhang*[3], *Eric Block*[3], *Guo-Qiang Chen*[1,2], *Hiroaki Matsunami*[4,5]
[1]Department of Pathophysiology, Key Laboratory of Cell Differentiation and Apoptosis of National Ministry of Education, Ruijin Hospital, Shanghai Jiao Tong University School of Medicine Shanghai, China, [2]Institute of Health Sciences, Shanghai Institutes for Biological Sciences of Chinese Academy of Sciences and Shanghai Jiao Tong University School of Medicine Shanghai, China, [3]Department of Chemistry, University at Albany, State University of New York Albany, NY, USA, [4]Department of Molecular Genetics and Microbiology, Duke University Medical Center Durham, NC, USA, [5]Department of Neurobiology, Duke University Medical Center Durham, NC, USA

The detection of volatile odorants is mediated by odorant receptors (ORs) in the olfactory sensory neurons (OSNs) of the nose, but the detailed mechanism of how a certain OR interacts with its ligand is largely unknown. Thiols, thioethers, and other low-molecular weight divalent sulfur compounds are notable in that they are extremely potent as odorants. Previously, it was proposed that metals may play a role in thiol-OR interactions, but whether metal ions change thiol-mediated OR activation is not known. (Methylthio)methanethiol (MTMT), a compound with adjacent thiol and thioether groups found specifically in male mouse urine, is a known semiochemical that attracts female mice. It has an intense odor, detected by humans at 100 parts per billion. Here we identify an OR that responds to MTMT and other structurally related sulfur compounds in heterologous cells. Importantly, we found that copper ion, but not other metal ions tested, is required for robust receptor activation. We propose that this effect is mediated by the interaction of an MTMT-copper complex with the OR domain facing the extracellular milieu.
Acknowledgements: This research is supported by grants to H.Z. from the National Natural Science Foundation of China (30970981), Shanghai Pujiang Program (09PJ1406900), Program for Innovative Research Team of Shanghai Municipal Education Commission, the Chen Guang Project funded by Shanghai Municipal Education Commission and Shanghai Education Development Foundation (2009CG15), and the Program for Professor of Special Appointment (Eastern Scholar) at Shanghai Institutions of Higher Learning, to H.M. from the National Institutes of Health, to J.Z. from National Basic Research Program of China (973 Program) (2011CB504001), and to E.B. from the National Science Foundation (CHE-0744578).

#14 **PLATFORM PRESENTATIONS: OLFACTION**

A Carnivore Odor Avoided by Prey

Stephen D Liberles[1], David M Ferrero[1], Jamie K Lemon[1], Daniela Fluegge[3], Stan L Pashkovski[4], Wayne J Korzan[1], Sandeep R Datta[4], Marc Spehr[3], Markus Fendt[2]
[1]Harvard Medical School/ Department of Cell Biology Boston, MA, USA, [2]Novartis Institutes for BioMedical Research/ Neuroscience Basel, Switzerland, [3]RWTH Aachen University/ Department of Chemosensation Aachen, Germany, [4]Harvard Medical School/ Department of Neurobiology Boston, MA, USA

Predator-prey relationships provide a classic paradigm for the study of innate animal behavior. Odors from carnivores elicit stereotyped fear and avoidance responses in rodents, although the predator-derived odors and rodent olfactory receptors involved are largely unknown. Here, we explore the molecular basis by which the olfactory system recognizes predator cues and elicits complex behavioral responses. We identified natural and synthetic ligands for 13 olfactory trace amine-associated receptors (TAARs) using a cAMP-based reporter gene assay, and found that one of these receptors selectively detects odors from several predators. We purified the TAAR agonist from bobcat (Lynx rufus) urine by silica gel chromatography, and identified it to be a biogenic amine, 2-phenylethylamine. Quantitative HPLC analysis of 2-phenylethylamine biosynthesis across 38 mammalian species indicates enriched production by numerous carnivores, with some producing >3,000 fold more than herbivores examined. Calcium imaging of neuronal responses in olfactory tissues slices identified a small population of carnivore odor-selective sensory neurons that also respond to 2-phenylethylamine. Two prey species, rat (Rattus norvegicus) and mouse (Mus musculus), avoid a 2-phenylethylamine odor source, and we developed methodology to deplete this chemical from a carnivore odor for loss-of-function behavioral studies, finding it to be required for a full avoidance response. These data indicate that mouse olfactory sensory neurons and chemosensory receptors have the capacity for interspecies cue recognition. One such cue, carnivore-derived 2-phenylethylamine, is a key component of a predator odor blend that triggers hard-wired aversion circuits in the rodent brain.
Acknowledgements: This work was supported by grants from the National Institute On Deafness And Other Communicative Disorders (SDL, Award Number R01DC010155), and the Deutsche Forschungsgemeinschaft (MS, SP724/2-1)

#15 **PLATFORM PRESENTATIONS: OLFACTION**

Trace amine-associated receptors map to a subset of dorsal glomeruli in the mouse

Rodrigo Pacifico[1], Brian Weiland[1], Caiying Guo[2], Dmitry Rinberg[2], Thomas Bozza[1]
[1]Northwestern University/Dept. of Neurobiology and Physiology Evanston, IL, USA, [2]Howard Hughes Medical Institute/Janelia Farm Research Campus Ashburn, VA, USA

Olfaction in mammals is mediated by a large repertoire (~1200 genes in mice) of G protein-coupled odorant receptors (ORs) which have been historically divided into two phylogenetically distinct groups, so called Class I and Class II ORs. Olfactory sensory neurons that express Class I or Class II ORs project to glomeruli in discrete domains of the dorsal olfactory bulb. Trace amine-associated receptors (TAARs) represent a third phylogenetic class of ORs (Class III,) a majority of which are expressed in the dorsal olfactory epithelium along with Class I and Class II ORs. TAARs respond *in vitro* to volatile amines, some of which are present in mouse urine and may convey social cues. However, little is known about the axonal projections and odorant response profiles of TAAR-expressing OSNs *in vivo*. We have characterized for the first time the axonal projections of OSNs expressing genetically-defined TAARs. To do this, we generated gene targeted mouse strains in which OSNs that express specific TAAR genes are labeled with fluorescent markers. We find that TAAR-specific OSNs project to a region of the caudal bulb near the interface between the dorsal Class I and Class II domains. Additionally, OSNs that express TAAR deletion alleles (in which a specific TAAR coding sequence is replaced with a marker gene) preferentially co-express other TAAR genes and innervate a circumscribed subset of dorsal glomeruli. Using *in vivo* optical imaging, we show that glomeruli within this region are activated by certain amine odorants. Taken together, the data suggest that dorsal TAARs are mapped to a third domain, which may respond selectively to a subset of volatile amines.
Acknowledgements: This project was supported by NIH/NIDCD grants R01DC009640-01A2 and R21DC010911-01

#16 **PLATFORM PRESENTATIONS: OLFACTION**

Inter-Glomerular Lateral Inhibition Suppresses Mitral Cell Output in a Timing-Dependent Fashion

Jennifer D Whitesell, Nathan E Schoppa
Neuroscience Program, Department of Physiology and Biophysics, and Rocky Mountain Taste and Smell Center, University of Colorado Anschutz Medical Campus Aurora, CO, USA

The OB includes dense networks of GABAergic interneurons, suggesting that lateral inhibitory interactions could impact glomerular input-output patterns. We used patch clamp recordings from mitral cells (MCs) in rat OB slices to test for the mechanisms of lateral inhibition. Electrical stimulation of a "conditioning" glomerulus (50-500 µA) that was 100-550 µm away from the "target" glomerulus of a MC decreased the excitatory synaptic current evoked by stimulation of OSNs at the target glomerulus by 38±7% (n=19, p=0.005). This effect was not mediated by GABA(B) receptors, since the bath solution contained 2 mM CGP55845. We performed microsurgical cuts to determine the cell-type(s) mediating this lateral inhibition and found that inhibition could still be observed in slices with a cut through the external plexiform layer (20±6% decrease, p=0.03, n=10), but a cut through the glomerular layer abolished lateral inhibition (1±2% decrease, p=0.5, n=9) implicating glomerular rather than granule cells. In addition, in recordings from external tufted (ET) cells, we observed GABAergic inhibitory post-synaptic currents (IPSCs; 45±6 pA at V_{hold} = 0 mV, n = 22) in response to electrical stimulation of a conditioning glomerulus, presumably arising from periglomerular cells. There was a close relationship between the duration of ET cell inhibition (half-width = 25±4 ms) and the time-interval between conditioning and target glomeruli stimulation (20 ms) that was most effective for suppression of MC excitation. Because ET cells have a prominent role in mediating feed-forward excitation between OSNs and

MCs (De Saint Jan et. al., 2009), these results suggest that lateral inhibitory input to ET cells may impact the OB's input-output relationship by preventing feed-forward excitation of MCs. Acknowledgements: NIDCD 2R01DC006640-06A1 (NES) NINDS 5T32NS007083-29 (JDW)

#17 **PLATFORM PRESENTATIONS: OLFACTION**

Neural Circuit Mechanisms for Pattern Detection and Feature Combination in Olfactory Cortex

Ian G Davison[1], Michael D Ehlers[1,2]
[1]Dept. of Neurobiology, Duke University Medical Center Durham, NC, USA, [2]Neuroscience Research Unit, Pfizer Global Research and Development Groton, CT, USA

Odors are initially encoded in the brain as a set of distinct physicochemical characteristics, but are ultimately perceived as a unified sensory object. It remains unclear how chemical features encoded by diverse odorant receptors and segregated glomeruli in the main olfactory bulb (MOB) are assembled into integrated cortical representations. Combining patterned optical microstimulation of MOB with in vivo electrophysiological recordings in anterior piriform cortex (PCx), we assessed how cortical neurons decode complex activity patterns distributed across MOB glomeruli. PCx firing was insensitive to single-glomerulus photostimulation. Instead, individual cells reported higher-order combinations of coactive glomeruli resembling odor-evoked sensory maps. Intracellular recordings revealed a corresponding network architecture providing each cortical neuron with weak synaptic input from a distinct and restricted subpopulation of MOB glomeruli. PCx neurons thus detect specific ensembles of activated glomeruli, providing an explicit neural representation of chemical feature combinations that are the hallmark of complex odor stimuli. Acknowledgements: This work was supported by NIH grant R01 MH086339 and the Howard Hughes Medical Institute (to M.D.E.).

#18 **PLATFORM PRESENTATIONS: OLFACTION**

Active Sampling Gates Intensity Coding in Olfactory Cortex

Anne-Marie M. Oswald, Nathaniel N. Urban
Carnegie Mellon University, Department of Biological Sciences, Center for the Neural Basis of Cognition Pittsburgh, PA, USA

An important issue in sensory neuroscience is understanding how changes in sampling behavior shape the activation of cortices and coding of sensory stimuli. In air-breathing animals, repeated cycles of inspiration and expiration temporally pattern olfactory input. During exploration, rodents vary the sampling rate of olfactory inputs from 1-3 Hz (passive breathing) to 5-12 Hz (active sniffing). Here we investigate how neurons in olfactory cortex respond to stimulus intensity when stimuli are delivered at active sniffing frequencies versus passive breathing frequencies. We recorded cortical neurons in a slice preparation of olfactory cortex. We quantified short-term depression mitral/tufted (M/T) cell to cortex synapses and used these results to simulate M/T population input. These population inputs were delivered at sampling frequencies consistent with active sniffing (8 Hz) and passive respiration (1 Hz). We found that cortical neurons code changes in stimulus intensity when population inputs are delivered at active but not passive frequencies. Increases in stimulus intensity are coded by increases in cortical firing rates that are phase-locked to simulated active sniffing cycles. Furthermore, the phase locking of cortical neurons is enhanced compared to the simulated M/T population. In contrast, cortical responses are invariant to changes in intensity and only weakly phase-locked to simulated inputs delivered at passive breathing frequencies. These cortical firing patterns are achieved through the differential recruitment of short-term depression at M/T-to-cortex synapses by inputs driven at active versus passive sampling frequencies. Taken together, our results show that changes in respiratory behavior gate the transfer of stimulus information between the olfactory bulb and cortex. Acknowledgements: RO3DC011375 to AO and R01DC0005798 and R01DC011184 to NU.

#19 **PLATFORM PRESENTATIONS: OLFACTION**

The Role of the Amyloid Precursor Protein in the Construction and Deconstruction of the Peripheral Olfactory System

Mark W. Albers
Massachusetts General Hospital Boston, MA, USA

Odor naming deficits presage the development of clinical symptoms in Alzheimer's disease (AD). Pathological hallmarks of AD are found in the olfactory bulbs and entorhinal cortices of asymptomatic elderly individuals, suggesting that the olfactory neural network is particularly susceptible to this disease.
To investigate the vulnerability of the peripheral olfactory neural circuit to early stages of AD, we generated a mouse line that overexpresses the Swedish mutation of human APP (hAPPsw) exclusively in a stochastic fraction of olfactory sensory neurons (OSNs) (<1%). Examination of these mice revealed a profound loss of M71-expressing and P2-expressing subpopulations of OSNs. We observed increased levels of activated caspase 3 immunostaining in the olfactory epithelia, predominantly in OSNs not expressing hAPPsw. Moreover, we found perturbed targeting of OSN axons in the olfactory bulb and reduced enantiomer discriminatory power in these mice. Expression of a cleavage product of hAPPsw, the A beta peptide, in sustentacular cells predominantly also altered axon targeting of sensory neurons in the olfactory bulb. Together, these results indicate that APP and the A beta peptide may act non-cell autonomously to mediate changes in the olfactory epithelium and olfactory bulb.
Suppression of hAPPsw expression reversed these phenotypes in adult mice. We are evaluating human olfactory epithelia and olfactory bulbs from symptomatic and asymptomatic individuals with AD pathology for the presence of these phenotypes. Finally, we have observed axon mistargeting phenotypes in APP null and APP haploinsufficient mice, suggesting that a delicate balance of APP and its cleavage products is critical for the formation and maintenance of the orderly glomerular map of OSN axons in the olfactory bulb. Acknowledgements: NIDCD K08, NIH New Innovator Award, Ellison Medical Foundation, American Federation of Aging Research

#20 **PLATFORM PRESENTATIONS: OLFACTION**

Network dysfunction, olfactory behavior impairments, and their reversibility in an Alzheimer's β-amyloidosis mouse model

Daniel W Wesson[1], Anne H Borkowski[1], Gary E Landreth[2], Ralph A Nixon[1], Efrat Levy[1], Donald A Wilson[1]
[1]Nathan Kline Institute & NYU School of Medicine Orangeburg, NY, USA, [2]Case Western Reserve Univ. School of Medicine, Dept. of Neurosciences Cleveland, OH, USA

The vulnerability of the olfactory system to Alzheimer's disease (AD) pathology and the high incidence of olfactory perceptual dysfunction in early stages of the disease makes the olfactory system a unique model for understanding mechanisms of synaptic and neural network dysfunction in AD. Here we demonstrate aberrant neural oscillations within the olfactory bulb (OB) and piriform cortex (PCX) of mice overexpressing human mutations of amyloid precursor protein (*APP*). Network dysfunction was evident starting at 3 months of age in APP mice, prior to the onset of significant behavioral impairments or comparable hippocampal network dysfunction. Coinciding with the onset of behavioral impairments, we found hyperactivity of odor-evoked responses in the PCX and enhanced coherence between the OB and PCX. In contrast, older APP mice with established disease-related pathology were characterized by hyporesponsive PCX odor-evoked activity and impaired behavior which were both recovered by treatment with a Liver-X Receptor (LXR) agonist. These results complement recent findings in other neural networks and suggest that disease-relevant network dysfunction can be transient and region specific, yet with lasting effects on cognition and behavior. Acknowledgements: This work was supported by National Institutes of Health grants DC003906 to D.A.W, AG017617 to R.A.N. and AG030482 to G.E.L.

#22 **SYMPOSIUM: NEW FRONTIERS IN CHEMESTHESIS**

Physiological basis of tingling paresthesia evoked by hydroxy-alpha-sanshool

Diana M Bautista
UC Berkeley, Department of Molecular & Cell Biology, Berkeley, CA, USA

Szechuan peppers and other members of the *Zanthoxylem* plant family have been used in traditional folk medicine to treat toothache and other types of trigeminal pain. In contrast to the intense burning pain associated with hot peppers of the *Capsicum* family, Szechuan peppers elicit a robust, benign numbing and tingling paresthesia, suggestive of an interaction with neurons involved in tactile sensation. Psychophysical studies in humans have shown that the alkylamide, hydroxy-alpha-sanshool, is the active ingredient in Szechuan peppers. We examined the effects of sanshool on cultured mouse somatosensory neurons and primary afferent fibers. We show that sanshool specifically activates two extremely sensitive, light touch receptors, the rapidly adapting A-beta fibers and the ultra-sensitive D-hairs. In addition, sanshool inhibits mechanical sensitivity of a subset of nociceptors, with no effect on thermal sensitivity. Thus, sanshool provides a novel pharmacological tool for discriminating functional subtypes of cutaneous mechanoreceptors. The identification of sanshool-sensitive fibers represents an essential first step in identifying the cellular and molecular mechanisms underlying tingling and numbing paresthesias. Acknowledgements: Burroughs Wellcome Fund, Pew Program in Biomedical Science, McKnight Scholar Program, NIH New Innovator Award.

#23 **SYMPOSIUM: NEW FRONTIERS IN CHEMESTHESIS**

Ocular sensory circuits and lacrimation

David A Bereiter, Keiichiro Okamoto
U of MN School of Dentistry Minneapolis, MN, USA

The ocular surface receives a dense supply from trigeminal sensory nerves whose main function is to protect the eye via reflexes such as lacrimation. Loss of normal lacrimation due to eye surgery, infection or disease is critical aspect of eye injury and dry eye disease. Tears are controlled by a "lacrimal functional unit": afferent sensory nerves, CNS pathways and autonomic efferent nerves. The basis for sensory transduction in the eye is well defined; however, less is known about the CNS pathways necessary for reflex lacrimation. Ocular sensory nerves terminate in a unique bimodal pattern in caudal brainstem at the interpolaris/caudalis transition (Vi/Vc) transition and Vc/upper cervical cord (Vc/C1) junction regions. Vi/Vc and Vc/C1 ocular neurons share many properties such as encoding chemical irritant concentration, but differ in many other aspects such as sensing the moisture status of the ocular surface, plasticity after inflammation, and response to opioid analgesics. CO2-evoked tears are prevented by synaptic blockade of the Vi/Vc, but not the Vc/C1 region, while only Vc/C1 neurons are become sensitized by ocular inflammation. Intraocular tissues also receive a rich supply from trigeminal nerves; however, less is known about their role ocular homeostasis. Bright light activates ocular neurons at the Vi/Vc and Vc/C1 regions via a circuit of accessory visual nuclei and increased autonomic outflow to the eye and independent of corneal nerves. Synaptic blockade of Vi/Vc, but not Vc/C1, prevents light-evoked lacrimation. In summary, second-order ocular neurons at the Vi/Vc transition and Vc/C1 junction regions encode the intensity of a wide variety of conventional and unconventional sensory signals, yet each region likely plays a unique role in maintaining normal ocular function. Acknowledgements: NIH NS26137

#24 **SYMPOSIUM: NEW FRONTIERS IN CHEMESTHESIS**

Unusual Pungency from Extra-Virgin Olive Oil via Tissue Specific Expression of TRPA1 Channel in the Human Oro-Pharynx

Catherine Peyrot des Gachons[1], Kunitoshi Uchida[2,3], Bruce Bryant[1], Asako Shima[2], Jeffrey B. Sperry[4], Luba Dankulich-Nagrudny[1], Makoto Tominaga[2,3], Amos B. Smith, III[1,4], Gary K. Beauchamp[1], Paul A.S. Breslin[1,5]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]Okazaki Institute for Integrative Bioscience Okazaki , Japan, [3]Department of Physiological Sciences, Graduate University for Advanced Studies Okazaki, Japan, [4]Department of Chemistry, University of Pennsylvania School of Medicine Philadelphia, PA, USA, [5]Department of Nutritional Sciences, Rutgers University School of Environmental and Biological Sciences New Brunswick, NJ, USA

Oleocanthal, a major phenolic compound in extra-virgin olive oil elicits an unusual oral pungency sensed almost exclusively in the throat. This contrasts with most other common oral irritants, such as capsaicin and alcohol, which irritate mucus membranes throughout the oral cavity and elsewhere. In vitro, oleocanthal selectively activates the hTRPA1 channel in HEK 293 cells and its ability to excite the trigeminal nervous system in rodents requires a functional TRPA1. The over-the-counter analgesic, ibuprofen, which elicits the same restricted pharyngeal irritation as oleocanthal, also specifically excites rodent sensory neurons via TRPA1. Human sensory studies and immunohistochemical analyses of human oral and nasal tissues reveal a complete overlap of the anatomical distribution of TRPA1 in humans with the characteristic regions irritated by oleocanthal. TRPA1 immunoreactivity was present in nerve endings of the nose and pharynx but not the tongue. These observations suggest that TRPA1 mediates the upper airway tissue sensitivity to oleocanthal. They also suggest that some canonical TRPA1 agonists, such as mustard oil, which irritate the anterior tongue in humans, may do so via interactions with a TRPA1 variant or heteromer that does not react with our antibody. The pharynx and naso-pharynx are privileged tissues. Many bitter tasting toxins are perceived more strongly in the posterior oral cavity than the anterior. The enrichment of posterior oral tissues with toxin and irritant receptors enables them to guard against ingestion and inhalation of potentially harmful compounds by stimulating defensive reflexes such as coughing, throat clearing, and general rejection. Acknowledgements: NIH DC02995 and DC06760

#25 **SYMPOSIUM: NEW FRONTIERS IN CHEMESTHESIS**

Nasal solitary chemosensory cells link irritation to inflammation

Marco Tizzano, Thomas E. Finger
University of Colorado Denver/Rocky Mountain Taste and Smell Center, Department of Cell and Developmental Biology Aurora, CO, USA

Airways are continually assaulted by harmful compounds carried on the incoming airstream. The trigeminal nerve responds to such compounds as irritants and evokes protective reflexes, including sneezing and apnea. The airway epithelium houses a population of trigeminally innervated solitary chemosensory cells (SCCs) that express T2R taste receptors along with their downstream signaling components: Gα-gustducin and TrpM5. We have described that nasal SCCs are necessary to evoke trigeminally mediated respiratory reflex reactions to the T2R-ligand denatonium benzoate and to acyl–homoserine lactones (AHL), quorum-sensing molecules of Gram-negative bacteria (Gulbransen 2008, Tizzano 2010). These studies showed the necessity for SCCs in triggering respiratory depression to certain types of irritants. We investigated here the possibility that SCC activators, e.g. denatonium, also trigger an inflammatory and immune response in the nasal cavity. We exposed the nasal passageways to denatonium benzoate (20mM) and injected the animals intravascularly with fluorescently-conjugated albumin. Within 30 min., we detected leakage of plasma albumin into the nasal epithelium. In addition, we find that by 1 hr following stimulation with denatonium, the dendritic cells of the epithelium take on an activated appearance and appear to move from their basal resting position to lie in close contact with SCCs on the treated side, but not control side of the nose. Genetic deletion of either Gα-gustducin or TrpM5, essential elements of the T2R transduction cascade, eliminates the plasma leakage and the activation of the DCs. These findings indicate that activation of the SCCs can provoke local inflammatory responses in the nasal epithelium, both by activation of nearby dendritic cells and by neurogenic mechanisms. Acknowledgements: This study was supported by NIH grants NIDCD R01 DC006070 & P30 DC04657 (to D.Restrepo. & T.E.F.), and DC009820 (to T.E.F. & S.C. Kinnamon).

#26 **SYMPOSIUM: NEW FRONTIERS IN CHEMESTHESIS**

Bitter taste receptors on airway smooth muscle: signaling, function and therapeutic applications

Deepak A Deshpande
University of Maryland Baltimore/Medicine Baltimore, MD, USA

We identified the expression of bitter taste receptors (TAS2Rs) on human airway smooth muscle (ASM) cells. Real-time PCR and immunofluorescence studies revealed the presence of 17 different subtypes of TAS2Rs with varying levels of expression. Stimulation of ASM cells with known bitter agonists such as chloroquine, quinine, saccharin and denatonium resulted in an increase in inositol triphosphate production and intracellular calcium ($[Ca^{2+}]_i$) concentration, similar to other Gq-coupled receptor agonists such as histamine and bradykinin. Interestingly, bitter tastants relaxed human and murine airways and ASM cells, whereas Gq- coupled receptor agonists (that elevate $[Ca^{2+}]_i$) contracted human and murine airways. Relaxation of airways or ASM cells induced by bitter tastants was not associated with cyclic adenosine monophosphate production or protein kinase A activation, but was attenuated by pretreatment with ibiriotoxin, an inhibitor of large-conductance Ca^{2+}-activated K^+ channels. Stimulation of ASM cells with bitter tastants resulted in robust hyperpolarization which was reversed by ibiriotoxin. Confocal microscopic evaluation of $[Ca^{2+}]_i$ elevation by bitter tastants demonstrated large calcium increases in the localized regions beneath the plasma membrane. Finally, aerosol exposure of TAS2R agonists reversed the bronchoconstriction in both normal and asthmatic mice with a higher efficacy than the ß-adrenergic agonists (commonly used anti-asthma drugs). In summary, bitter tastants relax ASM by localized calcium elevation and activation of BK_{Ca} channels. Collectively, these studies identified a novel class of receptors on the ASM and their functional effects which could be exploited to develop a new class of bronchodilators for the treatment of obstructive pulmonary diseases such as asthma and COPD. Acknowledgements: HL087560, HL045967 and HL071609

#29 **SYMPOSIUM: EXPANDING THE CANONICAL VIEW OF SYNAPTIC PROCESSING IN THE OLFACTORY BULB**

Deconstructing the Glomerular Input-Output Function

Michael T Shipley
Department of Anatomy & Neurobiology, Program in Neuroscience, University of Maryland School of Medicine Baltimore, MD, USA

Odor signals from olfactory sensory neurons are initially processed in glomeruli where information is transferred to principal output neurons, mitral/tufted (MT) cells. MT cells are regulated by inhibition from GABAergic granule cells (GCs). However they also receive inhibitory and excitatory inputs from glomerular circuits. We examined the relative contributions of glomerular and infraglomerular regulation of mitral cells in bulb slices. External tufted (ET) cells provide excitatory input to MT cells, contributing to development of long lasting depolarizations. This disynaptic excitatory drive parallels direct monosynaptic input as demonstrated by synaptic latencies and ultrastructural identification of ON synapses on identified mitral cells.
In addition, glomerular circuits generate rapid onset, strongly summating postsynaptic inhibition of MT cells. The interval between sensory-evoked excitation and intraglomerular inhibition creates a 'window' for a brief burst of action potentials. The balance and timing of this excitation-inhibition sequence determines how long the window is open and thus gates the magnitude and duration of mitral cell spiking. Indeed, recent studies indicate that substantial odor encoding occurs in the initial phase of the sniff cycle. Glomerular circuits also generate strong tonic inhibition of MT cells and olfactory nerve terminals. Tonic inhibition is due to spontaneous bursting of ET cells, which drive PG cells generating sustained GABA release. Tonic pre- and postsynaptic intraglomerular inhibition filters weak sensory inputs and set a threshold for spike activation in MT cells. The combination of tonic and sensory-evoked intraglomerular inhibition exerts strong control of glomerular output to determine the impact of sensory signals on all downstream olfactory circuits.
Acknowledgements: Supported by NIH DCCD005676, DCCD19015

#30 **SYMPOSIUM: EXPANDING THE CANONICAL VIEW OF SYNAPTIC PROCESSING IN THE OLFACTORY BULB**

Neural circuitry between sensory input and output mitral cells in the mammalian olfactory bulb

N. E. Schoppa
Dept. of Physiology and Biophysics, University of Colorado School of Medicine.

A prevailing view of the mammalian olfactory system is that input signals are transferred directly from olfactory sensory neurons (OSNs) to mitral cells (MCs) in the olfactory bulb, which are then processed through lateral inhibitory interactions between different glomeruli. In our study, done in rodent olfactory bulb slices, we investigated alternate forms of signaling, beginning by testing the assumption of direct OSN-to-MC transmission. Using two methods of stimulating OSNs, either electrically or via light activation of OSNs specifically expressing channelrhodopsin, we found a striking absence of fast electrical signals in MCs that could be attributed to direct OSN-to-MC transmission. Instead, OSN signaling onto MCs appeared to be through a multi-step mechanism in which tufted cells were intermediaries (OSN-to-tufted cell-to MC). Tufted cells also appeared to play a key role in mediating inhibition of MCs, as periglomerular cell inputs targeted onto these cells suppressed MC activity via both intra- and inter-glomerular mechanisms. Based on our results, we propose that a network of interacting tufted cells and periglomerular cells lying between OSNs and MCs engages in much of the processing of olfactory information, and that MCs may largely act as simple conveyors of already-processed signals to the cortex.

#31 **SYMPOSIUM: EXPANDING THE CANONICAL VIEW OF SYNAPTIC PROCESSING IN THE OLFACTORY BULB**

Odour representation and synaptic inhibition in the mouse olfactory bulb

Izumi Fukunaga, Jan T. Herb, Andreas T. Schaefer
SNWG Behavioural Neurophysiology, Max-Planck-Institute for Medical Research

Inhibitory interneurons in the olfactory bulb (OB) are thought to play an important role in shaping neuronal representation of odours. Recently, we have shown that altering granule cell-mediated inhibition in behaving animals affects their performance in difficult odour discrimination tasks (Neuron 2010, 65:399). Mechanistically, inhibitory inputs to mitral and tufted (M/T) cells have been suggested to have diverse functions, including enhancing contrast between similar odours, maintaining invariance of responses to a given odour over different concentrations, establishing temporal coherence and improving precision in spike timing. To study the role of phasic inputs mediated by GABAA receptors in shaping M/T responses, we applied a combination of GABAA-receptor agonist and antagonist, gabazine and muscimol at high concentrations with the aim of occluding synaptic activation of GABAA receptors. Whole cell patch clamp recordings of M/T cells in anaesthetised mice were carried out with biocytin in the pipette solution to recover morphologies and allow for unambiguous identification of cell type. Combined application of gabazine and muscimol resulted in stable, non-epileptic recording conditions, and preserved spontaneous firing and the input resistance of neurons. The effectiveness of the drug treatment was seen in a significant reduction of phasic activation of GABAA receptors, as measured by recurrent inhibition evoked by depolarising current injection. Such blocking of phasic inhibition unmasked different types of signal transformations: for example, in cells that showed strong excitatory responses to an odour during control, blocking phasic inhibition resulted in the removal of action potential adaptation, whereas inhibitory "off-responses" were generally suppressed. In approximately one third of all odour-cell pairs (59/197) pure phasic inhibition was observed in response to odorants. In virtually all of these cases, occluding synaptic activation of GABAA receptors resulted in a substantial depolarisation upon odour presentation in a concentration-dependent manner. Furthermore, if odours elicited no response under control conditions, no change was observed upon application of gabazine and muscimol. If odour conentration was increased under control conditions, in some cases odours that elicited phasic inhibition at low concentrations reverted to excitatory responses for higher

odour concentrations. Finally, selective hyperpolarisation of granule cells through specific expression of NPHR2 and light application did not alter odour-evoked phasic inhibitions. Thus, these findings are consistent with a prominent role for feed-forward inhibition onto M/T in shaping odour representation in the mammalian olfactory bulb.

#32 **SYMPOSIUM: EXPANDING THE CANONICAL VIEW OF SYNAPTIC PROCESSING IN THE OLFACTORY BULB**

The Benefits of Biophysical Diversity in Olfactory Bulb Mitral Cells

Nathan N. Urban[1]
[1]Department of Biological Sciences, Carnegie Mellon University Pittsburgh, PA, USA, [2]Center for the Neural Basis of Cognition Pittsburgh, PA, USA

Neurons are highly diverse in their properties. Even neurons of the same molecular type have notable intrinsic differences. Largely unknown, however, is the degree to which these differences impair or assist neural coding. We examined how olfactory bulb mitral cells respond to identical stimuli and found that each cell's spiking response was dictated by its unique biophysical fingerprint. We then show how this intrinsic heterogeneity influences the amount of information that can be encoded by populations of mitral cells, the ease with which mitral cells can synchronize, and the level of correlation in mitral cell spike trains. Our results indicate that intrinsic neuronal diversity is important for neural coding and is not simply the result of biological imprecision. Acknowledgements: R01DC0005798, R01DC11184

#33 **SYMPOSIUM: EXPANDING THE CANONICAL VIEW OF SYNAPTIC PROCESSING IN THE OLFACTORY BULB**

Understanding neuronal circuits of the mammalian olfactory bulb

Dinu F Albeanu
Cold Spring Harbor Laboratory Cold Spring Harbor, NY, USA

In the mammalian olfactory bulb (OB), sensory neurons expressing the same type of olfactory receptor (~10,000) converge in tight focus, forming clusters of synapses called glomeruli (~2,000). From each glomerulus, a few dozen mitral cells (principal output neurons of the OB) carry the output further to the cortex. The mitral cells typically have only one primary dendrite that projects to a single glomerulus but can sample inputs on their primary and secondary dendrites from functionally diverse glomeruli via several types of interneurons. Thus, a few dozen sister mitral cells share input from the same parent glomerulus, but may have different inhibitory surrounds. We have recently shown that these sister cells are similar to each other in terms of average firing rate changes in response to odors but differ substantially in the timing of their firing and thus may relay non-redundant information to the olfactory cortex. These temporal differences in firing could be due to cross glomerular interactions at the input layer itself or lateral inhibitory inputs and processing at mitral cell level or even centrifugal feedbacks. We are now systematically investigating the excitatory and inhibitory connections in the bulb to further dissect these mechanisms and the contribution of local lateral inputs to the input-output function. We are using an intersectional Cre/Lox approach to express Arch (an inhibitor of neuronal activity) in specific subsets of interneurons and using patterned illumination to reversibly modulate their activity. The focus is on two largely non-overlapping populations of interneurons, genetically defined as parvalbumin and somatostatin positive respectively. We simultaneously monitor the firing properties of mitral cell pairs using either tetrode arrays or optical recordings via GCAMP.

#33.5 **IFF LECTURE**

Belling the Cat: Understanding Late Transduction Mechanisms in the Taste Bud

Scott Herness
College of Dentistry, The Ohio State University, Columbus, OH

In the last decade, remarkable progress has been made on the once seemingly intractable problem of understanding how a taste receptor cell responds to taste stimuli. Groundbreaking experiments in the molecular biology of the taste bud have elucidated distinct receptors and associated downstream transduction cascades for all five taste qualities, each quality surprisingly transduced by a separate subpopulation of cells within the bud. Conducted in parallel with these studies were others directed at understanding the nature and identity of the neurotransmitter between the taste receptor cell and the afferent nerve. These results initially produced confusion when multiple signaling agents were discovered but eventually made clear that extensive cell-to-cell communication occurs among the cells of the bud. Collectively, these two approaches have led to the classification of transduction mechanisms into early and late events. Early transduction mechanisms refer to those events occurring between stimulus-induced receptor activation and the resultant depolarization of the taste receptor cell. Late transduction mechanisms, on the other hand, refer to the complex network of excitatory and inhibitory cell-to-cell communication pathways within the bud evoked after the initial depolarization. These events are mediated by multiple neurotransmitters, neuropeptides, and the corresponding receptors that are expressed in both autocrine and paracrine manners. They participate in the processing of gustatory information in manners such as gain modulation, lateral inhibition, and adaptation of the final afferent neural discharge. Hence the activity of single taste receptor cells is influenced not only through apical receptor activation but also through basolateral receptors. At present, late transduction events are only partially understood. With the exceptional progress in understanding early transduction, extracting how these late transduction hardwired pathways participate in the transduction cascades of taste quality processing is fast becoming the field's next "seemingly intractable" problem.

#34 PLATFORM PRESENTATIONS - POLAK YOUNG INVESTIGATOR AWARD WINNERS

Loss-of-function mutations in sodium channel Nav1.7 cause anosmia

Jan Weiss[1], Martina Pyrski[1], Eric Jacobi[1], Bernd Bufe[1], Vivienne Willnecker[2], Bernhard Schick[2], Philippe Zizzari[3], Samuel J. Gossage[4], Charles A. Greer[5], Trese Leinders-Zufall[1], Geoffrey Woods[6], John N. Wood[4], Frank Zufall[1]
[1]Department of Physiology, University of Saarland School of Medicine Homburg, Germany, [2]Department of Otolaryngology, University of Saarland School of Medicine Homburg, Germany, [3]Centre de Psychiatrie & Neurosciences, UMR 894 Inserm, Faculté de Médecine, Université Paris Descartes Paris, France, [4]Molecular Nociception Group, Wolfson Institute for Biomedical Research, University College London London, United Kingdom, [5]Department of Neurosurgery, Yale University School of Medicine New Haven, CT, USA, [6]Department of Medical Genetics, Cambridge Institute for Medical Research Cambridge, United Kingdom

Loss of function of the gene *SCN9A*, encoding the voltage-gated sodium channel Nav1.7, causes congenital inability to experience pain in humans. Here, we show that Nav1.7 is not only necessary for pain sensation but is also an essential and nonredundant requirement for odour perception in both mice and humans. We examined human *SCN9A* loss-of-function mutants and provide clear evidence that they are anosmic. To establish the essential role of Nav1.7 in odour perception, we generated conditional null mice in which Nav1.7 was removed from all olfactory sensory neurons (OSNs) and combined gene expression studies with cellular electrophysiological and behavioural analyses. We found that Nav1.7 occupies a critical location in the olfactory pathway, in the olfactory nerves and glomeruli, presynaptically to the first synapse in the olfactory system. To examine whether presynaptic activity of Nav1.7 underlies transmitter release in the olfactory glomerulus, we prepared acute olfactory bulb tissue slices and combined focal electric stimulation of the olfactory nerve layer with whole-cell patch-clamp recording of visually identified mitral/tufted (M/T) cells. In the absence of Nav1.7, OSNs still produce odour-evoked action potentials but fail to initiate synaptic signaling from OSN terminals to M/T cells. The mutant mice no longer show a wide range of vital, odour-guided behaviours including innate recognition of food and species-specific odours, odour discrimination and short-term odour learning, innate avoidance toward a predator odour, and maternal pup retrieval. Our study creates a mouse model of congenital general anosmia and offers new strategies to unravel the genetic basis of the human sense of smell. Acknowledgements: This work was supported by grants from the Deutsche Forschungsgemeinschaft, Volkswagen Foundation, BBSRC MRC Wellcome Trust and grant number No. R31-2008-000-10103-0 from the WCU project of the MEST and the NRF.

#35 PLATFORM PRESENTATIONS - POLAK YOUNG INVESTIGATOR AWARD WINNERS

Residual Chemoresponsiveness to acids in the superior laryngeal nerve in "tasteless" (P2X2/P2X3 double KO) mice

Tadahiro Ohkuri[1], Nao Horio[1], Thomas E. Finger[2], Yuzo Ninomiya[1]
[1]Dept. Oral Neuroscience, Graduate School of Dental Sciences, Kyushu University Fukuoka, Japan, [2]Rocky Mountain Taste & Smell Center, University of Colorado School of Medicine, Aurora, CO, USA

Mice lacking both P2X2 and P2X3 purinergic receptors exhibit nearly total lack of responses to all taste qualities in both the glossopharyngeal and chorda tympani nerves. Similarly, these mice exhibit near total loss of taste-related behaviors (increased intake for appetitive tastants; decreased intake of bitter tastants) except for a near-normal avoidance of acidic stimuli (Finger 2005; Hallock 2010). The persistence of avoidance of acids despite the loss of gustatory neural responses to sour, was postulated to be due to remnant responsiveness of the superior laryngeal nerve (SLN) which innerves the epithelium and taste buds of the larynx. Using immunohistochemistry, we confirm that a substantial population of peptidergic nerve fibers (presumably capsaicin-sensitive polymodal nociceptors) richly innervate the laryngeal epithelium. Chemoresponses of the larynx are attributable both to taste buds and to capsaicin receptors on the laryngeal nerve fibers (Arai et al 2010). In order to test whether the SLN of the P2X2/P2X3 dbl KO mice remains responsive to acids but not to other tastants, we recorded from the SLN in WT and dbl KO mice. The WT mice showed substantial responses to MSG (300 - 1000mM), sucrose (100 - 1000mM), Urea (300mM), and Denatonium (10mM) all of which were essentially absent in the P2X dbl KO animals. In contrast, the KO mice exhibited near-normal responses of the SLN to citric acid (50mM) although responses to acids ("sour") in both the chorda tympani and glossopharyngeal nerves were nearly absent. These results are consistent with the hypothesis that the residual avoidance of acidic solutions by P2X dbl KO mice may be attributable to the direct chemosensitivity of nerve fibers innervating the laryngeal epithelium, and not to taste. Acknowledgements: Supported by NIH Grants (5R01 DC007495 and 5P30 DC04657) to T.E.F. and by KAKENHI 18109013, 18077004 to Y.N.

#36 PLATFORM PRESENTATIONS - POLAK YOUNG INVESTIGATOR AWARD WINNERS

Cholinergic chemosensory cells in the trachea regulate breathing

Gabriela Krasteva[1], Brendan J Canning[2], Petra Hartmann[1], Tibor Z Veres[3], Tamara Papadakis[1], Christian Mühlfeld[1], Schliecker Kirstin[1], Yvone Y Tallini[4], Armin Braun[3], Holger Hackstein[5], Nelli Baal[5], Eberhard Weihe[6], Burkhard Schütz[6], Ines Ibanez-Tallon[7], Michael I Kotlikoff[4], Wolfgang Kummer[1]
[1]Justus-Liebig-University/Institute of Anatomy and Cell Biology Giessen, Germany, [2]2Johns Hopkins Asthma and Allergy Center Baltimore, MD, USA, [3]3Fraunhofer Institute for Toxicology and Experimental Medicine Hannover, Germany, [4]College of Veterinary Medicine/Dept. of Biomedical Sciences Ithaca, NY, USA, [5]Justus-Liebig-University/Institute for Clinical Immunology and Transfusion Medicine Giessen, Germany, [6]Philipps-University Marburg/Institute for Anatomy and Cell Biology Marburg, Germany, [7]Max-Delbrück-Centre for Molecular Medicine Berlin, Germany

Brush cells are suspected to serve a chemosensory function. Here, we investigated the possibility that brush cells in the mouse trachea produce acetylcholine (ACh). Using mice expressing eGFP under the control of the promoter of the ACh synthesizing enzyme, choline acetyltransferase (ChAT), we identified solitary cholinergic cells in the mouse tracheal epithelium as brush cells by their immunoreactivity for villin and their characteristic ultrastructure. They also expressed the vesicular ACh transporter and proteins of the taste transduction pathway (α-gustducin and $PLC_{\beta 2}$). Messenger RNA for taste receptor 105 involved in perception of the bitter substance cycloheximide was detected in ChAT-eGFP cells isolated by FACS. CLSM-analyses revealed direct contacts with CGRP-immunoreactive nerve fibres. Using another transgenic mouse model that expresses eGFP under the control of the promotor for the a3-subunit of the nicotinic ACh receptor, we identified a subpopulation of C-fibres as cholinoceptive. Retrograde neuronal tracing identified airway-projecting sensory neurons with this chemical coding in the jugular-nodose-complex and in cervico-thoracic DRG. Respiratory pattern was measured after tracheal stimulation in a newly established model in spontaneously breathing anesthetized mice. DMPP, a nicotinic agonist, caused a drop in respiratoty rate which was augmented by inhibition of nicotinic receptors with mecamylamine, a nicotinic receptor antagonist. Cycloheximide elicited an epithelium-dependent drop in respiratory rate which was abolished by pretreatment with mecamylamine (p=0.019). We conclude that tracheal brush cells are chemosensory, cholinergic cells that transmit changes in the luminal microenvironment of the airways to the CNS via ACh release and nicotinic stimulation of sensory neurons. Acknowledgements: This work was supported by a Young Investigator Grant from the Medical Faculty of the Justus-Liebig-University (GK), Giessen, Germany, Universities of Giessen & Marburg Lung Center (WK), von Behring-Röntgen-Stiftung (GK), and the Deutsche Forschungsgemeinschaft (WK). BJ Canning was supported by a grant from the National Institutes of Health USA (HL083192).

#37 PLATFORM PRESENTATIONS - POLAK YOUNG INVESTIGATOR AWARD WINNERS

The significance of convergent inputs from olfactory receptor neurons to the second-order neuron on olfactory processing in the *Drosophila* antennal lobe

Vikas Bhandawat[1], Rachel Wilson[2]
[1]Duke University Durham , NC, USA, [2]Harvard Medical School Boston, MA, USA

A conserved feature of olfactory systems across the animal kingdom is the large convergence from the olfactory receptor neurons (ORNs) to the second-order neuron. What is the role of this convergence in olfactory processing? It is usually assumed that this large convergence allows the olfactory system to increase its sensitivity to odors, but empirical data is lacking. How does the odor sensitivity of an animal depend upon the convergence ratio between the ORNs and second order neurons? More generally, how does information transmission from the first-order neuron to the second-order neuron depend on the convergence ratio? We ask this question in the context of the fly olfactory system, which offers the experimental advantages of genetic accessibility, an organized anatomy, and a quantifiable pool of ORNs. Each second-order neuron (called projection neuron or PN) receives input from a population of ~60 ORNs. In this study, we genetically reduced the convergence ratio between the ORNs and PNs and asked how the convergence ratio affects the strength of PN responses as well as the trial-to-trial variability in the PN responses. We found that convergence ratio had a dramatic effect on the strength of the odor responses in the PNs, but had surprisingly little effect on the trial-to-trial variability in PN responses. We will discuss the circuit mechanism that underlies the dependence of PNs response on the convergence ratio. We will also discuss the implication of these findings on the detection threshold for odors, as well as, its importance in olfactory processing. Acknowledgements: Charles King Fellowship (VB) NIH (R01DC008174)(RW) McKnight Scholar Award(RW) Sloan Foundation Research Fellowship (RW) Beckman Young Investigator Award (RW)

#38 PLATFORM PRESENTATIONS - POLAK YOUNG INVESTIGATOR AWARD WINNERS

The Piriform Cortex Utilizes Different Microcircuits to Process Cortical and Amygdaloid Synaptic Inputs

Victor M Luna
NIH Bethesda, MD, USA

The posterior piriform cortex (pPC) integrates synaptic inputs from a variety of cortical and subcortical structures to form olfactory representations. To do this, the pPC must theoretically be able to delineate the anatomical origin of incoming synaptic signals in order to assess their computational value. However, it is not known if and how the pPC performs this function. To address this issue, mouse anterior piriform cortex (aPC; a source of cortical input) or basolateral amygdala (BLA; a source of subcortical input) were infected with adenoassociated virus expressing channelrhodopsin-2. The aPC and BLA were chosen because they send disparate types of information to the pPC, with the BLA sending inputs necessary for forming emotionally-charged percepts (e.g. fear-based learning). Combining optogenetics with whole-cell electrophysiology allowed for photostimulation of either aPC or BLA fibers only and recording of evoked excitatory postsynaptic currents (EPSC) in pyramidal cells (PYR) and interneurons (IN) in pPC slices. Using strength of EPSCs as a measure of functional connectivity, it was found that aPC and BLA fibers activated different pPC neuronal ensembles. aPC fibers excited all pPC neurons with Layer 2/3 (L2/3) PYRs and INs receiving the greatest amounts of excitation. In contrast, BLA fibers were more selective, evoking EPSCs preferentially on certain subtypes of L3 INs while providing very little excitatory drive to most PYRs and INs. Thus, the pPC is able to extract and delineate information conveyed by aPC versus BLA by processing their synaptic inputs through different microcircuit channels. This mechanism allows the pPC to assign computational value to incoming synaptic signals based on their anatomical origin and to generate unique outputs for each distinct type of afferent input.

#39 PLATFORM PRESENTATIONS - POLAK YOUNG INVESTIGATOR AWARD WINNERS

Does Free Glutamate in Infant Formula Promote Satiation and Satiety?

Alison K Ventura, Sehris Khawaja, Gary K Beauchamp, Julie A Mennella
Monell Chemical Senses Center Philadelphia, PA, USA

Striking discrepancies exist for the free amino acid profiles of the types of formulas most commonly fed to infants. We recently discovered that infants randomized to feed a formula high in free amino acids (extensive protein hydrolysate formula [PHF]) during the first 7 months of life consumed less formula to satiation and gained less weight across the study period compared to infants fed an isocaloric formula low in free amino acids (cow milk formula [CMF]). Also, the weight gain trajectory of PHF-fed infants resembled that of breast-fed infants whereas CMF-fed infants had an accelerated trajectory. One explanation for these findings is that certain free amino acids (e.g., glutamate) stimulate sensory receptors in the gastrointestinal tract to signal satiation. The present study tested this hypothesis by examining whether the higher level of free glutamate in PHF compared to CMF (7472 vs. 109g/100kcal) promotes greater satiation and satiety. To this end, 1- to 3-month-old infants were observed feeding two formula meals on three separate test days. In counterbalanced order, infants fed CMF, PHF, or CMF with added free glutamate to satiation during the first meal. When infants signaled hunger again, they were fed CMF until satiation. Primary dependent measures included volume of formula consumed during the first meal and the satiety ratio (interval between first and second meal divided by amount consumed at the first meal). Preliminary analyses revealed infants consumed less and were satiated longer when they fed the formulas containing higher levels of free glutamate when compared to CMF. Results from this study suggest that free glutamate is at least partially responsible for the satiating effects of PHF and call into question the claim that formula-fed infants cannot self-regulate energy intake.
Acknowledgements: The project described was supported by a grant from Ajinomoto, Inc. and grants HD37119 and 1F32HD063343-01A1 from the Eunice Kennedy Shriver National Institute of Child Health and Human Development. The content is solely the responsibility of the authors and does not necessarily represent the official views of the Eunice Kennedy Shriver National Institute of Child Health and Human Development or the National Institutes of Health.

#41 SYMPOSIUM: IONOTROPIC SENSORY RECEPTORS

The molecular basis for water taste in Drosophila

Peter Cameron, Makoto Hiroi, John Ngai, Kristin Scott
Department of Molecular and Cell Biology and Helen Wills Neuroscience Institute, University of California, Berkeley, Berkeley, CA

The detection of water and the regulation of water intake are essential for animals to maintain proper osmotic homeostasis. Drosophila and other insects have gustatory sensory neurons that mediate the recognition of external water sources, but little is known about the underlying molecular mechanism for water taste detection. We identified a member of the Degenerin/Epithelial Sodium Channel family, ppk28, as an osmo-sensitive ion channel that mediates the cellular and behavioral response to water. We use molecular, cellular, calcium imaging and electrophysiological approaches to show that ppk28 is expressed in water-sensing neurons, loss of ppk28 abolishes water sensitivity, and ectopic expression of ppk28 confers water sensitivity to bitter-sensing gustatory neurons in the fly and to heterologous cells. These studies link an osmo-sensitive ion channel to water taste detection and drinking behavior, providing the framework for examining the molecular basis for water detection in other animals.

#42 SYMPOSIUM: IONOTROPIC SENSORY RECEPTORS

The TRP channel painless mediates gustatory DEET sensation in Drosophila melanogaster

Jason C. Caldwell, Yifan Xu, Allison D. Weaver, W. Daniel Tracey
Departments of Anesthesiology, Cell Biology, and Neurobiology. Duke University, Durham, NC, USA

Individual insect gustatory receptor neurons are typified by their physiological responses to sweet, bitter, salty, carbonation or water. The precise taste molecules that activate a particular neuron are thought to be determined by the precise ensemble of seven transmembrane receptor gustatory receptor proteins expressed in the sensory dendrite. In the case of Drosophila bitter receptor neurons, the TRPA family ion channels Painless and dTRPA1 are also expressed. Evidence suggests that expression of dTRPA1 and Painless in these cells plays an important role in the detection and avoidance of noxious isothiocyanate (ITCs) compounds. Similarly, mammalian TRPA1 is a receptor for ITCs and other noxious compounds in nociceptor neurons. Here, we show that the painless gene is also required for avoidance of the insect repellent DEET. Drosophila mutant for painless show a complete failure to avoid DEET and in some cases are actually attracted to this normally repellent compound. In addition, we have cloned the painless orthologue from the malaria vector Anopheles gambiae. Transgenic Drosophila expressing this mosquito painless gene in a painless mutant background functionally rescued for DEET avoidance. Our results suggest that the Painless protein is an evolutionarily conserved component that is used by insects in the detection of the broad spectrum chemical repellent DEET. The Painless channel thus represents an ideal target for the future development of novel insect repellent compounds.

#43 SYMPOSIUM: IONOTROPIC SENSORY RECEPTORS

Sour sensations: a matter of taste and pain

Emily Liman
USC/Neurobiology Los Angeles, CA, USA

Of the five tastes, sour is perhaps the most mysterious. Sour is evoked by severely acidic substances, which activate both taste and pain pathways, and are generally aversive. Previous work has established that a subset of taste cells that express the ion channel PKD2L1 mediate sour taste, while sensory neurons in the trigeminal ganglion mediate the pain response. However, the ionic mechanisms that generate either sensory response are incompletely understood. Using mice genetically engineered to express YFP under the PKD2L1 promoter, we now show that a

proton conductance specific to the PKD2L1 expressing cells is sufficient to drive action potentials and Ca^{2+} elevation in response to acid stimuli. This mechanism is in contrast to the mechanism of acid sensing in the pain system, where gating by extracellular and intracellular protons of Na+ permeable channels contributes to the sensory response. We hypothesize that for sour taste transduction, a proton channel, as opposed to a proton-gated Na^+ channel, would be able to track changes in the concentrations of acids within the oral cavity, without confounding contributions from Na^+ ions which vary widely in concentration and are detected separately as saltiness Acknowledgements: NIH R01DC004564

#44 **SYMPOSIUM: IONOTROPIC SENSORY RECEPTORS**

Ionotropic Mechanoreceptors

M. B. Goodman
Department of Molecular and Cellular Physiology, Stanford University School of Medicine, Stanford, CA

The ability to detect touch is conserved from echinoderms to humans. It relies on specialized mechanoreceptor neurons that vary in their sensitivity and association with accessory structures. Despite its importance and conservation across taxa, very little is known about how touch works. We seek to improve understanding by studying the nematode C. elegans, a simple animal that has only 30 mechanoreceptor neurons. Our work focuses on two classes of mechanoreceptor neurons: the 6 non-ciliated touch receptor neurons (TRNs) that detect touch applied to the body wall and the paired ciliated ASH neurons that detect noxious mechanical stimuli applied to the nose. Genetic analysis has revealed ion channel genes needed for TRN and ASH function. To learn the precise cellular function of such channel proteins and to investigate their gating mechanisms, we combine genetic dissection with in vivo electrophysiology and biomechanical analysis. The picture emerging from our recent work is that touch activates closely related ionotropic receptors in the nonciliated TRNs and the ciliated ASH neurons, which differ by 100-fold in their sensitivity to external force. Challenges for the future include understanding the basis for differences in sensitivity and the biophysics of mechanotransduction channel gating. Key Words: C. elegans, mechanotransduction, ion channels, electrophysiology

#45 **SYMPOSIUM: IONOTROPIC SENSORY RECEPTORS**

Piezo proteins are essential components of mechanically-activated cation channels

Bertrand Coste[1], Ardem Patapoutian[1,2]
[1]The Scripps Research Institute, Department of Cell Biology La Jolla, CA, USA, [2]Genomics Institute of the Novartis Research Foundation San Diego, CA, USA

Mechanical stimuli drive many physiological processes, including touch and pain sensation, hearing, and blood pressure regulation. Mechanically-activated (MA) cation channel activities have been recorded in many cells, but the responsible molecules have not been identified. Using reverse genetic approach, we have identified a two member family of genes that are required for the expression of MA currents. Piezo1 (*Fam38A*) and related Piezo2 (*Fam38B*) are vertebrate multipass transmembrane proteins with homologs in invertebrates, plants, and protozoa. Overexpression of mouse Piezo1 or Piezo2 induced two kinetically-distinct MA currents. Piezos are expressed in several tissues, and knockdown of Piezo2 in dorsal root ganglia neurons specifically reduced rapidly-adapting MA currents. We propose that Piezos are components of mechanically-activated cation channels. Acknowledgements: This work was supported by grants from NIH and the Novartis Research Foundation. B. Coste was the recipient of an American Heart Association postdoctoral fellowship.

#45.5 **CLINICAL LUNCHEON**

The Sentinel Function of the Chemical Senses in Health & Disease

Pamela Dalton, PhD, MPH
Monell Chemical Senses Center

Just as the gustatory system can signal to alert and protect us from the ingestion of toxins, the chemosensory systems in the nose and upper airways serve a critical protective role — not only to detect but also to detoxify many inhaled exogenous pollutants. However, unlike the gustatory system which can be relatively robust to damage, the capacity of the chemosensory systems in the airways can be overwhelmed by significant acute or chronic exposures. Exposure to many airborne pollutants, induces inflammation and morphological changes that alter olfactory and nasal trigeminal sensitivity. This talk will review clinical studies in pollutant-exposed populations, including those exposed on 9/11 and beyond at the World Trade Center, and discuss the implications for the observed changes in chemosensory function.

#47 **SYMPOSIUM - ODOR-BASED SOCIAL BEHAVIOR IN MAMMALS: SIGNALS, BRAIN AND BEHAVIOR**

Olfactory sensing via immune receptors

Ivan Rodriguez
University of Geneva Geneva, Switzerland

Mammalian species rely heavily on olfaction to adequately interact between individuals. At the core of this sensory system are large superfamilies of G-coupled receptors that are present on dendrites of main olfactory and vomeronasal sensory neurons. These receptors define the agonist profile of sensory neuron populations, and thus define specific neural circuits, some of which are highly specialized. We recently uncovered a family of formyl peptide receptor-like, whose members are exclusively expressed by vomeronasal sensory neurons. Their corresponding genes are characterized by monogenic transcription and a punctate expression pattern in the sensory neuroepithelium. In vitro expression of these receptors provides sensitivity to disease/inflammation-related ligands. Finally, axons emanating from neurons expressing the same formyl peptide receptor-like converge in the accessory olfactory bulb, where they form a specific topographical projection map. Taken together, these data suggest the existence of a specific olfactory circuit in rodents, potentially involved in the identification of pathogenic states, or in the discrimination of pathogens.

#48 **SYMPOSIUM - ODOR-BASED SOCIAL BEHAVIOR IN MAMMALS: SIGNALS, BRAIN AND BEHAVIOR**

The functional organization of the accessory olfactory bulb
Timothy E. Holy
Washington University in St. Louis School of Medicine/Anatomy & Neurobiology St. Louis, MO, USA

The mouse accessory olfactory system specializes in detecting and analyzing chemical signals produced by other animals. Its sensory neurons, residing in the vomeronasal organ, detect a variety of cues that range from small molecules to sizable peptides. From analyses of an important natural stimulus urine from female mice we identified a family of highly active compounds, the sulfated steroids. Recently, we have made considerable progress in understanding how sulfated steroids are represented by the collective activity of populations of vomeronasal sensory neurons. This talk will focus on the next stage of the pathway, the accessory olfactory bulb. Using a combination of calcium imaging and electrophysiology, we are taking the first steps to elucidate the spatial and functional organization of the accessory olfactory bulb. We have found that most sulfated steroids activate multiple glomeruli in the anterior region. Glomeruli responding to one ligand frequently display striking co-localizations with glomeruli detecting other compounds. Post-synaptic to the glomeruli, the majority of steroid-responsive neurons can be well-described as receiving excitatory input from a selective subset of sensory neurons, arguing against widespread "random" wiring in the accessory olfactory bulb. Acknowledgements: NIH-NIDCD R01 DC005964 NIH-NIDCD R01 DC010381 Mathers Foundation

#49 **SYMPOSIUM - ODOR-BASED SOCIAL BEHAVIOR IN MAMMALS: SIGNALS, BRAIN AND BEHAVIOR**

Darcin: a pheromone that stimulates innate and learned sexual attraction in mice
Jane L. Hurst[1], Sarah A. Roberts[1], Robert J. Beynon[2]
[1]Mammalian Behaviour & Evolution Group , [2]Protein Function Group, Institute of Integrative Biology, University of Liverpool, Liverpool, UK

Scents play an integral role in mediating reproductive interactions in many mammals, allowing animals to recognize and locate individual conspecifics of the opposite sex and to assess the attractiveness of potential mates. In common with many other species, male mice advertise their location and competitive dominance through urinary scent marks deposited around defended territories. These urine marks contain a high concentration of major urinary proteins (MUPs). We show that an atypical male-specific MUP, which we named darcin, elicits the highly repeatable innate sexual attraction of female mice to spend time near male urinary scent marks. By contrast, females fail to show innate sexual attraction towards other MUPs or to airborne volatiles from male urine. Contact with darcin also stimulates strong and rapid associative learning of volatiles in an individual male's urinary scent, such that females are subsequently attracted to the airborne urinary odor of that particular male but not to airborne odors of other males. Thus, darcin allows female sexual attraction to be innate but also selective towards individual males. The airborne odor that females learn is determined in part by the individual-specific pattern of MUPs expressed in a male's urine. These proteins bind low molecular weight hydrophobic urinary volatiles and slow their release from scent marks. Manipulation of an individual male's MUP pattern changes the airborne volatiles learned, but a male's urine must contain the pheromone darcin to stimulate such learning. Darcin exemplifies a pheromone that can drive the flexible individual-specific social responses that are typical of mammals.

#50 **SYMPOSIUM - ODOR-BASED SOCIAL BEHAVIOR IN MAMMALS: SIGNALS, BRAIN AND BEHAVIOR**

Mechanisms of olfactory-regulated stereotypic behavior
Lisa Stowers
The Scripps Research Institute/Department of Cell Biology La Jolla, CA, USA

We are studying how subsets of neurons in the olfactory system specify behavior. Pheromone ligands activate dedicated subsets of neurons to generate instinctive behavior which provides a powerful experimental system to study neural function. Our approach is unique in that we have developed quantifiable analysis of the mouse's natural behavior. As biochemical assays are used to map and elucidate metabolic pathways, we use innate behavior as a functional assay to identify the corresponding ligand cues and cognate neurons that generate behavior. We are directing our experiments to identify underlying neural mechanisms that encode behavior using molecular probes, genetic manipulation, and functional imaging. We expect that restricting our investigation to a single behavior will enable us to identify the neural code for that behavior. Ultimately, however, that approach would limit our ability to define the unique neural features of one behavior from the common mechanisms of all behavior promoting circuits. Therefore, we are identifying the ligands that direct four different innate behaviors in order to clearly define general neural that enable the olfactory system to regulate stereotyped behavior. Our behaviors of interest include aggression, fear, mating, and pup-suckling. We expect this focused multi-circuit strategy to advance our understanding of the neural principles that generate behavior. Acknowledgements: NIH-NIDCD, Skaggs Foundation

#51 **SYMPOSIUM - ODOR-BASED SOCIAL BEHAVIOR IN MAMMALS: SIGNALS, BRAIN AND BEHAVIOR**

How Mammalian Females Olfactorily Broadcast the Source of Milk to their Offspring
Benoist Schaal
CNRS-CSGA Dijon, France

Neonatal mammals are exposed to a ruthless selective skimming at birth. Therefore, any way to reduce their energy expenditure and hasten their acceptance to ingest milk may have been gainful over evolutionary time. Thus, it is essential for mammalian, including human, females to display a cutaneous interface structure that is sensorily conspicuous and executively feasible for their fragile and inexperienced newborns. The females' strategy to increase the conspicuousness of nipples could only exploit the newborns' most functionally advanced sensory systems, *viz*. somesthesis and olfaction. In all mammals (except monotremes), selection has accordingly shaped tactilely/olfactorily obvious

mammary structures (*papillae*, nipples or teats). This evolutionary trend has worked either by mustering odor-producing structures or processes on, or close to, mammaries, or indirectly by relying on maternal behavioral propensities to secondarily create olfactory traces on them. These predictions will be surveyed in some mammalian representatives that have received empirical attention among marsupials, rodents, lagomorphs, ungulates, carnivores and primates. It appears that broadcasting chemical cues and/or signals from the mammae is a pan-mammalian reproductive strategy to control arousal in neonates, drive their attention and attraction to the mother, provide precise localizatory guidance toward, motivate oral grasping of, and sucking on, the papilla, and finally to boost up rapid learning. However, the ways by which these mammary chemical cues are produced or assembled are highly complex within species and diverse between species, offering an interesting ground for comparative analyses in olfactory communication and perceptual development. Acknowledgements: Supported by grants from CNRS and the Council of Region Bourgogne.

#52 **SYMPOSIUM - ODOR-BASED SOCIAL BEHAVIOR IN MAMMALS: SIGNALS, BRAIN AND BEHAVIOR**

Olfaction regulation of maternal behavior in sheep

Frederic Levy
'Behavior, Neurobiology, Adaptation' Unit, INRA Nouzilly, France

In mammals, olfactory cues are extensively used in many aspects of maternal care to ensure the coordination of mother-infant interactions and consequently the normal development of the offspring. Outside the period of parturition and lactation, non-pregnant females find the odor of young aversive. On the contrary at the time of parturition, a shift in the hedonic value of infantile odors occurs so that the young now become a very potent stimulus and this sensorial processing constitutes an important part of the maternal motivational system. In sheep, olfactory cues provided by the amniotic fluids are highly attractive to the parturient female and initiate the onset of licking and the full display of maternal behaviour. The paraventricular nucleus of the hypothalamus and the medial preoptic area through activation of the oxytocinergic system are key structures for regulating the expression of all components of maternal responsiveness. Moreover, infants' odors provide a basis for individual recognition by their mothers and some species (ungulates, humans) have developed highly specialized mechanisms for processing of the infant signals. In sheep, the selective recognition process in mother relies on odor cues from the own lamb, is mediated by the main olfactory bulb (MOB), and is rapidly established after 4 hours of mother-young contact. Extensive electrophysiological and neurochemical changes occurring in the MOB are part of the learning mechanisms; in particular noradrenaline release is essential for selective recognition of lambs. In addition, neurogenesis in the MOB is down-regulated by the olfactory learning process. Finally, the basal forebrain cholinergic system and the cortical and medial amygdala are involved in lamb olfactory recognition. Acknowledgements: This work was financially supported by INRA PHASE division and the PTR Institute Pasteur/INRA N°319.

#54 **SYMPOSIUM: BASIC TASTES: WHY FIVE?**

Are Free Fatty Acids Effective Taste Stimuli in Humans?

Richard D. Mattes, Bhushan V. Kulkarni, Robin M. Tucker
Purdue University/Foods and Nutrition West Lafayette, IN, USA

There is little controversy that dietary fats may be detected by visual, tactile, olfactory and auditory cues, but uncertainty remains for taste. Principle concerns focus on the origin of the effective stimulus, presumed to be free fatty acids (FFA)), difficulty in isolating taste from somatosensory cues (FFA contribute tactile and irritant cues) and lack of a clear lexicon for the sensation (so questions persist about whether it can be described by other primary taste qualities). However, accumulating evidence on these and other facets of the issue supports a taste contribution to fat detection. First, recent studies document unequivocally, that oral process of an array of fat-containing foods varying in physical form promotes micromolar concentrations of FFA in saliva. Thus, the purported effective signal is present under physiological conditions. Second, detection thresholds have been reliably measured for a variety of FFA in humans while controlling, albeit not perfectly, non-gustatory cues. Third, much like MSG, graded FFA concentrations can be monotonically scaled for intensity and sensations are not described by other common taste qualities. Fourth, multiple putative transduction mechanism have been identified, some in human lingual tissue. Studies are currently underway comparing FFA thresholds on oral gustatory and non-gustatory tissue as one test of their involvement. Fifth, gustatory nerve cuts in animal models impair behavioral responses to dietary fats. Sixth, neuro-imaging studies support fat detection independent of its tactile cues. Finally, a wide array of fat-specific, physiological responses are evoked by oral fat exposure, revealing a role of oral FFA signaling on lipid metabolism. Collectively, these data are consistent with a taste component for FFA. Acknowledgements: USDA HATCH Grant #IND030455H

#55 **SYMPOSIUM: BASIC TASTES: WHY FIVE?**

Calcium-Specific Taste

Stuart A. McCaughey[1], Michael G. Tordoff[2]
[1]Ball State University Muncie, IN, USA, [2]Monell Chemical Senses Center Philadelphia, PA, USA

Calcium is the most abundant mineral in the body and is essential for a wide range of important physiological events. It has been well-established that animals are able to adjust their intake of calcium based on need. Formerly, it was thought that this behavioral regulation was based entirely on learning about post-ingestive effects. However, our work suggested that the sense of taste is important for directing calcium-deprived rats toward their needed ion. These studies revived important issues related to the taste of calcium; namely, whether "calcium-like" represents a unique taste quality distinct from other prototypical tastes, and, if so, what mechanisms are responsible for the gustatory transduction of calcium. There is now converging evidence from several species that $CaCl_2$ has a unique taste and can be distinguished from most non-calcium compounds, with the possible exception of $MgCl_2$, on the basis of taste alone.

Recently T1R3 has been implicated as a taste receptor for calcium. Another proposed receptor is the calcium-sensing receptor (CaSR), which is found in taste tissue and is known to monitor calcium levels elsewhere in the body. Knowledge of the receptor mechanisms for calcium taste may lead to improvements in the palatability of high-calcium foods, which tend to be under-consumed by most people.

#56 **SYMPOSIUM: BASIC TASTES: WHY FIVE?**

Water as an Independent Taste Modality: An Old Idea with New Evidence

Patricia M. Di Lorenzo, Andrew M. Rosen
Binghamton University, Psychology Binghamton, NY, USA

The idea that there are a finite number of taste qualities (defined as groups of chemicals that taste alike) is as old as the study of taste itself. Historically, there has been only four recognized taste qualities: sweet, sour, salty and bitter. As time and technology have advanced, however, the idea of four "basic" taste qualities has been challenged. Most notably, the acknowledgement of umami as a basic taste quality, first evidenced by psychophysical data over a hundred years ago, was cemented by the discovery of umami-specific receptors on the tongue. More recently, as the prospect of additional taste qualities has acquired momentum, ideas about how we perceive and parse the taste world may have to be revised. For example, although the idea that we can perceive water as an independent taste is not new, mounting molecular and physiological evidence has provided fresh support. A water-dedicated transduction mechanism has been identified in the peripheral nervous system and water-responsive fibers in the peripheral taste nerves have been reported. In addition, water-responsive as well as water-specific neurons have been described in gustatory nuclei in the central nervous system. In our own work, we have recorded such neurons in the nucleus of the solitary tract and the parabrachial nucleus of the pons, respectively the first two central relay nuclei in the rodent brainstem, in both anesthetized and awake preparations. Although the size of the representation is not large, we argue that it is distinctive and similar in character to the representation of other taste qualities. It is possible then, that water constitutes an independent taste modality encoded by neurons that may be key elements in the regulatory system for fluids. Acknowledgements: Supported by NIDCD grant DC006914 to PMD.

#57 **SYMPOSIUM: BASIC TASTES: WHY FIVE?**

Separate Tastes for Sugar, Maltodextrin and Starch

Anthony Sclafani
Brooklyn College - City University of New York Brooklyn, NY, USA

The sweet taste quality helps humans and other mammals identify sugars. The peripheral taste response to sugars is mediated by the T1R2/T1R3 taste receptor. Other chemosensors may exist for the detection of starch, the most abundant carbohydrate in nature. Some species appear to perceive starch-derived glucose polymers (maltodextrins; e.g., polycose) as having a taste that is distinct from that of sucrose. This is well documented by behavioral and electrophysiological studies of rodents. For instance, rats are attracted to polycose and sucrose at low concentrations but do not cross-generalize conditioned taste aversions between the two carbohydrates. Studies of knockout mice indicate that the maltodextrin taste is independent of the T1R2 and T1R3 sweet receptors but dependent upon gustducin and TRPM5 signaling proteins. Additional evidence indicates that maltodextrin taste may be distinct from starch taste. Conditioned aversions to pure starch and polycose do not cross-generalize in rats. In addition, gustducin knockout (KO) mice display impaired polycose but not starch preference, whereas TRPM5 KO mice are indifferent to both polycose and starch. Humans are not attracted to pure starch or starch-derived glucose polymers, but recent findings suggest that they may detect glucose polymers. Exercise performance was enhanced by oral stimulation with glucose or maltodextrin but not by non-nutritive sweeteners. The maltodextrin taste receptor in rodents remains to be identified and its existence in humans warrants investigation. Much more work is needed to elucidate starch chemosensation and the functional significance of multiple carbohydrate sensors. Acknowledgements: NIDDK Grant DK031135

#59 **PLATFORM PRESENTATIONS: TASTE**

Wnt/β-catenin signaling controls the renewal of differentiated taste cells of adult mice

Dany Gaillard[1], Sarah E Millar[2], Fei Liu[3], Linda A Barlow[1]
[1]Department of Cell & Developmental Biology-Rocky Mountain Taste & Smell Center, University of Colorado Denver, School of Medicine Aurora, CO, USA, [2]Departments of Dermatology and Cell & Developmental Biology, University of Pennsylvania School of Medicine Philadelphia, PA, USA, [3]Institute for Regenerative Medicine at Scott & White Hospital, Texas A&M University System Health Science Center Temple, TX, USA

In adult mice, cells within taste buds are continually renewed. Signaling pathways including Sonic Hedgehog, BMP, and Notch are thought to control this turnover, but specific molecular mechanisms have not been elucidated. Moreover, while Wnt/β-catenin signaling regulates embryonic taste bud development (Liu et al., 2007), its involvement in adult taste cell turnover is unexplored. We used conditional doxycycline-inducible Cre recombination of either exons 2-6 or exon 3 of the β-catenin gene to induce β-catenin loss- (LOF) or gain- (GOF) of function, respectively, in the lingual epithelium of adult mice. After one month of doxycycline, β-catenin LOF induced a significant

reduction in α-gustducin-IR (type II) cells in taste buds in the circumvallate papilla (CVP). Moreover, we found that proliferation of extragemmal basal cells in the CVP was dramatically reduced, as determined by the proliferation marker Ki67. In the β-catenin GOF mice, we found, paradoxically, that α-gustducin-IR cells are also reduced, but additionally, that NCAM-IR (type III) cells disappear after 2 weeks of doxycycline. Yet in β-catenin GOF tissue, proliferation appears grossly normal. Because proliferation is reduced when β-catenin is lost from basal epithelial cells, β-catenin is likely required for proliferation of extragemmal basal cells, which are the progenitor population for taste buds; thus in the absence of proliferation, new taste cells are not generated. Beta-catenin GOF also results in fewer differentiated taste cells, but we hypothesize that this is because elevated β-catenin induces continued progenitor proliferation in lieu of taste cell differentiation. In sum, our data implicate β-catenin signaling in the renewal of type II and III taste cells.
Acknowledgements: Supported by NIH/NIDCD DC008373 and ARRA DC008373-03S1 to LAB

#60 **PLATFORM PRESENTATIONS: TASTE**

COMPREHENSIVE MAPPING OF FUNCTIONAL SITES FOR AGONISTS AND INHIBITORS OF THE BITTER TASTE RECEPTOR TAS2R16 BY SHOTGUN MUTAGENESIS

Please Note: *This presentation will be given by Paul A. S. Breslin*

Joseph B. Rucker[1], Anu Thomas[1], Suzanne Alarcon[3], Tiffani A. Greene[1], Eli Berdougo[1], Wely B. Floriano[4,5], Benjamin J. Doranz[1], Paul A. S. Breslin[2,3]
[1]Integral Molecular, Inc. Philadelphia, PA, USA, [2]Monell Chemical Senses Center Philadelphia, PA, USA, [3]Rutgers University New Brunswick, NJ, USA, [4]SciReal Grand Marais, MN, USA, [5]Lakehead University Thunder Bay, ON, Canada

Bitter tastes are detected at the cellular level by a diverse family of taste receptors (TAS2Rs) belonging to the G protein-coupled receptor (GPCR) superfamily. Because GPCRs are challenging targets for direct structural determination, such as by x-ray crystallography, defining the structural features of TAS2Rs that are responsible for binding ligands is challenging. We have developed a high-throughput strategy called Shotgun Mutagenesis Mapping for structure-function analysis of TAS2Rs and other GPCRs. Shotgun Mutagenesis Mapping enables a complete and unbiased residue-by-residue evaluation of the contribution of each amino acid to ligand-mediated function. We created a comprehensive TAS2R16 mutation library with defined point mutations at every amino acid in the receptor and screened each clone's functional activity in response to the ligand salicin using a fluorescent Ca^{2+}-flux signaling assay. We identified critical amino acids throughout the protein that, when substituted, abrogate salicin dependent cell signaling. These residues define the ligand binding site for salicin on TAS2R16 as well as defining residues important for signal transduction. During these studies, we also unexpectedly identified an inhibitor with activity against TAS2R16 and several additional TAS2Rs that diminished perceived bitterness of salicin in humans. We identified key point mutants that suppressed the action of the inhibitor without affecting the activity of salicin, which, together with additional pharmacological experiments, suggest an allosteric mechanism of action for this TAS2R16 inhibitor. Overall, our results demonstrate the utility of Shotgun Mutagenesis Mapping for defining ligand binding sites on complex receptors and guiding the structural understanding of bitter taste receptor function.
Acknowledgements: NIH DC010105 to J.B.R. NIH DC002995 to P.A.S.B.

#61 **PLATFORM PRESENTATIONS: TASTE**

A proton current drives action potentials in genetically identified sour taste cells

Rui Chang, Hang Waters, Emily Liman
University of Southern California, Department of Biological Sciences, Section of Neurobiology Los Angeles, CA, USA

Five tastes have been identified. Of these, sour is associated with acidity, a characteristic of fermented food, and as such is usually aversive. Recent results demonstrated that sour stimuli are detected by a subset of taste cells that express the TRP channl PKD2L1, as selective elimination of these cells eliminates nerve responses to sour stimuli. The PKD2L1/PKD1L3 channel was previously reported to be a sour receptor; however, we could not detect any direct activation of this channel in heterologously expressed cells, even under conditions as acidic as pH 2.5. Instead, as previously reported, we detected a delayed Ca^{2+} response after washing off the acids in ~30% of transfected cells. That this "off" response persisted for tens of seconds and could not be elicited more than once in the same cells indicates that the PKD2L1/PKD1L3 complex may not serve as a sour receptor. To identify acid-sensitive conductance unique to sour cells, we created genetically modified mice in which sour cells were marked by expression of YFP under the control of the PKD2L1 promoter. To measure responses to sour stimuli we developed a method in which suction electrode recording is combined with UV photolysis of NPE-caged proton. Using these methods, we report that responses to sour stimuli are not mediated by Na^+ permeable channels as previously thought, but instead are mediated by a proton conductance specific to PKD2L1-expressing taste cells. This conductance is sufficient to drive action potential firing in response to acid stimuli, is enriched in the apical membrane of PKD2L1-expressing taste cells and is not affected by targeted deletion of the PKD1L3 gene. We conclude that, during sour transduction, protons enter through an apical proton conductance to directly depolarize the taste cell membrane.
Acknowledgements: DC004564

#62 **PLATFORM PRESENTATIONS: TASTE**

Fatty Acids Activate Type II and a Subset of Type III Mouse Taste Cells

Timothy Gilbertson, Pin Liu
Utah State University/Biology & The Center for Advanced Nutrition Logan, UT, USA

Type II cells are generally accepted as the receptor cells for sweet, bitter and umami tastants and they express the appropriate transduction machinery including PLCß2, TRPM5, and IP_3R3, all of which have been used to identify this cell type. Our recent data have shown that TRPM5 plays a critical role in the transduction pathway for linoleic acid at the cellular and behavioral levels implicating the involvement of Type II cells in this process. We have primarily used functional calcium imaging to determine if Type II cells were the only fatty acid (FA)-responsive cell type. To test this idea, we have used responses to high KCl to identify type III cells and responses to a tastant mixture to identify type II cells. FAs are primarily able to elicit $[Ca^{2+}]_i$ responses in a proportion of Type II cells, however, a small but significant subset of Type III cells also respond to FAs. Because we previously questioned the utility of using KCl to functionally identify Type III cells, these findings have been verified in transgenic mice expressing green fluorescent protein under control of the PLCß2 (Type II) or GAD67 (Type III) promoter. We find a significant proportion of Type II cells and some Type III cells respond to a variety of FAs, including caproic, myristic, oleic, linoleic, and arachidonic acids, representing short, medium and long chain, saturated and unsaturated fatty acids. Few cells appeared to be completely specific for individual classes of fatty acids. While we hypothesize that Type II cells are the primary FA-sensitive cells, Type III cells appear to contribute to the transduction of fatty acids in taste cells. The mechanism for FA transduction in Type III taste cells remains an open question as is the relative importance of the Type III pathway to FA reception *in vivo*. Acknowledgements: Supported by NIH DK059611 and International Flavors & Fragrances.

#63 **PLATFORM PRESENTATIONS: TASTE**

Salivary PYY: a putative bypass to satiety

Daniela Hurtado[1], Andres Acosta[1], Oleg Gorbatyuk[2], Michael La Sala[1], David Duncan[1], George Aslanidi[1], Martha Campbell-Thompson[3], Shawn C. Dotson[4], Lei Zhang[5], Herbert Herzog[5], Bruce J. Baum[6], Sergei Zolotukhin[1]
[1]University of Florida/Pediatrics Gainesville, FL, USA, [2]University of Florida/Molecular Genetics and Microbiology Gainesville, FL, USA, [3]University of Florida/Pathology Gainesville, FL, USA, [4]University of Florida/Neuroscience and Psychiatry Gainesville, FL, USA, [5]Garvan Institute of Medical Research Sydney, Australia, [6]Molecular Physiology and Therapeutics Branch Bethesda, MD, USA

The objective of the study was to identify and characterize functions of Peptide YY (PYY) in saliva. PYY_{3-36} is a satiation hormone released postprandially into the bloodstream from L-endocrine cells in the gut epithelia. In the current report, we demonstrate PYY_{3-36} is also present in murine as well as in human saliva. In mice, salivary PYY_{3-36} derives from plasma and it is also synthesized in the taste receptor cells in taste buds of the tongue. Moreover, the cognate receptor Y2R is abundantly expressed in cells of the tongue epithelia and von Ebner's gland. The acute augmentation of salivary PYY_{3-36} induced stronger satiation as demonstrated in feeding behavioral studies. The anorexigenic action of salivary PYY_{3-36} was corroborated by an increase in neuronal activity in the paraventricular and arcuate nuclei. Moreover, unlike intraperitoneal administration, the augmentation of salivary PYY_{3-36} does not induce conditioned taste aversion (CTA), the fact supported by the relatively low level of activation in neurons in the area postrema of the brain stem. In diet-induced obese (DIO) mice, the chronic augmentation of salivary PYY was achieved by using PYY transgene delivery. Over the course of 8 weeks, after a single treatment, DIO mice had lost 23% of free feeding body weight. Thus this study provides evidence for new functions of the previously characterized gut peptide PYY_{3-36} suggesting a potential simple and efficient alternative therapeutic approach for the treatment of obesity. Acknowledgements: The research was supported in part by the Children's Miracle Network Foundation.

#64 **PLATFORM PRESENTATIONS: TASTE**

Tasting outside the mouth favors perception of bitterness relative to sweetness

Danielle J. Nachtigal[1], Juyun Lim[3], Barry Green[1,2]
[1]The John B. Pierce Laboratory New Haven, CT, USA, [2]Yale University School of Medicine New Haven, CT, USA, [3]Department of Food Science and Technology, Oregon State University Corvallis, OR, USA

The ability of sucrose to mask bitterness contradicts the assumed function of bitter taste as a signal of potential poisons. During informal testing we observed that sweetness, but not bitterness, seemed weaker when tasted with the tongue tip outside compared to inside the mouth. The implications of this observation for masking of bitterness by sweetness were tested by asking Ss to rate the sweetness and bitterness of 2 concentrations of sucrose and QHCl and their mixtures in 2 conditions: when the tongue was dipped for 5s in a 10-ml solution ('Dip'), or when the tongue was dipped in the sample for 1s then retracted into the mouth for 4s ('Retract'). Sucrose was rated significantly sweeter in the 'Retract' condition compared to the 'Dip' condition, whereas the bitterness of QHCl was rated significantly higher in the 'Dip' condition. Consistent with these trends, sucrose-QHCl mixtures were rated as more bitter in the 'Dip' condition than in the 'Retract' condition. A second experiment tested the hypothesis that the higher bitterness of the mixture outside the mouth was due in part to a rapid adaptation of sweetness in that mode of tasting. Ss again rated sweetness and bitterness, this time after 0, 3, 10 or 20s exposures to an adapting stimulus of sucrose or QHCl. The adapting and test stimuli were sampled by dipping the tongue into the solutions ('Dip'), or sipping and holding them in the mouth ('Sip'). The results confirmed that sweetness adapted more rapidly than bitterness in the 'Dip' condition but not in the 'Sip' condition. These findings suggest that sampling a stimulus with the tongue outside the mouth, as might occur in nature when encountering an unfamiliar potential food, increases the ability to detect possible poisons via a selective adaptation of sweetness, which unmasks bitterness. Acknowledgements: NIH RO1 DC005002

#65 PLATFORM PRESENTATIONS: TASTE

The Neural Correlates of Taste Mixtures

Julie A. Boyle[1], Jurgen Germann[2], Michael Petrides[1]
[1]Montreal Neurological Institute Montreal, QC, Canada,
[2]Hospital for Sick Children Toronton, ON, Canada

In everyday life humans commonly consume foods that contain more than one tastant. The aim of our study was to investigate whether subjects' brains activate different areas based on whether they are perceiving a single tastant or a mixture of two tastants. Eight healthy right-handed university-aged subjects (4 women) underwent a functional magnetic resonance imaging scan in which they were presented with either sucrose (S), quinine (Q) or a mixture of sucrose and quinine (SQ). Both individual tastants and the mixture were presented at isointense concentrations. Distilled water was used as a baseline condition. Contrasting the binary mixture and the single tastants (SQ- [S+Q]) resulted in activation of the left middle cingulate and lateral orbitofrontal cortex as well as the right anterior cingulate. A contrast of single tastant with the binary mixture ([S+Q] – SQ) did not yield any significant activations. Based on our current findings we conclude that binary taste mixtures and their individual components are processed differently by the human brain. Acknowledgements: This research was supported by grant from the Canadian Institutes of Health Research awarded to MP.

#66 PLATFORM PRESENTATIONS: TASTE

Opposing influences of flavor-evoked response in midbrain and medial orbital cortex vs. lateral prefrontal cortex on ad lib food intake

Sarah Nolan-Poupart[1], Maria G Veldhuizen[1,2], Dana M Small[1,2]
[1]The John B. Pierce Laboratory New Haven, CT, USA,
[2]Yale University School of Medicine New Haven, CT, USA

In a previous study (Small et al., 2001) we identified brain regions where response to consumption of Lindt chocolate decreased or increased as a function of satiety. We suggested that those regions with decreased response encoded the reward value of the chocolate (e.g. midbrain and medial orbitofrontal cortex mOFC), whereas the regions with increased response provided inhibitory signals mediating meal termination (ventral later prefrontal cortex vlPFC). What is unknown is whether responses in these regions evoked during flavor perception predicts subsequent ad lib intake of the food associated with that flavor. To investigate this we used fMRI to measure brain response to chocolate milkshake and a tasteless control solution in 20 healthy subjects. Subjects rated the pleasantness of these stimuli, as well as their hunger before and after the functional runs. A within-subject analysis of variance (SPM5) comparing milkshake to tasteless produced bilateral activation in the insular cortex corresponding to primary taste cortex. A regression analysis showed that activity in the mOFC and midbrain correlated positively with intake. This relationship was not influenced by pleasantness, or hunger. In contrast, a negative association was found between intake and vlPFC activation that was significantly influenced by ratings of perceived pleasantness but not hunger. We conclude that higher responses in the midbrain and mOFC and lower responses in the vlPFC are associated with greater ad lib intake. Since hunger does not influence these responses, we suggest that the network underlies hedonic rather than homeostatic feeding. In contrast, perceived pleasantness significantly modulated response in vlPFC. These findings confirm opposing influences of medial OFC vs. vlPFC on intake of a palatable food. Acknowledgements: ROI DK085579

POSTER PRESENTATIONS

#P1 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Preference for Dried-Bonito *Dashi* (a Traditional Japanese Broth) in Rodents

Takashi Kondoh, Tetsuro Matsunaga, Hanae Yamazaki
AJINOMOTO Integrative Research for Advanced Dieting, Graduate School of Agriculture, Kyoto University Kyoto, Japan

The dried-bonito (DB) *dashi* is a traditional Japanese broth that enhances palatability of various cuisines due to its specific flavor. The *dashi* is the mixture of various taste and olfactory substances as well as macromolecules. To select a suitable animal model for investigating mechanisms of preference for *dashi*, here we have investigated concentration-preference functions of DB *dashi* in various rodents using the 48-hr two-bottle preference test. The animals used were the adult male C57BL/6 mice, ICR mice and Sprague-Dawley (SD) rats. The commercial DB *dashi* employed were the "*Hondzukuri Ichiban-dashi Katsuo*" (Ajinomoto, Japan) which is 5- to 10-fold dense-taste broth of hot-water extracted DB. As the dry matter components was 4% (w/w) on a weight basis, the commercial *dashi* is considered as 4% *dashi* solution. In all animal groups, preference for *dashi* was commonly observed between 0.4% and 4% solutions and the most palatable concentrations were between 1.2% and 2% solutions. However, the maximal preference varied among animals with the order of SD rats (preference ratio, 99%) > ICR mice (80%) > C57BL/6 mice (65%). In addition, total daily intakes (water + *dashi* solutions) in the SD rats and ICR mice increased significantly when tested palatable solutions (0.4-4%) compared with low (0.004-0.2%) solutions, while C57BL/6 mice showed no such alterations in any concentrations. These results suggested that there are strain and species differences in preference for DB *dashi* in rodents. It is worth interesting that C57BL/6 mice showed only weak preference for *dashi* while they are known to show strong preference for umami (glutamate) taste. In conclusion, we recommend to select the SD rats as one of the best animal models to advance studies concerning preference for DB *dashi* in rodents.

#P2 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Experience with Sapid Fluids Stimulates MSG Solution Preference in Mice

Karen Ackroff, Anthony Sclafani
Brooklyn College of CUNY Brooklyn, NY, USA

C57BL/6 mice are reported to prefer MSG over a range of concentrations in 48-h two-bottle tests. These animals had prior experience with other sapid solutions. In our first experiment, naive B6 mice failed to prefer to MSG at any concentration tested (0.1- 450 mM). To explore the effects of experience, the same mice were next given forced exposure to 300 mM MSG (for 4 days as the sole fluid source) and then retested. They now exhibited significant preferences for 1-300 mM MSG. New groups of naive mice were exposed to 0, 10, 100, or 300 mm MSG; only experience with 300 mM significantly increased subsequent MSG intake. Other naïve mice were given experience with 8% sucrose, 8% Polycose or 0.8% sucralose. Only the sucrose and Polycose groups subsequently preferred MSG. The preference threshold at which MSG intake significantly exceeded water intake was 1 mM after MSG experience, 10 mM after sucrose, and 100 mM after Polycose experience. These thresholds were inversely related to total preexposure intakes. The reported generalization of MSG and sucrose responses in rodents under some conditions suggests that they share some taste properties, which may be related to the T1R3 receptor common to the sweet and umami taste receptors. Polycose taste, however, is not mediated by the T1R3 receptor. The lack of effect of sucralose indicates that sweet taste alone is not sufficient to enhance MSG preference. Together these results suggest that a combination of oral and post-oral effects may be responsible for the experience effect, with MSG itself the most potent stimulus. Acknowledgements: Supported by the Ajinomoto Amino Acid Research Program.

#P3 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Effect of chorda tympani nerve transection on salt taste perception in mice

Glen J Golden[1], Yutaka Ishiwatari[1,2], Maria L Theodorides[1], Alexander A Bachmanov[1]
[1]Monell Chemical Senses Center Philadelphia, PA, USA,
[2]Institute of Life Sciences, Ajinomoto Co., Inc. Kawasaki, Japan

Effects of gustatory nerve transection on salt taste have been studied extensively in rats and hamsters, but have not been well explored in the mouse. We examined the effects of chorda tympani nerve transection on NaCl taste preferences and thresholds in outbred CD-1 mice using a high-throughput phenotyping method developed in our lab. To measure taste thresholds, mice were conditioned by oral self-administration of LiCl and then presented with NaCl conentration series in two-bottle preference tests. LiCl-conditioned and control NaCl-exposed mice were given bilateral transections of the chorda tympani nerve (LiCl-CTX, NaCl-CTX) or were left intact as controls (LiCl-CNT, NaCl-CNT). After recovery from surgery, mice received a concentration series of NaCl (0 – 300 mM) in 48-h two-bottle tests. Chorda tympani transection increased NaCl taste thresholds in LiCl-conditioned mice and eliminated avoidance of concentrated NaCl in control NaCl-exposed mice. This demonstrates that in mice the chorda tympani nerve is important for detection and recognition of NaCl taste and conveys a hedonically aversive sensation evoked by high concentrations of NaCl. The results of this experiment also show that the method of high-throughput phenotyping of salt taste thresholds is suitable for detecting changes in the taste periphery in mouse genetic studies. Acknowledgements: This work was supported by the NIH training grant NIDCD 5T32DC000014-30 (GJG) and NIH grant R01 DC00882 (AAB).

#P4 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

An investigation of the relationship between umami and salt tastes using the Microtiter Operant Gustometer (MOG), a high throughput operant taste discrimination assay

Kyle Palmer, Daniel J. Long, Raymond Salemme
Redpoint Bio Corp, Discovery Research Ewing, NJ, USA

A synergistic association between the basic tastes of salt and umami has been suggested from the effects of adding fermented protein to sodium-reduced foods. It is unclear if the association is an actual synergistic effect on salt taste or if the reduced-salt foods are made more palatable by fermented protein mixtures. Few studies have systematically characterized salt-umami interactions and the relationship between the two tastes remains ambiguous. Using the MOG, a high throughput operant taste discrimination technology, we have trained two cohorts of rats to discriminate a sodium cue (100 mM NaCl) and an umami cue (3 mM IMP+3 mM MSG+10 mM amiloride), respectively. Sodium- and umami-trained rats readily discriminated their respective training cues from sour (10 mM citric acid), bitter (10 mM quinine), sweet (100 mM sucrose) tastes and water. All rats discriminated sodium from umami taste cues with no cross-generalization observed between the cohorts. A full dose response function for sodium taste of NaCl solutions was established with the sodium cohort yielding an EC50 of approximately 30 mM. Sodium taste of MSG (without IMP or amiloride) across a range of concentrations also was evaluated using the sodium-trained cohort. The lower concentrations (2-38 mM) of MSG were reported as "sodium-like" by the rats, but higher concentrations (75-300 mM) did not generalize to the sodium cue. However, a sodium-taste dose-response function was evident, yielding an EC50 (20 mM) that was not significantly different than that of NaCl. Furthermore, adding 3 mM IMP+3 mM MSG to the NaCl concentration range had no impact on the NaCl dose-response function for sodium-trained rats. Our data indicate that umami taste *per se*, does not enhance the taste of sodium, but possibly could make low-sodium foods more appetitive.

#P5 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Use of the Conditioned Taste Aversion Generalization Procedure to Assess the Contribution of the TRPV1 Channel to Salt Taste Quality in Mice

Kimberly R. Smith, Yada Treesukosol, Alisa Millet, Robert J. Contreras, Alan C. Spector
Department of Psychology, Program in Neuroscience, Florida State University Tallahassee, FL, USA

At least two transduction mechanisms are involved in salt taste—the sodium selective epithelial sodium channel, which amiloride (AMIL) blocks, and an amiloride-insensitive cation nonselective pathway(s). Prior electrophysiological evidence from the chorda tympani nerve (CT) has implicated the TRPV1 channel as a major component of amiloride-insensitive salt taste transduction. However, TRPV1 knock-out (KO) mice have normal taste detection thresholds to NaCl and KCl with and without AMIL treatment. Here we conditioned a taste aversion to 0.5 M NaCl prepared with AMIL (CS) and examined taste generalization profiles. Thirsty WT and KO mice (9/group) were presented with 15 min of the CS followed by injection (ip) of 3.0 mEq/kg of either LiCl or NaCl on 3 conditioning trials. These animals were later tested in a gustometer that delivered 5-s trials of test stimuli (0.5 M NaCl, 0.5 M sodium gluconate (NaGlu), 0.5 M KCl, 0.5 M NH_4Cl, 13.25 mM citric acid (CA), 0.3 mM quinine, and 1.0 M sucrose) in randomized blocks with a 5-lick water rinse between each stimulus presentation. Both LiCl-injected WT and KO mice avoided KCl, NH_4Cl, CA, and quinine suggesting a non-TRPV1 amiloride-insensitive channel(s) is contributing to salt taste. Whereas LiCl-injected WT mice significantly avoided NaCl and NaGlu, KO mice did not, suggesting that TRPV1 contributes to oral perception of sodium salts, at least at high concentrations. However, electrophysiological recordings of the CT measured in a subset of these animals (WT=2; KO=4) revealed relatively normal AMIL-induced suppression of sodium responses which contrasts with prior findings in the literature. Thus, whether the effect of TRPV1 deletion on sodium salt perception in our study is based on gustatory signals, at least in the CT, remains to be resolved. Acknowledgements: NIH R01-DC004574 to A.C.S.

#P6 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Investigations Into the Mechanism Underlying the Super-saltiness of Sodium Carbonate to Rats

Steven J St. John, Bennett Garfinkel, Shakirra Meghjee
Department of Psychology, Rollins College Winter Park, FL, USA

Sodium appetite is a fascinating perceptual phenomenon in which organisms that are physiologically deficient in sodium seek out and consume salts at concentrations and in amounts exceeding those consumed when they are physiologically replete. Work by many investigators over the last 80 years confirms that this salt appetite is unlearned, taste-based, and "salty-specific"; that is, organisms ingest substances based on how salty they are (Schulkin, J Comp Physiol Psychol 96:628 1982). Extending some classic studies by Morrison (e.g., Physiol Behav 8:25 1972), we recently reported (St. John, Marshall, & Krauskopf, AChemS, 2009) that sodium-deprived rats respond to Na_2CO_3 as if it were ten times as salty as NaCl. We report now on two studies using Sprague-Dawley rats designed to investigate the mechanism of this "super-saltiness". In Experiment 1, thirsty rats were tested in a brief-access licking paradigm for willingness to ingest NaCl, Na_2CO_3, and pH-modified NaCl (3 – 715 mM) to assess whether the high pH of Na_2CO_3 contributed to increased salt-sensitivity. In contrast to the hypothesis, rats avoided Na_2CO_3 at concentrations ten times lower than NaCl regardless of pH (~7 or ~11.5). In Experiment 2, thirsty rats were presented with NaCl and Na_2CO_3 (7 – 1540 mM) mixed in one of four concentrations of amiloride (0 – 100 M) to assess the possibility that the epithelial sodium channel has a higher affinity for Na_2CO_3 than NaCl (c.f., Lyall et al., J Physiol 120:793 2002). Amiloride had a slightly more potent effect on Na_2CO_3 responses than NaCl responses, but high doses of amiloride did not eliminate the behavioral disparity between the two salts. The mechanism for the perceptual salience of Na_2CO_3 remains obscure.

#P7 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Sucralose Avoidance Predicts Sensitivity to Sweet and Bitter Tastants

Gregory C Loney, Christopher J Carballo, James C Smith, Lisa A Eckel
Program in Neuroscience, Florida State University Tallahassee, FL, USA

Rats vary in their preference for the artificial sweetener sucralose. At concentrations > 0.01 g/L, rats either prefer sucralose over water or they avoid it. Here, we determined if this bimodal response profile is unique to sucralose or if sucralose acceptance predicts the behavioral response to other basic or binary tastants. Two groups of rats were categorized as either sucralose preferrers (SP) or sucralose avoiders (SA). In the first group, we examined the rats' unconditioned licking responses to brief (30 s) presentations of a range of sucrose, NaCl, and quinine (QHCl) concentrations (0.015, 0.03, 0.06, 0.13, 0.25, 0.5, 1.0 M for sucrose/NaCl and the same values in mM for QHCl). The concentrations of each tastant were presented at random (testing order: sucrose, NaCl, QHCl). The second group was given a series of two-bottle, 24-h preference tests consisting of water and a 0.25 M sucrose solution adulterated with increasing concentrations of QHCl (0.015-1.0mM). In the brief-access tests, SA licked more and initiated more trials to lower concentrations of sucrose and licked less and initiated fewer trials to higher concentrations of sucrose, relative to SP (Ps<0.05). In comparison, the licking profiles to NaCl and QHCl did not differ between SP and SA. In the 24-h preference tests, all rats decreased their preference for the sucrose-QHCl mixture as the concentration of QHCl increased (Ps<0.05). SA also generalized their avoidance to the sucrose/QHCl mixtures, demonstrating a reduced preference, relative to SP, for mixtures containing > 0.13 mM quinine (Ps<0.05). We conclude that the rat's behavioral response to sucralose predicts their acceptance of sweet and bitter tastants and that SA display enhanced sensitivity to high concentrations of sucrose and QHCl, relative to SP. Acknowledgements: NIH DK73936

#P8 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

TRPM5 knockout mice are sensitive to the aversive postingestive effects of bitter compounds

Mariana Q Magalhães[1,2], Xueying Ren[1,2], Jozélia Ferreira[1,2], Ivan E de Araujo[1,2]
[1]The John B. Pierce Laboratory New Haven, CT, USA,
[2]Department of Psychiatry, Yale University New Haven, CT, USA

Besides their repulsive gustatory properties, bitter compounds have also the ability to promote acquired aversion via postingestive signals [1]. Because chemosensory cells of the gastrointestinal tract have been shown to express signaling elements that mediate bitter taste in the oral cavity [2], it has been proposed that bitter taste-like signaling in the gut may support acquired behavioral aversions to bitter compounds elicited by postingestive factors [1]. The cation channel TRPM5, which is expressed in both the oral cavity and gut [3], is required for normal perception of bitter taste via gustatory pathways [4,5]. We therefore tested the possibility that TRPM5 knockout mice are impaired in their ability to develop postingestive-driven aversions to bitter compounds. Our results show that, while TRPM5 knockout mice displayed behavioral indifference to a range of bitter tastants during short-term preference tests, they nevertheless retained the ability to develop aversions to these tastants during longer-term exposure. Our results therefore suggest that the mechanisms involved in the detection of aversive bitter compounds by gastrointestinal chemosensors may fundamentally differ from those of the oral cavity.
[1] Glendinning et al Physiol Behav 2008 93:757-65 [2] Rozengurt E Am J Physiol 2006 291:G171-7 [3] Bezençon et al Chem Senses 2007 32:41-9 [4] Zhang et al Cell 2003 112:293-301 [5] Damak et al Chem Senses 2006 31:253-64 Acknowledgements: NIDCD, Pierce Laboratories, CNPq (Brazil)

#P9 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

The Role of Post-Ingestive Cues in Feeding Preferences by Mice that Lack Taste Capabilities

Jennifer M Stratford, Thomas E Finger
U. of Colorado Denver Anschutz Medical Campus Aurora, CO, USA

The gustatory nerves of mice lacking P2X2 and P2X3 purinergic receptor subunits (P2X-KO) are unresponsive to taste stimulation (Finger et al., 2005). Surprisingly, P2X-KO mice show residual behavioral responses to concentrated tastants, presumably from post-ingestive detection. Therefore, the current study tested whether tastant-evoked gut signaling is functional in P2X-KO mice. WT and P2X-KO mice were given eight training sessions on alternating days either with an odor alone or with a different odor mixed with 150 mM monosodium glutamate (MSG). Then all animals were given preference tests consisting of both odors without MSG. Both WT and P2X-KO animals preferred the odor previously paired with MSG showing post-ingestive cues were detected and associated with an odor. Further, we measured MSG-evoked brain activation, by expression of the immediate early gene c-Fos (cFLI), in the n. of the solitary tract (nTS) - the primary taste/viscerosensory nucleus. In rostral, gustatory levels of the nTS, significantly less cFLI was present in P2X-KO animals compared to WT controls. In contrast, in the caudal, viscerosensory part of nTS, cFLI did not differ between WT and P2X-KO mice. Together, these results suggest that P2X-KO mice can form preferences based on post-ingestive cues and that post-ingestive detection of MSG does not require purinergic signaling that is crucial for the taste system. Acknowledgements: Supported by NIH and 3ARP grants to TEF.

#P10 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Abrupt Changes in Temperature Influence Single-Cell Responses of Geniculate Ganglion Neurons to Chemical Stimulation in Rats

Alexandre A Nikonov, Robert J Contreras
Florida State University,Department of Psychology & Program in Neuroscience Tallahassee, FL, USA

Temperature influences responses of geniculate ganglion (GG) neurons to chemical stimulation of fungiform taste buds. However, our prior studies were limited methodologically to a gradual 1°C/s change in temperature, while in common experience chemical stimulation often co-occurs with an abrupt substantial change temperature. Thus, we recorded stimulus-evoked lingual potentials (electrogustogram; EGG) simultaneously with single-cell 2.5-s neural responses from 10 narrowly tuned (4 NaCl-best; 4 MSG-best; 2 sucrose-best) and 11 broadly tuned (5 citric acid-best, 6 NaCl-Quinine generalist) neurons from anesthetized male rats. We adapted the tongue to 35°C and recorded EGG and GG responses to 100 mM NaCl, 100 mM MSG, 500 mM sucrose, 10 mM citric acid, 20 mM quinine HCl, and 100 mM KCl at three different stimulus temperatures of 10°- 15°, 23°- 25°, 38°- 40°C with a two-channel temperature control system. Artificial saliva (15mM NaCl, 22mM KCl, 3mM $CaCl^2$, 0.6mM $MgCl^2$) served as the rinse solution and solvent for all taste stimuli. Our preliminary findings show that NaCl-best neurons responded to cool 10°- 15°C 100 mM NaCl, but the response was largely due to an abrupt change in temperature and not to the NaCl stimulus; on the other hand NaCl-best neurons responded to warm 38°- 40°C 100 mM NaCl, but the response was mostly due to NaCl and not to a change in stimulus temperature. Sucrose-best neurons were unresponsive to cool sucrose, but responded to warm sucrose and off-responses to quinine and MSG. Although cooling by itself is an excellent stimulus for citric acid-best neurons, stimulation in cool 10°-15°C (and warm) solution reduced GG neural responses to citric acid. In general, the response patterns of neurons became less distinct when presented significantly below and above body temperature. Acknowledgements: NIH grant R01 DC004785

#P11 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Solution Temperature Alters Preference for and Acceptance of Water and Sweet Solutions

Ann-Marie Torregrossa, Michelle B. Bales, Joseph M. Breza, Thomas A. Houpt, James C. Smith, Robert J. Contreras
Florida State University Tallahassee, FL, USA

Temperature modulates sweet-taste perception in humans and the neural discharges of sweet-sensitive peripheral neurons in hamsters. However, little work has been done examining the role of temperature in rodent ingestive behavior. Using cages modified with Peltier-controlled lickometers we conducted two experiments to investigate the influence of temperature on the behavioral responses to water and sucrose solutions. In our first study we tested water-deprived and non-deprived rats (within subjects design) in a series of 10-min 2-bottle preference tests between 10°C water and water or sucrose (0.03-1.0M) at increasing temperatures (10-40°C). Both water-deprived and non-deprived rats preferred the 10°C water over warmer water as well as 20 and 10°C sucrose over 10°C water. Surprisingly, water deprived rats preferred 10°C water over warm (30°C-40°C) sucrose while non-deprived rats preferred sucrose at these temperatures. In the second experiment, using a modified Davis rig, non-deprived rats were presented with water or a single sucrose concentration (0.05-0.2M) at four temperatures (10-40°C). Bottles were presented 4 times each for 30s with 10s intervals between presentations. The solution concentration varied across test days. Lick rate to water was inversely related to temperature, with maximal lick rate at 10°C and lowest at 40°C. Lick rate to sucrose revealed an interaction of temperature and concentration: lick rate increased with sucrose concentration, but also showed an inverted-U response with respect to temperature, with maximal licking at 20 and 30°C and declining at both higher and lower temperatures. These studies demonstrate that solution temperature and deprivation state profoundly influence taste-mediated licking responses to water and sucrose solution in rats. Acknowledgements: NIH 5 T32 DC000044, NIDCD 04607, DC-004785 & DC-010110

#P12 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Perception of orosensory stimuli: influence of temperature and sources of individual variation

Martha R. Bajec, Gary J. Pickering
Department of Biological Sciences, Brock University
St. Catharines, ON, Canada

Propylthiouracil (PROP) responsiveness has long been used as an index of individual variation in oral sensation as general orosensory responsiveness to a variety of stimuli associates with the ability and degree to which individuals perceive PROP's bitterness. Thermal taste, the phenomenon where thermal tasters (TTs) perceive taste sensations from the application of thermal stimuli to the tongue, was recently described. Thermal taster status (TTS) appears to function as a marker of individual variation in orosensory perception as TTs are more responsive to prototypical orosensory stimuli and flavor attributes in complex beverages than thermal non-tasters. Here we examined the influence of TTS and PROP taster status (PTS), an expression of individuals' PROP responsiveness, on the relationship between stimulus temperature and orosensory perception. Perceptually equi-intense stimuli eliciting sweet, sour, bitter, and astringent sensations presented at 5°C and 35°C were evaluated by forty-four subjects (age 18-45, 15 males) using time-intensity methodology. As expected, a trend of TTs reporting higher perceived maximum intensities was observed for all stimuli. While some differences in maximum perceived intensity were found between PTS groups for the different stimuli, they were not as expected. As previously reported, TTS and PTS interactions were not observed. Interestingly, temperature influenced the maximum perceived intensity from astringent, bitter, and sour stimuli, but not from the sweet stimulus. We conclude that further investigation into the influence of TTS and PTS on orosensory perception in ecologically valid but well controlled conditions using time-intensity methodology are necessary.
Acknowledgements: This work was supported by a NSERC DG (GJP), a NSERC PGS-D (MRB), and the Pangborn Sensory Science Scholarship (MRB).

#P13 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Individual Differences in the Avidity for Calcium and Saccharin are Influenced by Variation in *Itpr3* or in a Nearby Gene on Mouse Chromosome 17

Hillary T. Ellis, Hongguang Shao, Danielle R. Reed, Michael G. Tordoff
Monell Chemical Senses Center Philadelphia, PA, USA

Calcium preferences differ markedly among inbred strains of mice. Of 40 strains surveyed, the BTBR T^{+} *tf*/J (BTBR) strain has among the highest, and the NZW Lac/J (NZW) has among the lowest, calcium preferences (Physiol Behav. 91:632-643, 2007). To identify the genetic variation underlying this strain difference, we first conducted a genome screen: The BTBR and NZW strains were intercrossed to produce 610 F_2 hybrid mice, which were phenotyped using two-bottle choice tests with several taste solutions and genotyped with 625 SNP markers spanning the entire genome. Interval mapping revealed a quantitative trait locus on chromosome 17 with remarkably strong linkage to preferences for 50 mM $CaCl_2$ (LOD = 45), 2 mM saccharin (LOD = 101), and other taste compounds. The NZW allele of this QTL is dominant; it decreases $CaCl_2$ preference and increases saccharin preference. To isolate the gene(s) responsible, we developed a congenic strain by introgressing the NZW allele onto the BTBR background. After 11 generations of successive backcrossing, the region of interest is ~1.4 Mbp. Mice that are heterozygous NZW/BTBR (n=14) in the congenic interval have significantly higher preferences for 2 mM saccharin (72% ± 5 vs. 45% ± 3) and lower preferences for 50 mM $CaCl_2$ (25% ± 6 vs. 44% ± 2) than mice that are homozygous BTBR/BTBR (n=64). Of the 44 genes within the congenic interval, the strongest candidate to mediate these phenotypes is *Itpr3*, a gene that has been previously linked to taste perception of sweet, umami, and bitter tastes. Our results show that large differences among mice in taste preferences can be accounted for by natural variation in *Itpr3* (or in a very small genetic region nearby). Acknowledgements: NIH DK-46791

#P14 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Aversion to Sucrose octaacetate by Laboratory Mice is controlled by a Polygenic System

David A Blizard[1], Ayako Ishii[2], Tsuyoshi Koide[2], Aki Takahashi[2], Toshihiko Shiroishi [4], Thomas P Hettinger[3], Marion E Frank[3], Lawrence D Savoy[3], Bradley K Formaker[3], Sezen Yertutanol[1], Arimantas Lionikas[5]
[1]Pennsylvania State University/Department of Biobehavioral Health University Park, PA, USA, [2]National Institute of Genetics/Mouse Genomics Resource Laboratory Mishima, Japan, [3]Department of Oral Health and Diagnostic Sciences/University of Connecticut Health Center Farmington, CT, USA, [4]National Institute of Genetics/Mammalian Genetics Laboratory Mishima, Japan, [5]University of Aberdeen/School of Medical Sciences Aberdeen, Scotland

Based on crosses among inbred mouse strains derived principally from *M. m. domesticus* sucrose octaacetate (SOA) aversion has been thought for many years to be controlled by a single genetic locus (*Soa*) located on distal chromosome 6. To expand knowledge of the genetic basis underlying SOA aversion, we have studied *M. m. molossinus* chromosomes derived from the MSM inbred strain on a *M. m. domesticus* (C57BL/6J: B6) host background (using 2-bottle preference procedures, MSM mice strongly avoided 0.1 mM and 1 mM SOA while B6 mice were indifferent to 0.1 mM and exhibited slight aversion to 1 mM SOA). Preference tests of 16 available consomic strains implicated chromosomes 2, 4 and 15 in SOA aversion in addition to the prominent effect of the established *Soa* locus on chromosome 6. The originally defined *Soa* locus is presumably associated with one or more members of the cluster of *Tas2r* genes on distal chromosome 6 that code for bitter taste receptors. Our results point to the possible role of known *Tas2r* genes on chromosomes 2 and 15 as well as genes not coding for bitter receptors (Chr 4), in SOA aversion. SOA aversion emerges from this consomic screen as being definitively under polygenic control. The genetic diversity captured by the MSM model system is shown to be a valuable tool to complement the limited genetic variation in commonly used stocks derived from *Mus m. domesticus*. The value of presenting other ionic (quinine, denatonium, KCl) and non-ionic bitter solutions (caffeine, salicin, nicotine) to the SOA-averse consomics will be discussed. Acknowledgements: This research was supported by NIH grant R01 DC004099 and a Japan Supplement to M.E Frank from the National Institute of Deafness and Communication Disorders and by KAKENHI (Grant-in-Aid for Scientific Research) from the Ministry of Education, Culture, Sports, Science and Technology of Japan, the Japan Society for the Promotion of Science (JSPS), and the Research Organization of Information and Systems, Transdisciplinary Research Integration Center to T. Koide, A. Takahashi and T. Shiroishi.

#P15 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Lactisole influences calcium taste

Laura K. Alarcon, Michael G. Tordoff
Monell Chemical Senses Center Philadelphia, PA, USA

Studies with mice suggest that T1R3, the sweet and umami taste receptor subunit, also mediates calcium taste transduction. Lactisole [Sodium 2-(4-methoxyphenoxy) propanoate] binds with the transmembrane domain of human T1R3 to reduce the intensity of sweeteners and monosodium glutamate. To determine whether lactisole also influences human calcium taste perception, we asked subjects to sample calcium salts and other taste compounds mixed with water or lactisole. The subjects rated the solutions' intensities, how much they liked each sample, and how sweet, sour, salty, bitter, and "other" they tasted. Relative to ratings when the solutions were given alone, lactisole reduced "otherness" and increased the sourness and overall intensity of calcium lactate, $CaCl_2$, and calcium gluconate but not several other compounds (NH4Cl, KCl, KSO4, MgSO4, quinine hydrochloride, urea, caffeine, or sucrose). These data suggest that either T1R3 acts as a calcium taste receptor or that lactisole acts on another receptor to influence calcium taste (or both). The results thus provide the first functional evidence that calcium taste has its own specific transduction mechanism in humans. Acknowledgements: Supported by NIH DK-46791

#P16 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Fat Taste: Qualitative and Quantitative Analysis of Salivary Free Fatty Acids in Humans

Bhushan V. Kulkarni, Richard D. Mattes
Purdue University West Lafayette, IN, USA

Free fatty acids (FFA) are proposed to be the ligands for various purported fat taste receptors. However, unlike rodents, humans may lack functional levels of lingual lipase to hydrolyze these molecules from triacylglycerols, the primary form of dietary fat. Hence, questions have been raised about the ecological significance of an oral FFA signaling system in humans. The present study sought to quantify and characterize FFA present in the oral cavity after oral processing of foods varying in nutrient content (i.e., fat type (saturated, monounsaturated or polyunsaturated) and concentration) and physical form (solid, semi-solid and fluid) to determine if adequate concentrations of FFA are present to evoke a "taste" response. Participants chewed standard amount of almond, walnut, coconut, almond butter and olive oil at a rate of a bite per second for 1 minute and then expectorated the sample. Quantitative and qualitative analyses of salivary FFA from the expectorated food samples were conducted by gas chromatography - mass spectrometry. Concentrations of salivary FFA were significantly increased after oral processing of each food sample as compared to their levels in the mechanically stimulated saliva. Palmitic acid, oleic acid, linoleic acid, and stearic acid were found to be the main salivary FFA. Salivary FFA concentrations in the expectorated food samples ranged from 30 to 60 micromolar and their profile reflected the FFA composition of the food. Thus, these findings indicate the presence of ligand for an oral FFA signaling system. Future studies with taste receptor cells are required to determine the adequacy of the salivary FFA concentrations seen in this study to evoke a signal in humans. Acknowledgements: USDA HATCH # IND084055

#P17 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Experience with Na-cyclamate affects human taste sensitivity for high-intensity sweeteners differently

Julia Sabin, Alexa T Navasero, Bennett R Collins, Elizabeth T Rosen, Michael S Zemel, Miranda Valerio, Todd P Livdahl, Linda M Kennedy
Clark University Worcester, MA, USA

Treatment with Na-cyclamate (Na-c) significantly increases human taste sensitivity for glucose, fructose, and maltose, and leads to increases approaching significance for sucrose (Gonzalez et al., 2007, 2008; Collins et al., 2010). Human psychophysical, hamster chorda tympani and *Drosophila melanogaster* neurophysiology data suggest mechanisms in the peripheral nervous system (Faurion et al., 2002; Hassan et al., 2006; Gonzalez et al, 2009). We have suggested that binding of the treatment compound with the receptor molecule (T1R3 in the case of humans) leads to changes in binding or other steps in the receptor response to the test compound. Here we tested whether Na-c treatment leads to a similar increase in taste sensitivity for Na-c, sucralose (sucrl), and D-tryptophan (D-tryp). Na-c is a salt of cyclamic acid that binds T1R3. Sucrl is sucrose modified with 2 Cl atoms and D-tryp is an amino acid; modeling data suggest sucrl and D-tryp bind the Venus flytraps of T1R2 and T1R3 (Morini et al., 2005). Subjects rinsed their tongues with 4 mM Na-c or water for 10 sec once a day for 10 days. On day 11 or 12, they tasted a concentration series of Na-c, sucrl, or D-tryp, each concentration paired with water, and indicated which of each pair was "the sweetener." Subjects treated with Na-c showed significant increased sensitivity for Na-c and sucrl, but significant decreased sensitivity for D-tryp. These results indicate that the mechanism(s) for experience-induced changes affect(s) stimulation by various sweeteners differently. Acknowledgements: Supported by NIH NIDCD R15DC009042 to LMK.

#P18 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Inhibition of Human Sweet Taste Perception By the Lipid Lowering Drug Clofibrate

Matthew C Kochem[1], Bedrich Mosinger[2], Suzanne M Alarcon[2], Robert F Margolskee[2], Paul AS Breslin[1,2]
[1]Rutgers University Department of Nutritional Sciences New Brunswick, NY, USA, [2]Monell Chemical Senses Center Philadelphia, NY, USA

The T1R2-T1R3 receptor is believed to be the principal carbohydrate sensing taste receptor in humans and is inhibited *in vitro* by lactisole. We recently showed that clofibric acid, a lipid lowering drug structurally related to lactisole, also inhibits T1R2-T1R3 activity *in vitro.* Here we sought to determine if this fibrate drug inhibits sweet taste perception in humans. 14 participants rated the sweetness intensity of four sweeteners (Na cyclamate, sucralose, acesulfame-K, sucrose) across a broad range of concentrations. Each sweetener was prepared neat and with 1.37 mM of the T1R2-T1R3 inhibitors, Na lactisole and Na clofibrate. In general, every subject showed sweetness inhibition by both inhibitors for all four sweeteners. Compared to lactisole, clofibrate demonstrated a very similar inhibitory potency and pattern with Na cyclamate. But clofibrate was a more potent inhibitor of sucralose for most individuals and tended to be a slightly more effective inhibitor than lactisole of acesulfame-K. Sucrose inhibition by the two inhibitors varied across individuals. Consistent with competitive binding, the inhibition of sucrose and cyclamate was diminished with increasing concentration of sweetener. With sucralose and acesulfame-K, however, the inhibition appeared somewhat less competitive. In conclusion, the lipid lowering drug clofibrate inhibits perception of sweetness in humans and is, thus, a carbohydrate receptor inhibitor *in vivo*. Whereas clofibrate generally tended to be a more potent inhibitor of sweetness than lactisole, effects varied with sweetener and individual participant. Next, we will compare the *in vivo* perceptual responses of these subjects with the functional performance of their individual T1R2-T1R3 receptors *in vitro*. Acknowledgements: NIH DC02995

#P19 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Structural Analogues of Homoeriodictyol as Bitter Taste Modifiers: Structure-Activity Concepts by Sensory Analysis

Jakob P. Ley, Katharina V. Reichelt, Gerhard E. Krammer
1 Holzminden, Germany

Bitter taste is a huge challenge for product development in food industry because pharmaceutical bitter reduction strategies are limited due to legal or technical restrictions. In the past we found some caffeine bitter maskers based on homoeriodictyol (HED) by sensory screening (1-3). To elucidate the gustophors, we i) varied and simplified the structure of HED and ii) tested their activity against various bitterants. Various flavanones, other flavonoids, benzoic acid amides, gingerdiones, and deoxybenzoins were screened for their activity against different bitterants (caffeine, quinine, salicin, naringin). Sensory analysis was performed with a trained panel (>=12) using a blinded and randomized duo comparison by rating on a scale from 1 - 10. Averaged ratings for blind and test samples were compared. HED and eriodictyol (ED) showed about 40-50 % reduction of bitter ratings in a 500 ppm caffeine solution at a concentration of 100 ppm. All structural modifications or simplifications led to decrease of activity. Only vanillic acid vanillyl amide, 4,4'-dihydroxy-3-methoxy-deoxybenzoin and gingerdione-[2] showed significant masking effects of about 30 %. The masking pattern against 5 ppm quinine and 100 ppm salicin showed some similarities. ED was more active against naringenin compared to HED. Surprisingly, combinations of HED and sterubin showed suppression of the expected combined masking effects. Generally, at least one vanillyl moiety in combination with a 2,4-dihydroxyphenyl group seem to be mandatory for bitter masking pattern. The active molecules therefore may modulate at least one of the broadly tuned of the 25 human bitter receptors. (1) Ley, J. P. et al. *JAFC.* 2005, *53*, 6061 (2) Ley, J. P. et al. *JAFC* 2006, *54*, 8574 (3) Ley, J. P. et al. *JAFC* 2008, *56*, 6656

#P20 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Reliability of a Brief Spatial Test for Assessment of Gustatory Function

Susan E. Coldwell[1,2], Mark T. Drangsholt[1,2], Kimberly H. Huggins[1,2], Gayle Garson[1], Mary K. Scott[2], Mary K. Hagstrom[3], Linda LeResche[2]
[1]University of Washington Department of Dental Public Health Sciences Seattle, WA, USA, [2]University of Washington Department of Oral Medicine Seattle, WA, USA, [3]University of Washington Regional Clinical Dental Research Center Seattle, WA, USA

Assessment of gustation by application of tastants directly to the anterior tongue, along with whole-mouth testing, has been recommended for inclusion in the NIH Toolbox. This method can reveal damage to the chorda tympani branch of the facial nerve. However, data on test-retest reliability are lacking. As part of a larger case-control study, we assessed temporal stability of general Labeled Magnitude Scale (gLMS) ratings for training items (lights and sounds), tastants applied directly to the anterior tongue, and tastants given whole mouth. Participants were 18 patients with burning mouth syndrome (age 33 to 77 yrs, 1 male) and 24 controls (age 23 to 72 yrs, 6 male), both groups tested twice; and 24 controls (age 28 to 68 yrs, 4 male) tested once. Participants used the gLMS to rate the intensity of the room light, a dimly lit restaurant, and the brightest light ever seen. Next, participants rated intensity of a whisper, a conversation, and the loudest sound ever heard. A cotton swab was then used to directly apply tastants (1 M sucrose, 1 M NaCl, 0.032 M citric acid, 0.001 M quinine HCl) once each to the lateral, dorsal surface of the anterior right and then left of the tongue. After tongue testing, participants tasted each solution once by whole mouth. Testing was repeated 7 to 70 (median 15) days later. For participants tested twice, intraclass correlations (ICCs) between ratings ranged from 0.53 to 0.85 for lights and sounds. ICCs for tongue tip testing ranged from 0.46 to 0.69. ICCs for whole mouth testing ranged from 0.54 to 0.69. For the 48 controls, ICCs between gLMS ratings for the same tastant applied to the right and left sides of the tongue ranged from 0.87 to 0.91 within a session. This brief gustatory assessment method yields reasonable reliability. Acknowledgements: Supported by federal funds from the National Institute of Dental and Craniofacial Research, 3R21DE018768-02S1 (PI, Mark Drangsholt) and by Grant UL1RR025014 from the NIH National Center for Research Resources.

#P21 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Validation of Commercial PROP Taste Strips for the NIH Toolbox

Hetvi Desai[1], Susan E. Coldwell[2], James W. Griffith[3], Lloyd Hastings[4], Gregory S. Smutzer[1]
[1]Biology Department, Temple University Philadelphia, PA, USA, [2]Dental Public Health Sciences, University of Washington Seattle, WA, USA, [3]Department of Medical Social Sciences, Northwestern University Chicago, IL, USA, [4]Osmic Enterprises, Inc. Cincinnati, OH, USA

Edible taste strips allow the delivery of precise amounts of tastants to the oral cavity. The goal of this study was to validate the use of edible taste strips prepared to industry manufacturing standards for examining 6-*n*-propylthiouracil (PROP) bitter taste function in humans. This validation was carried out by presenting participants with 1 inch x 1.25 inch taste strips that contained 800 nmoles of PROP. Participants then reported the intensity and quality of the perceived taste. Taste intensity values for 800 nmole PROP strips were compared to three PROP and three NaCl solutions (Tepper *et al.*, *Physiol. & Behav*., 2001). Taste intensity results for PROP strips, PROP solutions, and NaCl solutions were then compared to haplotype analysis of the *TAS2R38* bitter taste receptor gene. Participants with at least one PAV allele readily discriminated 800 nmole PROP strips from control strips, and detected PROP strips as bitter tasting [$t(21) = 6.96$, $p < 0.001$]. For PROP tasters, 800 nmole PROP strips resulted in a general Labeled Magnitude Scale (gLMS; 0-100 scale) average value of 41 (n = 22). PAV/PAV subjects reported an average gLMS of 48 (n = 11), and PAV heterozygotes reported an average gLMS of 34 (n = 11). In contrast, non-tasters with an AVI/AVI genotype averaged 11 (n = 9) for the 800 nmole PROP strips. Control taste strips averaged 3 for all genotypes [$t(8) = 1.82$, NS]. Intensity ratings for 3.20 mmolar PROP solutions by PAV homozygotes or

heterozygotes was higher (60) than for PROP strips (t(21) = 4.16, p <0.001), whereas average gLMS rating for 0.32 mmolar PROP solutions by tasters was comparable to strips (37) [t(21) = 0.76, NS]. These results indicate that edible taste strips made to industry standards exhibit low background taste, and are suitable for examining PROP taster status in humans. Acknowledgements: Supported by federal funds from the Blueprint for Neuroscience Research and the Basic Behavioral and Social Science Opportunity Network (OppNet), National Institutes of Health under Contract No. HHS-N-260-2006-00007-C (PI, R. Gershon), and by NIDCD 2R44 DC007291.

#P22 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Development of an Electronic Tongue (ET) to Evaluate the Bitterness Intensity of Rx and OTC Formulations

Marie O. Richardson[1], Lisa A. Glover[1], Phil B. Stern[2], David Clapham[3], Ken A. Saunders[4], Andrey. Legin[5], Dmitry Kirsanov[5], Evgeny Legin[5], Boris Seleznev[5], Alisa Rudnitskaya[6]
[1]GlaxoSmithKline Consumer Healthcare Weybridge, United Kingdom, [2]GlaxoSmithKline Consumer Healthcare Parsippany, NJ, USA, [3]GlaxoSmithKline Pharmaceutical Ware, United Kingdom, [4]GlaxoSmithKline R&D Stevenage, United Kingdom, [5]University of St Petersburg St Petersburg, Russia, [6]University of Aveiro Aveiro, Portugal

Masking the bitterness of actives to make pharmaceutical formulations more palatable has long been a goal for GSK. To be able to mask the taste of a bitter component, one must be able to evaluate its bitterness intensity first. Sensory evaluation by a human panel is often a difficult exercise due to the lack of a full toxicology profile at early stages of development. Even once this profile is established, there is a desire to reduce the amount of human tasting to the minimum required. GSK has set up a partnership with the University of St Petersburg to develop an e-tongue with the aim of using the instrument in the evaluation of the bitter taste of pharmaceutical molecules. GSK needs a system with robust sensors to compare data across time, hence the choice to develop its own e-tongue. The bitterness intensity of 8 bitter tasting substances of different nature was evaluated by e-tongue and correlated with the values obtained by a sensory panel by PLS regression. The predicted MRE was 15 % (RMSE=0.72) with an error of only 5% (RMSE=0.27) for the ultra bitter active azelastine HCl. No prediction was possible for caffeine, paracetamol and KNO_3. The first two substances, being non ionic, were hard to detect by the sensors and no other inorganic salts were present in the calibration model for KNO_3. Accuracy in the bitterness prediction was achieved by lowering the original pH 7 to pH 6 and to pH4. The data obtained were merged into a three dimensional set (substance x sensors x pH) and the calibration model was calculated using 3-way PLS regression. The MRE became 12% (RMSE=0.51). The 3D model was not optimal for azelastine HCl (MRE = 22%) or naratriptan (MRE=23%) but is a promising approach for the prediction of the bitterness intensity using an e-tongue. Further developments are underway. Acknowledgements: none

#P23 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Citizen-Science in a Community-Based Chemoreception Laboratory

Meghan M Sloan, Devin Walecka, Keely B Sudhoff, Brian Hostetler, Nicole L Garneau, Bridget Coughlin
Denver Museum of Nature and Science Denver, CO, USA

The Denver Museum of Nature & Science is training, utilizing and educating citizen-scientists in a community-based laboratory. The research goal is to study the Genetics of Taste in the framework of health. Fifty-three enthusiastic volunteer citizen-scientists are investigating relationships between genetic ancestry, the ability to taste bitter substances and body composition. The citizen-scientists are involved in all aspects of the NIH-funded study and are certified by the agency to conduct human subject research. They recruit and enroll visitors to participate, collect background information, a DNA sample, body composition information, taste phenotype and a photograph of a blue-stained tongue. In addition, the Museum is training citizen scientists to analyze data using techniques such as DNA purifications, gel electrophoresis, PCR and UV-Vis spectrometry. To date, the citizen-scientists have enrolled over eleven hundred subjects. Importantly, over the course of 18 months, we have retained a 100% satisfaction rate in a museum-administered evaluation of the citizen-science program within the health sciences department. In conclusion, a community-based lab is viable and capable of producing competive scientific data. We have shown that citizen-scientists with non-science backgrounds can effectively and accurately articulate the basis of genetics, the intricacies of DNA, and physiology of taste papillae. This is a shining example of research being translated to the public domain. With the volunteer generated data, we can draw correlations between fungiform papillae density, body composition and genotype for the *TAS2R38* gene. Acknowledgements: R25 RR025066-02 NIH NCRR SEPA

#P24 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Expression of functional N-terminal domain of human T1R2 taste receptor

Maud Sigoillot, Elodie Maîtrepierre, Laurence Le Pessot, Loïc Briand
Centre des Sciences du Goût et de l'Alimentation, UMR6265 CNRS, UMR1324 INRA, Université de Bourgogne Dijon, France

The sweet taste receptor is a heterodimer composed of two distinct subunits called T1R2 and T1R3 (class C of G protein-coupled receptors). These subunits possess a large extracellular N-terminal domain (NTD) linked to the seven-transmembrane domain by a shorter cysteine-rich region. T1R2 NTD has been shown to interact with sucrose and some non-caloric sweeteners like sucralose, aspartame and neotame. However, the binding properties of T1R2 NTD remain largely unknown. To study the binding specificity of T1R2 subunit, a large amount of purified NTDs is suitable for biochemical and structural studies. Here, we report the successful expression and characterization of the human T1R2 NTD. The protein was overexpressed using

Escherichia coli as insoluble aggregated protein (inclusion bodies). The protein was solubilized and in vitro refolded using suitable buffer and additives. T1R2 NTD was then purified and characterized. Circular dichroism and fluorescence spectroscopy demonstrated that T1R2 NTD is properly refolded and able to bind sweet compounds with physiological relevant affinities. Owing to the large amount of produced protein, T1R2 NTD binding properties have been investigated using isothermal titration microcalorimetry. Interestingly, thermodynamic data revealed distinct mechanisms of binding for some sweeteners. Our expression strategy will allow large-scale production of human functional T1R2 NTD suitable for cristallographic studies.
Acknowledgements: This work was supported by INRA and Burgundy council (Région Bourgogne) grants to E.M. & M.S.

#P25 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Generation and characterization of T1R2-LacZ knock-in mouse

Ken Iwatsuki[1], Reiko Ichikawa[1], Masatoshi Nomura[2], Atsushi Shibata[2], Hisayuki Uneyama[1], Kunio Torii[1]
[1]Institute for Innovation, Ajinomoto Co. Inc. Kawasaki, Japan, [2]Dep. Med. and Bioregulatory Science, Kyushu University Fukuoka, Japan

Taste cells are chemosensory epithelial cells that sense distinct taste quality such as umami, sweet, bitter, sour and salty. Taste cells utilize G protein-coupled receptors to detect umami, sweet and bitter taste whereas ion channels are responsible for detecting salty and sour taste. Among these taste receptors, taste receptor type 2, T1R2 (or Tas1r2), has been identified as a sole sweet taste receptor in mammals that mediates sweet signals upon dimerization with T1R3. However, because of limited availability of reliable antibodies and low expression level of G protein-coupled receptors, it is uneasy to identify the cell-types that express these receptors in non-taste tissues. In this study, we have generated a T1R2-LacZ reporter knock-in mouse to investigate tissue distribution of T1R2 at a single-cell level. We found that the LacZ gene expression in these mice was faithful to the expression of T1R2 in the taste tissue and in the gastrointestinal tract where T1R3 expression has been reported. Surprisingly, T1R2 expression was also found in the testis. Mice homozygous for *T1R2* deletion lacked T1R2 protein analyzed by the antibody raised against T1R2 peptide sequences. In summary, the T1R2 knock-in mouse is a powerful tool to analyze the putative targets for sweeteners as well as to study the physiological roles of T1R2 in detecting sugars.

#P26 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Molecular Modeling of the Human T1R2 Venus Fly Trap Domain

Silvia Osuna[1], Marcel Swart[1,2], Miquel Sola[1], Eugeni Roura[3]
[1]Institut de Quimica Computacional i Departament de Quimica, Universitat de Girona Girona, Spain, [2]Institucio Catalana de Recerca i Estudis Avancats (ICREA) Barcelona, Spain, [3]Centre for Nutrition and Food Sciences, QAAFI, The University of Queensland Brisbane, Australia

Sweet taste has evolved to enhance intake of dietary carbohydrates but sweet perception varies across species reflecting the adaptation to ecological niches and diets. The development of a molecular model for the human sweet heterodimer receptor T1R2/T1R3 will be a usefull tool to help understand the receptor-ligand interactions and ultimately in predicting species differences. Reportedly the main ligand binding site is located in the extracellular N-terminal Venus Fly Trap domain (VFTD) of the T1R2 subunit. We have developed a molecular model of the hT1R2 VFTD using QuantumBioChemistry program and the Amber force field. Our model predicts that the charge of the candidate sweetener determines the affinity to the hT1R2 VFTD. Cationic ligands bind strongly, neutral compounds (i.e. sugars) show weaker binding and the negatively charged sweeteners are repelled. The study of the interactive amino acid residues from the VFTD led us to locate two different binding sites: an inner site at the joint of the VFTD and an outer site similar to what has been described in the umami T1R1. The inner binding site of our model contains most of the amino acid residues described in previous studies but also some residues not identified before such as the ARG317 that we believe may play an important species-dependent role. The outer binding site is able to bind several compounds almost as strongly as the inner binding site. Moreover the model predicts a synergistic effect resulting from a compounds binding the outer site when a ligand is already present in the inner site. In that perspective our results also confirm previous publications. In summary, we report the involvement of ARG317 in the hT1R2 VFTD and the existence of two ligand binding sites that function in a similar fashion as the umami T1R1.
Acknowledgements: MICINN (CTQ2008-03077/BQU, CTQ2008-06532/BQU), DIUE (2009SGR637, 2009SGR528)

#P27 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Transgenic Mice Expressing a Humanized Taste Receptor

Kevin M. Redding, Robert F. Margolskee, Bedrich Mosinger
Monell Chemical Senses Center Philadelphia, PA, USA

T1R3, a critical subunit of the sweet and umami (amino acid) sensing receptors, is expressed in taste cells, enteroendocrine cells of the gastrointestinal tract, pancreatic islets, testis and brain. To get insights into the function of the human receptor *in vivo* we generated transgenic animals expressing a humanized form of T1R3. Humanized T1R3 was introduced into a genetically engineered mouse line that lacks endogenous T1R3 (T1R3KO). Therefore only the humanized receptor is present in the mouse and any responses would reflect the function of the human type receptor. In contrast to mouse T1R3, the humanized receptor can be blocked by human-specific inhibitors such as lactisole, fibrates and phenoxy herbicides and activated by the human-specific sweetener cyclamate. These animals will serve as a model and tool to study in detail long-term metabolic, endocrine and developmental effects of human-specific compounds, a task that is not possible in unmodified rodents and difficult if not impossible to conduct directly in humans. The DNA construct used to generate transgenic animals also contains a GFP marker so that we can easily detect where T1R3 is expressed. We have established two transgenic lines and are currently analyzing the expression of the transgene in various tissues and their behavioral responses to sweeteners.

#P28 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Ablation of type I taste bud cells

Feng Li[1], Jie Cao[1], Dieter Riethmacher[2], Minliang Zhou[1], Liquan Huang[1]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]University of Southampton, Human Genetics Division Southampton, United Kingdom

Type I cells constitute the most abundant group of cells in mammalian taste buds. These cells are known to express glutatmate-aspartate transporter (GLAST) and nucleoside triphosphate diphosphohydrolase (NTPDase) that remove and degrade excess transmitters from intragemmal space. Thus these cells are believed to play a supportive role in taste bud structure and physiology. Recent studies, however, indicate that these cells may also be involved in the response to salty stimuli. To further investigate the contributions of these type I cells to taste bud structure, salty taste sensation and signal transmission regulation, we set out to generate transgenic mice containing type I cells that can be inducibly ablated. We prepared a transgene construct by replacing the coding sequence for mouse NTPDase2 in a bacterial artificial chromosome (BAC) clone with the sequence for the reverse tetracycline-controlled transactivator (rtTA), which then was injected into fertilized mouse eggs. Founder mice carrying this transgene (ENT2-rtTA) were bred with B6 mice and progeny crossed with transgenic lines carrying TetO-Cre, Rosa26-stop-lacZ, Rosa26-stop-DTA or TetO-DTA (diphtheria A toxin). Histological analysis indicated that following the induction of rtTA activity with doxycycline, the triple transgenic (ENT2-rtTA/TetO-Cre/Rosa26-stop-lacZ) mice expressed beta-galactosidase in the taste buds and nerve fibers that have been shown to express NTPDase2, whereas the bi-transgenic (ENT2-rtTA/TetO-DTA) mice displayed altered gene expression patterns. Further characterization of these mice will provide additional information on type I cells' role in taste bud structure and function. Acknowledgements: Supported by a grant from NIH/NIDCD (DC007487).

#P29 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Unconditioned licking responses to glucose, maltose and maltotriose but not Polycose, are severely blunted in mice lacking T1R2, T1R3 or both protein subunits

Yada Treesukosol, Kimberly R. Smith, Alan C. Spector
Department of Psychology & Program in Neuroscience, Florida State University Tallahassee, FL, USA

We previously reported that T1R2 or T1R3 knockout (KO) mice display concentration-dependent licking of Polycose, a mixture of glucose polymers of various chain lengths, in brief access taste tests (25 min, 5-s trials). Here we assessed whether there was a gradient of stimulus effectiveness related to glucose chain length in WT, T1R2 KO, T1R3 KO, and T1R2/3 double KO mice. On the first test session, whereas WT mice receiving glucose, maltose (2 glucose units), maltotriose (3 glucose units) or Polycose all displayed sigmoidally increasing concentration-response functions, the functions for all KO groups were flat except those for Polycose. But by the third test session, some T1R2 KO and T1R3 KO mice showed concentration-dependent responses to maltose and maltotriose suggesting that testing experience influenced responsiveness. A smaller subset of T1R2/3 double KO mice showed concentration-dependent licking of maltotriose in the third session. The number of trials T1R2 KO and T1R3 KO mice initiated to Polycose did not significantly differ from those of their WT controls, but was significantly lower for T1R2/3 KO mice. All KO genotypes displayed virtually flat concentration-response functions for saccharin and monotonically rising functions for Polycose in further testing. These findings are consistent with the view that the T1R2+3 heterodimer is the principal taste receptor for "sweet" ligands. Polycose may be weakly stimulating the T1R2+3 receptor, but the data support the hypothesis that a novel receptor(s), with an optimal ligand greater than 3 glucose moieties, may mediate polysaccharide taste. These results also suggest that KO mice can associate remaining oral cues with positive postingestive events even in a brief access test when repeatedly tested across sessions with a caloric compound. Acknowledgements: Supported by NIH R01-DC004574 to A.C.S.

#P30 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

The A2B adenosine receptor is required for sweet taste in posterior tongue

Sue C. Kinnamon[1,3], Arian Baquero[1,3], Shinji Katoaka[2,3], Nicole Shultz[2,3], Thomas E. Finger[2,3]
[1]University of Colorado Denver/Otolaryngology Aurora, CO, USA, [2]University of Colorado Denver/Cell & Developmental Biology Aurora, CO, USA, [3]Rocky Mountain Taste and Smell Center Aurora, CO, USA

ATP is released from Type II taste cells in response to bitter, sweet, and umami taste stimuli to activate ionotropic P2X receptors on afferent nerve fibers and metabotropic P2Y receptors on adjacent taste cells. The released ATP is degraded to ADP by NTPDase2 on Type I taste cells and is further broken down to adenosine by intrinsic nucleotidases. Adenosine can further act on one or more metabotropic adenosine receptors: A1, A2A, A2B, and A3. We recently reported that the adenosine receptor A2B is expressed on a subset of taste cells (Katoaka & Finger, Achems Abs., 2008). Here, we have used RT-PCR, in situ hybridization, and immunocytochemistry on A2BR KO/LacZ mice to show that A2B is exclusively expressed in posterior tongue, and is co-localized with the subset of taste cells that expresses Galpha14, likely the sweet-responsive Type II taste cells (Tizzano et al., BMC Neurosci. 2008 9:110). To test whether A2B affects taste function, we have recorded from the glossopharyngeal (GL) and chorda tympani (CT) nerves of A2BR KO and wildtype (WT) mice. Surprisingly, A2BR KO mice completely lacked GL nerve responses to sucrose (300 mM, 500 mM, and 1M), although responses to citric acid (10 mM), NaCl (100 mM), quinine (30 mM) and NH4Cl (100 mM) were normal. Chorda typani nerve responses of KO mice to all stimuli, including sucrose, were not different from responses in WT mice. These data suggest that the A2BR is required for sweet taste function in posterior tongue. Further experiments will be required to determine if adenosine acts in an autocrine fashion to potentiate the release of ATP, or directly to modulate the sweet taste transduction mechanism. Acknowledgements: DC007495, DC006021, and P30 DC004657

#P31

POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE

Mice lacking T1R3 exhibit impaired glucose tolerance and a deficient insulin response

Tatsuyuki Takahashi[1], C. Shawn Dotson[2], Maartje C. P. Geraedts[1], Steven D. Munger[1,3]
[1]Department of Anatomy & Neurobiology, University of Maryland School of Medicine Baltimore, MD, USA, [2]Departments of Neuroscience and Psychiatry, University of Florida College of Medicine & Center for Smell and Taste, University of Florida Gainesville, FL, USA, [3]Department of Medicine, Division of Endocrinology, Diabetes and Nutrition, University of Maryland School of Medicine Baltimore, MD, USA

T1R family receptors expressed in taste bud cells are essential for the normal detection of sweet and umami taste stimuli. However, these receptors are also found in other tissues including several endocrine cell types that contribute to the control of glucose homeostasis (e.g., glucagon-like peptide-1 (GLP-1)-secreting intestinal enteroendocrine L cells and insulin-secreting pancreatic β cells). Indeed, T1R3 has been implicated in the glucose-dependent secretion of GLP-1 (Kokrashvili, 2009), which can impact insulin biosynthesis and secretion. However, the role that T1Rs play in glucose homeostasis remains unclear. To better understand the contribution of T1Rs to this process, we measured blood glucose and insulin levels (by glucose meter and ELISA, respectively) before and during a 2-hr oral glucose tolerance test in T1r3 $^{+/+}$, $^{+/-}$ and $^{-/-}$ mice. Animals (4 mo. old) were fasted for 18 hr prior to the test. Blood samples were obtained from tail bleeds prior to and at several timepoints after a glucose gavage (5 g/kg). Resting glucose and insulin levels were identical across genotype, as were body weight and food intake. However, glucose levels post gavage were higher in T1r3$^{-/-}$ mice (P=0.04) and showed a delay in recovery from peak, while insulin levels were significantly lower (P=0.005) and did not exhibit the initial peak (10-40 min) observed in $^{+/+}$ and $^{+/-}$ littermates. These results, which are consistent with those from α-gustducin null mice (Jang, 2007), suggest that T1R3 plays a critical role in the regulation of glucose homeostasis. Acknowledgements: NIDCD (DC010110) Ajinomoto Amino Acid Research Program

#P32

POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE

Characterization of β-D-Glucopyranoside Binding Site of Human Bitter Taste Receptor hTAS2R16

Takanobu Sakurai[1,2], Takumi Misaka[2], Masaji Ishiguro[3], Yohei Ueno[2], Shinji Matsuo[1], Yoshiro Ishimaru[2], Tomiko Asakura[2], Keiko Abe[2]
[1]General Research Institute of Food Science and Technology, Nissin Foods Holdings Co., Ltd. Shiga, Japan, [2]Department of Applied Biological Chemistry, Graduate School of Agricultural and Life Sciences, The University of Tokyo Tokyo, Japan, [3]Niigata University of Pharmacy and Applied Life Sciences Niigata, Japan

Humans recognize thousands of bitter compounds by only 25 members of the hTAS2R family of G-protein-coupled receptors (GPCRs). However, structural information on these receptors is limited. To address the molecular basis of bitter tastant discrimination by hTAS2Rs, we performed ligand docking simulation and functional analysis using a series of point mutants of hTAS2R16 as a receptor for β-D-glucopyranosides including salicin. The docking simulation using a structural model of rhodopsin photointermediate, metharodopsin, as template predicted two types of candidate binding structures for a salicin-hTAS2R16 complex. In both models, salicin was located in the same putative binding pocket formed by TM3, TM5, and TM6 with different binding modes. Mutational experiments revealed a probable salicin-hTAS2R16 binding mode and some amino acid residues involved in the recognition of salicin[1)]. On the other hand, gentiobiose, which is disaccharide, composed of two D-glucoses with β-(1-6) glycosidic linkage is known to elicit bitterness although it is a sugar. Since gentiobiose is regarded as one of β-D-glucopyranosides, the participation of hTAS2R16 in this bitter taste sensation was predicted. Functional analysis showed that gentiobiose activated hTAS2R16, but not human sweet taste receptor hT1R2/hT1R3[2)]. Also, our mutational experiments using several β-D-glucopyranosides, including gentiobiose suggested that hTAS2R16 activation was especially induced by the recognition of the cytoplasmic side of each D-glucose moiety located in the hTAS2R16 binding site[1,2)]. This finding can explain why hTAS2R16 recognizes many kinds of bitter β-D-glucopyranosides. 1) Sakurai et al., J. Biol. Chem. 285 (2010) 28373-28378. 2) Sakurai et al., Biochem. Biophys. Res. Commun. 402 (2010) 595-601. Acknowledgements: This study was supported by the Research and Development Program for New Bio-industry Initiatives

#P33

POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE

Molecular evolution of a bitter taste receptor gene in primates

Stephen P. Wooding
McDermott Center for Human Growth and Development, University of Texas Southwestern Medical Center Dallas, TX, USA

Bitter taste receptors (TAS2Rs) play a key role in enabling animals to avoid toxins in the environment, especially toxic defense compounds synthesized by plants. The importance of this role suggests that TAS2Rs have likely have been under strong pressures from natural selection. To investigate these pressures, we used molecular evolutionary analyses to test for signatures of selection in TAS2R38, which responds to plant toxins with potent anti-thyroid properties. Whole-gene sequences collected from 40 diverse primate species exhibited high levels of diversity including coding and non-coding nucleotide substitutions, premature stop signals, and frameshifts. The overall relative rate of coding (i.e., amino acid changing) mutation, ω, was high in primate *TAS2R38* (ω=0.62) compared to other genes, but was significantly lower than expected under neutrality ($p<4.0x10^{-9}$), indicating that purifying natural selection has preserved the basic structure of TAS2R38 throughout primate history. Comparisons among functional domains revealed low rates of change in internal loops (ω=0.52; $p<7.0x10^{-5}$), which mediate interactions with downstream components of the taste transduction cascade, and high rates in external loops (ω=1.19), which mediate ligand recognition, suggesting that the receptor has adaptively altered its responses to ligands over time. Differences were also present

among species groups. While mutation rates were significantly constrained in New World monkeys and cercopithecines, they were consistent with neutral expectations in apes and leaf-eating monkeys, suggesting that selection pressures on TAS2R38 may be moderated by cognitive or physiological variables. Taken together, signatures of natural selection on TAS2R38 in primates point to a history of rapid yet restrained adaptation to toxic challenges.

#P34 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Taste Function in Mice with a Targeted Mutation of the *Gpr113* Gene

Theodore M. Nelson[1], Natalia Bosak[1], Nelson D. LopezJimenez[2], Lino Tessarollo[3], Masashi Inoue[4], Susan L. Sullivan[2], Alexander A. Bachmanov[1]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]Laboratory of Molecular Biology, National Institute on Deafness and Other Communication Disorders, National Institutes of Health Rockville, MD, USA, [3]Neural Development Group, Mouse Cancer Genetics Program, Center for Cancer Research, National Cancer Institute Frederick, MD, USA, [4]Laboratory of Cellular Neurobiology, Department of Life Science, Tokyo University of Pharmacy and Life Science Tokyo, Japan

Gpr113 encodes a G-protein-coupled receptor (GPCR) belonging to family 2B, characterized by a long N-terminal domain including a hormone-binding domain. GPR113 expression is restricted to a subset of taste receptor cells that express TPRM5 and T1R3, but not T1R1, T1R2, or gustducin. Hormone-binding domains are a common feature of several GPCRs that bind various hormone peptides, suggesting one ligand for GPR113 may be a peptide. Due to its long extracellular domain it is probable that GPR113 is involved in additional interactions. Because GPR113 is expressed in taste receptor cells, candidate ligands for GPR113 include both endogenous peptides and environmentally encountered taste stimuli. In order to assess the importance of GPR113, we genetically engineered mutant mice with a disrupted *Gpr113* gene and examined taste function of these mice using behavioral and neurophysiological approaches. We measured preference ratios for concentration series of citric acid, NaCl, inosine monophosphate, quinine, sucralose, $CaCl_2$, glycine, monosodium glutamate, ethanol, and Polycose in 48-h two bottle tests. We found no significant differences in behavioral taste responses between *Gpr113* mutant animals and wild-type controls. Additionally, electrophysiological recordings of taste-evoked activity in both the chorda tympani and glossopharyngeal nerves reveal that mutant mice have unaltered responses to a variety of taste stimuli representing all major qualities. In conclusion, disruption of *Gpr113* does not alter behavioral and neural taste responses. Our results suggest that GPR113 likely does not function as a receptor for taste ligands, but do not preclude its involvement in the detection of endogenous peptide hormones. Acknowledgements: Supported by NIH grant R01 DC00882 (AAB).

#P35 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Effect of cAMP on the NaCl Chorda Tympani (CT) Taste Nerve Response Profile of Young Rats

Vijay Lyall, Tam-Hao T. Phan, Shobha Mummalaneni, John A. DeSimone
Virginia Commonwealth University/Physiology & Biophysics Richmond, VA, USA

In rats, the benzamil (Bz)-sensitive NaCl response is not present at birth and develops between roughly 10 and 45 days of age. The increase in gustatory Na^+ sensitivity is most likely due to a progressive addition of newly synthesized functional Bz-sensitive epithelial Na^+ channels (ENaCs) or the redistribution of the a, b and g ENaC subunits from the intracellular to apical domain. However, in the developing rats, the signaling mechanisms that are involved in the incorporation of functional ENaCs in the apical membrane of taste cells have not been investigated. We hypothesize that an increase in taste cell cAMP is an essential step in the conversion of the CT response profile observed in young rats to the CT response profile observed in adult rats. To test this hypothesis, NaCl CT responses were monitored in anesthetized young rats before and after topical lingual application of a membrane permeable cAMP, 8-chlorophenylthio (CPT)-cAMP. CT responses were recorded under zero current clamp or under lingual voltage clamp conditions. Our results show that NaCl CT responses in 19 to 23 days old rats are Bz-insensitive but are blocked by cetylpyridinium chloride (CPC), a specific modulator of the TRPV1t, a putative non-specific salt taste receptor. Topical lingual application of 8-CPT-cAMP (2.5-20 mM) for 10 min induced a CPC-insensitive but Bz-sensitive increase in the NaCl CT response and the apical Na^+ conductance in taste cells calculated by increased voltage-sensitivity of the NaCl CT response post-cAMP treatment. These results suggest that an increase in taste cell cAMP level is required for the incorporation of active Bz-sensitive ENaCs in the apical membrane of salt sensing taste cells in young rats. Acknowledgements: Supported by the National Institute on Deafness and other Communication Disorders grants (DC-000122 and DC-005981) to VL.

#P36 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Sour Taste Stimulates GABA Secretion from Mouse Presynaptic (Type III) Taste Cells

Yijen A. Huang[1], Stephen D. Roper[1,2]
[1]Department of Physiology & Biophysics, Miller School of Medicine, University of Miami Miami, FL, USA, [2]Program in Neuroscience, Miller School of Medicine, University of Miami Miami, FL, USA

Cell-to-cell communication in taste buds plays an important role in taste transduction. For instance, serotonin, a paracrine transmitter in taste buds, exerts negative feedback onto, and inhibits ATP secretion from Receptor (Type II) cells. Recently, γ-aminobutyric acid (GABA), its synthetic enzymes, and GABA receptors were identified in taste buds (Cao et al., 2009; Starostik et al., 2010; Dvoryanchikov, Huang et al., 2011). GABA reduced taste-evoked ATP secretion from Receptor cells and was

considered to be an inhibitory transmitter in taste buds. However, to date, the stimuli that trigger GABA release and the identity of the taste cells that release GABA are not well understood. In the present study, CHO cells transfected with $GABA_B$ R1 receptors ("GABA biosensors") were loaded with Fura 2 to detect GABA released from isolated mouse taste buds and taste cells. Biosensors responded (Ca^{2+}) to low concentrations of GABA (30~100 nM) but not to KCl depolarization (50 mM), taste compounds (cycloheximide, saccharin, denatonium, SC45647, or sucralose), or acetic acid (10 mM, pH 5.0). Taste buds released GABA when they were stimulated with bath-applied KCl or acetic acid. CGP55845, a $GABA_B$ receptor antagonist, reversibly abolished these Ca^{2+} responses in the biosensors and verified that taste buds had released GABA. In contrast to acid stimulation, sweet and bitter taste compounds did not evoke GABA release from taste buds. Lastly, we isolated and identified Presynaptic (Type III) cells from GAD1-GFP mice. KCl and acetic acid, but not ATP (up to 10 mM), triggered GABA release from these cells. The data indicate that sour taste stimulates Presynaptic cells to release GABA, ultimately inhibiting Receptor (Type II) cells. Acknowledgements: Supported by NIH/NIDCD 1R01DC007630 (SDR).

#P37 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Effect of guanosine monophosphate (GMP) on taste perception of L-amino acids and D-Ala by mice

Yuko Murata[1], Alexander A. Bachmanov[2]
[1]National Research Institute of Fisheries Science, FRA Yokohama, Japan, [2]Monell Chemical Senses Center Philadelphia, PA, USA

In vitro heterologous expression studies showed that most L-amino acids and D-Ala activate the mouse T1R1+T1R3 receptor when they are mixed with IMP, even though some of these amino acids do not activate T1R1+T1R3 without IMP (Nelson *et al*., 2002). Consistent with this, our previous studies with mice showed that conditioned taste aversion (CTA) to L-Met, L-Val or D-Ala (without GMP or IMP) did not generalize to MSG mixtures, but CTA to L-Met, L-Val and D-Ala mixed with IMP generalized to MSG mixtures. Like IMP, GMP also has synergistic effects on umami taste of MSG in humans (Yamaguchi, 1967) and on taste responses of the rat chorda tympani nerve to various amino acids (Yoshii, 1987). The goal of our study was to examine whether addition of GMP changes taste quality perception of L-Met, L-Val and D-Ala. We have addressed this question using the CTA technique. Separate groups of C57BL/6J mice were exposed to one of three conditioned stimuli (50 mM L-Met + 2.5 mM GMP, 50 mM L-Val + 2.5 mM GMP, 50mM D-Ala + 2.5mM GMP) or to water (control) and injected with LiCl to form CTA. Conditioned mice were presented with the conditioned stimuli, and a mixture of 50 mM MSG and μ30 M amiloride (Ami; added to block sodium taste) with or without 2.5 mM IMP (i.e., MSG+IMP+Ami and MSG+Ami), and their lick responses were recorded. CTA to each of these amino acids mixed with GMP generalized to MSG+Ami and MSG+IMP+Ami. These results suggest that addition of GMP changes the taste quality of L-Met, L-Val and D-Ala in a fashion similar to effects of IMP. Acknowledgements: Supported by Fisheries Research Agency (Yokohama, Japan) research grant (YM) and NIH grant DC 00882 (AAB)

#P38 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Molecular and Cellular Pathways of NaCl perception in *C. elegans*

Gert Jansen, Oluwatoroti Umuerri, Martijn Dekkers, Renate Hukema
Erasmus MC, Dept of Cell Biology Rotterdam, Netherlands

The nematode *C. elegans* is attracted to 0.1 to 200 mM NaCl and avoids higher NaCl concentrations. However, after prolonged exposure to NaCl the animals avoid all concentrations of NaCl, a behavior called gustatory plasticity. Attraction to NaCl is mainly mediated by one pair of sensory neurons, ASE, and involves cGMP and Ca^{2+} signaling using the guanylate cyclases GCY-14 and GCY-22, the cyclic nucleotide gated (CNG) channel TAX-2/TAX-4 and calcineurin TAX-6/CNB-1. Osmotic avoidance is mediated by the ASH neurons and requires the G subunit ODR-3 and the TRPV channel subunit OSM-9. We have identified six new genes involved in attraction to NaCl, which were positioned in two genetic pathways. One pathway involves *tax-2*, *tax-4*, *tax-6* and the MAPK genes *nsy-1* and *sek-1*. The second pathway consists of *tax-2* and the CNG channel subunit *cng-3*, *osm-9*, *odr-3* and *gcy-35*. The behavior of these "salt-taste" mutants and our gustatory plasticity mutants suggests that *C. elegans'* response to NaCl is determined by a balance between attraction and avoidance. Using Ca^{2+} imaging we found that the ASE neurons of naïve animals respond to both low and high NaCl concentrations. The ASH neurons only respond to high NaCl concentrations. However, after prolonged exposure to NaCl, the ASE neurons are desensitized, while the ASH neurons are sensitized. Sensitization of ASH requires signals from the ASE neurons. Our results suggest that naïve *C. elegans* are attracted to all NaCl concentrations, predominantly mediated by ASE, but that this attraction is overruled by osmotic avoidance, mediated by ASH. Pre-exposure to 100 mM NaCl in the absence of food, sensed by the ASE and other neurons, changes this core circuit, resulting in desensitization of attraction and sensitization of avoidance. Acknowledgements: This work is supported by a grant from NWO/ALW.

#P39 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

The effect of calcium-sensing receptor agonists on taste responses to calcium solutions in mice

Chandra M. Cherukuri[1], Nathan L. Roach[1], Micheal G. Tordoff[2], Stuart A. McCaughey[1]
[1]Ball State University Muncie, IN, USA, [2]Monell Chemical Senses Center Philadelphia, PA, USA

The calcium-sensing receptor (CaSR) is found in taste tissue in rodents, where it may be involved in gustatory transduction of calcium ions. CaSR has also been implicated in the mediation of taste enhancement ("kokumi" taste) in humans. We performed electrophysiological experiments in C57BL/6J mice so as to test CaSR's role in calcium taste more specifically. Measurements were made of responses evoked in the chorda tympani (CT) nerve by $CaCl_2$ and other solutions applied to the oral cavity. We examined the effects of two CaSR agonists, spermine and glutathione. Glutathione at 100 μM had negligible effects on taste-evoked CT

responses. Spermine at 330 μM caused a generalized decrease in responsiveness of the CT, as evidenced by smaller responses to an NH_4Cl reference stimulus. There was also a larger and more specific suppression of CT responsiveness, after factoring out the generalized effect, which was limited primarily to solutions containing calcium or magnesium. This Ca/Mg-specific effect was especially robust for the later, tonic portion of the response. Our results do not support a role for CaSR in mediating taste enhancement (i.e., kokumi taste). Our spermine data were complex and allow for a possible role of CaSR in calcium taste, although additional work will be needed to clarify important issues, including spermine's generalized effect.
Acknowledgements: Funding provided by NIH RO1 DK-46791

#P40 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Cyclophosphamide Interference of Taste Functions of Mice
Nabanita Mukherjee, Eugene R. Delay
Dept. Biology, University of Vermont Burlington, VT, USA

Cyclophosphamide (CYP) is a commonly prescribed chemotherapy drug which is a DNA alkylating agent attacking guanine base pairs and interfering mainly with the S-phase of the cell cycle. It has adverse side effects including loss or alterations in taste sensation which can lead to malnutrition and poor recovery. Sensory cells within taste buds have a high turnover rate, hence may be susceptable to chemotherapy drug treatment. We hypothesize that CYP damages the DNA of taste progenitor cells and transitory amplifying cells, causing these cells to die or arrest their cell cycle until DNA repair is completed, thereby disrupting the normal replacement of sensory cells within taste buds. We tested this hypothesis using behavioral and immunohistochemical methods. We trained C57BL/J6 mice to discriminate between the tastes of monosodium glutamate and inosital monophosphate, then injected the mice with CYP (75 mg/kg, IP). They were then tested for 16 days and found that discrimination performance was disrupted for up to 4 days after injection, then again 9-12 days after injection. BrdU labeling of lingual epithelium revealed a disturbance of proliferative cells in response to CYP which appears to delay renewal of functional taste cells. H and E staining and DIC images of tongue sections revealed elongated spaces and fewer taste cells within taste buds after CYP injection. Double labelling with keratinocyte markers K8 and K14 indicated that the turnover rate of the cells is affected. These results suggest that normal cell renewal is affected after CYP injection. H and E staining of Von-Ebner glands revealed minimum changes in morphology of the gland after the injection. These findings support our hypothotheis that CYP interfers with taste by disrupting the normal cell replacement cycle of taste buds. Acknowledgements: Vermont Genetics Network through Grant P20RR16462 and by NSF IBS Grant to ERD.

#P41 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Defects in the Taste Structure and Function in MRL/lpr Autoimmune Disease Mice
Agnes Kim, Pu Feng, Tadahiro Ohkuri, Daniel Sauers, Zachary J. Cohn, Jinghua Chai, Theodore Nelson, Alexander Bachmanov, Joseph Brand, Liquan Huang, Hong Wang
Monell Chemical Senses Center Philadelphia, PA, USA

Many pathological conditions associated with inflammation can alter taste sensation. The mechanism by which inflammation contributes to taste dysfunction remains largely unknown. Recently we showed that acute inflammation induced by lipopolysaccharide altered the gene expression profile and inhibited progenitor cell proliferation in the taste epithelium. Here, we report our findings on the peripheral taste structure and taste function in MRL/lpr mice, a model for autoimmune disease with chronic inflammation. MRL/lpr mice develop a spontaneous autoimmune disease that resembles systemic lupus erythematosus in human. The taste tissues of MRL/lpr mice showed increased infiltration of T lymphocytes and elevated expression of several inflammatory cytokines, including TNF-alpha and interferon-gamma. Histological studies revealed that taste buds in the circumvallate papillae of MRL/lpr mice were smaller than those of wild type congenic control (MRL/+/+) mice. Real-time RT-PCR analysis showed that the expression levels of several type II taste cell markers, such as gustducin, TrpM5, and NeuroD, were significantly lower in the taste epithelia of MRL/lpr mice than in control mice. On the other hand, the expression levels of the type III cell markers SNAP25 and PKD2L1 remained comparable in MRL/lpr and control mice. Immunohistochemical analysis showed a significant reduction in the number of gustducin-positive taste receptor cells in the taste buds of MRL/lpr mice. In brief-access tests, MRL/lpr mice exhibited decreased responsiveness to bitter, sweet, and umami compounds, but normal responsiveness to salty and sour compounds. These results suggest that in MRL/lpr mice the autoimmune disease, accompanied by chronic inflammation, selectively affects the structure and function of type II taste receptor cells.
Acknowledgements: This study is supported by NIH/NIDCD grants DC010012 and DC007487.

#P42 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Taste Receptor Gene Expression in Patients with Taste Disorders
Ryoji Hirai, Minoru Ikeda, Keiko Onoda
Department of Otolaryngology- Head & Neck Surgery, Nihon University School of Medicine. Tokyo, Japan

Purpose We have investigated the expression of taste receptor genes (*T2R* and *TAS2R*) in tongue tissue using RT-PCR method. The purpose of this study was to examine changes in the expression of taste receptor genes in patients who had loss of taste and those who had phantogeusia. **Methods** The tongue tissue was collected from the foliate papillae by a simple scraping method, and total RNA was extracted using TRIzol. The reverse transcription reaction was performed using SuperScript3 and

PCR was performed using Ex Taq. The electrophoresis was done using an Agilent 2100 Bioanalyzer and the presence or absence of gene expression of *T2R3, 8, 9, 10, 13, 16, TAS2R40, 42, 43* and *48* was examined. Statistical analysis was performed by using Fisher's exact probability test. **Subjects** The subjects of this study consisted of 51 patients with loss of bitter taste and 43 patients with phantogeusia. A control group was also included consisting of 24 subjects with normal taste test results and no symptoms of taste disorders. **Results** The patients with loss of bitter taste showed a significant decrease in the frequency of expression of taste receptor genes *T2R8* (p=0.002), *10* (p=0.001), *13* (p=0.003), *16* (p<0.001), *TAS2R40* (p<0.001), and *48* (p=0.007) compared to the control group. The frequency of gene expression of *TAS2R42* (p=0.008) and *T2R3* (p=0.033) were significantly increased in patients with phantogeusia. **Conclusion** Among the 10 genes examined in this study, expression of some genes was decreased in patients with loss of taste while expression of other genes was increased in patients with phantogeusia, and conflicting changes in gene expression were observed. It was suggested that conflicting changes in taste receptor gene expression were involved in the pathogenesis of the loss of taste and phantogeusia. Acknowledgements: Our study was supported by a Grant-in-Aid for Scientific Research (C) from the Ministry of Education, Science, Culture, and Sports in Japan (No. 22591922), and a Nihon University Composite Research Grant (No. 08-024).

#P43 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Cellular Mechanisms of Taste Cell Loss Following Head & Neck Irradiation

Ha M. Nguyen[1,2], Brendan W. Ross[1,2], Mary E. Reyland[3], Linda A. Barlow[1,2]
[1]Dept of Cell & Developmental Biology, University of Colorado Anschutz Medical Campus Aurora, CO, USA, [2]Rocky Mountain Taste and Smell Center, University of Colorado Anschutz Medical Campus Aurora, CO, USA, [3]Dept of Craniofacial Biology, University of Colorado Anschutz Medical Campus Aurora, CO, USA

Radiotherapy-induced taste loss is a well-known side effect for head and neck cancer patients, but why it occurs is not understood. One possibility is that radiation targets proliferating epithelial cells located adjacent to taste buds; these cells are responsible for continual renewal of taste cells in adults. We examined the homeostatic kinetics of these taste progenitors, and found that 80% of basal cells in the circumvallate epithelium are actively cycling, 15% are in S phase, and 10% are in M phase. We then irradiated (8Gy) adult mice and assessed cell cycle kinetics of the progenitor population. Proliferating cells drop dramatically (<20%) at 1-3 days post-irradiation (dpi), but cycling cells return to control levels by 6 dpi. Cells in S and M phase also drop initially (5% and 4%), but then transiently overshoot control levels, indicating that during recovery from radiation injury, the cell cycle accelerates and/or the proliferative pool is synchronized as cycling resumes. To determine if shifts in kinetics affect taste cell renewal, we monitored entry of new cells into taste buds. Significantly fewer new cells were found in buds immediately post-irradiation, whereas entry of new cells exceeded control levels during recovery. Finally we examined the status of differentiated taste cells post-irradiation. Taste cell number did not differ from controls at 3dpi, but was reduced at 7 dpi. In sum, our data support a model where irradiation targets taste progenitor cells and transiently interupts delivery of new cells into taste buds; while older taste cells are lost via normal attrition, resulting in a delayed and transitory reduction in differentiated taste cells. We hypothesize that this mechanism may underlie radiotherapy-induced taste loss in patients. Acknowledgements: NIDCD DC03947 LAB, NIDCR DE015648 to MER

#P44 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Flavor Fusion Nullification as a Presentation of Hypoguesia

Alan R. Hirsch, Gurprit S. Bains, Alam Asiri
Smell & Taste Treatment & Research Foundation Chicago, IL, USA

Objectives: An unusual manifestation of hypoguesia is described. **Methods/Results: Case 1:** 56 year old women, one year prior to presentation noted a two week period of reduction of taste to the point of total absence, which has persisted since then, as confirmed on threshold and suprathreshold taste testing. She denied trouble smelling and normosmia was confirmed with the QSIT and the BSIT. With thiamine and folic acid she noted a return of taste of 10-15% of normal. In particular, when foods were presented individually, flavors were perceived as normal. However, when these same foods were mixed together, there was no perceived flavor. For instance, she was able to taste blue cheese or onions alone without difficulty, but when placed on hamburger there was no taste at all. Similarly celery sticks alone had a normal flavor, but when placed in a chicken salad no taste, including all components of the salad, was perceived. **Case 2:** 38 year old male with a 4 month progressive loss of ability to taste to 50% of normal. He denied trouble smelling and normosmia was confirmed by UPSIT and QSIT. He noted that individually he was able to delineate flavors, but not when in combinations. Individually he can taste green pepper, mushroom and mozzarella cheese but when combined in a pizza, he tastes nothing. Similarly he can taste cookie dough or chocolate chips alone but when baked into a chocolate chip cookie, there is no taste at all. He can taste sausage, bread, turkey, and cheese individually, but when mixed as a sandwich it is totally flavorless, like eating cardboard. **Conclusions:** Common culinary concoctions integrate myriad flavors. These combinations may act in a hypo-additive manner to inhibit perceptions of certain flavors. This effect may be intensified in those with hypoguesia. Acknowledgements: None

#P45 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Long-term olfactory, neurocognitive and morphological consequences of chemotherapy for childhood leukemia

Franziska Krone[1], Marina Genschaft[2], Thomas Hübner[3], Franziska Plessow[4], Vasiliki N. Ikonomidou[5], Nasreddin Abolmaali[5], Meinolf Suttorp[2], Chrysanthy Ikonomidou[7], Clemens Kirschbaum[4], Michael N. Smolka[3], Thomas Hummel[1]
[1]Interdisciplinary Center for Smell & Taste, Dept. of Otorhinolaryngology, University of Dresden Dresden, Germany, [2]Dept. of Pediatrics, University of Dresden Dresden, Germany,

[3]Section of Systems Neuroscience, Department of Psychiatry, University of Dresden Dresden, Germany, [4]Dept. of Psychology, University of Dresden Dresden, Germany, [5]Dept. of Electrical and Computer Engineering, The Volgenau School of Information Technology and Engineering, George Mason University Fairfax, VA, USA, [6]Institute of Radiology, University of Dresden Dresden, Germany, [7]Dept. of Neurology and Waisman Center, University of Wisconsin Madison, WI, USA

Acute lymphatic leukemia (ALL) is the most common cancer in childhood. Due to an intensive chemotherapy more than 80% of the children can be cured, but the treatment can cause a wide range of side effects. Aim of this study was to investigate the long-term consequences of chemotherapy for childhood ALL on olfactory and cognitive function as well as brain morphology. A total of 19 ALL-survivors and 19 age- and sex-matched healthy controls (age range 15-23 years; 8 men, 11 women) participated. All diseased subjects developed ALL without a central nervous involvement before their 10th birthday and had no current evidence of disease. They were treated according to the "BFM protocol" (n=14) or "Co-ALL protocol" (n=5) with multiple intrathecal injections of methotrexate. All participants performed detailed lateralized investigation of the olfactory function using the "Sniffin' Sticks" odor threshold and odor identification test. Furthermore subjects received a neuropsychological evaluation of hippocampus-dependent memory functions, executive function and attention. For the structural brain analysis a T1-weighted magnet resonance imaging (MRI) was used. Comparing ALL and control group no significant differences in olfactory function were found. In contrast, neuropsychological testing revealed significant impairments in verbal memory, the ability to shield current action goals against distraction and sustained attention as well as increased impulsivity with continuous cognitive demand in ALL subjects. MRI data analyses showed a significant reduction of the mean volumes of hippocampus and amygdale as well as the white matter density in the putamen. The results indicate that ALL chemotherapy induces structural and neurocognitive damage, however, no major impairment of the olfactory function was observed.

#P46 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Rescuing Flavor Perception in the Elderly

Jennifer J. Stamps, Linda M. Bartoshuk
University of Florida Center for Smell and Taste Gainesville, FL, USA

Malnutrition is relatively high in the elderly and is the leading cause of death in patients with Alzheimer's disease. Thus it is important to evaluate flavor perception in these groups to determine if dietary changes are influenced by olfactory, taste, and/or trigeminal dysfunction. In our previous study (N=46) of normal participants of various ages (Stamps and Bartoshuk, AChemS, 2010), we found that those with taste loss experienced a loss of retronasal olfaction (i.e., flavor) for some foods and not others and that foods with a strong trigeminal component were the least vulnerable. For this study we recruited an additional cohort including normal participants of various ages (N=46) as well as a small sample of Alzheimer's patients (N=10) to see if we could rescue flavor by adding a trigeminal component to a food previously shown to have lost flavor with the loss of taste function (grape jelly). Cayenne pepper was added to grape jelly by weight, doubling the amount for each increment. The burn of the resulting jelly samples increased significantly until the third increment which did not increase the burn further. As predicted, ANOVA showed that the retronasal perception of the grape flavor increased as the burn increased; orthonasal olfaction did not change and the taste of the jelly did not change. This is consistent with our suggestion that flavor loss produced by taste damage may be rescued with trigeminal stimulation. Interestingly, the older participants (including the Alzheimer's patients) had significantly fewer fungiform papillae than the young participants suggesting a source of taste damage in this sample that deserves further study. Finding a way to overcome flavor loss due to a loss of taste function has important therapeutic implications. Acknowledgements: We thank NIDCD for support via DC283 and the NCRR for support to the Clinical and Translational Science Institute at the University of Florida (award TL1RR029888)

#P47 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Unlike Roux-en-Y Gastric Bypass Ileal Interposition Does Not Alter Sweet Taste Preference in High Fat Diet-Induced Obese Rats

Andras Hajnal[1,2], Mingjie Sun[1,2], Nikhil K. Acharya[1], Benjamin Bauchwitz[1], Ann M. Rogers[2]
[1]Neural & Behavioral Sciences, Penn State Univ. Coll. Med. Hershey, PA, USA, [2]Surgery, Penn State Milton S. Hershey Medical Center Hershey, PA, USA

Recently we have demonstrated effects of Roux-en-Y gastric bypass surgery (GBS) to reduce sweet taste preference and improve sucrose-elicited central gustatory responses in obese rats (Hajnal et al. AJP 2010). We hypothesized the effects of GBS on taste were related to improved postprandial GI hormone signaling. Whereas both GBS and ileal interposition (IT) result in increased glucagon-like peptide 1 (GLP-1) and peptide YY (PYY) signaling and improved blood glucose control, only GBS results in sustained weight loss. To investigate plausible effects of IT on taste and preference functions for sucrose, we performed brief-access (10-s) lickometer and 24-hr 2-bottle tests in high fat diet-induced obese (DIO, 18 weeks on 60 kcal% fat, high-energy diet) male Sprague Dawley rats (n=9). An additional group of DIO rats underwent GBS (n=5). The surgical controls received enterotomies without reducing and bypassing the stomach (n=9) or interposing the ileal segment (n=9). Whereas GBS in DIO rats resulted in significant (-27% of pre-op. baseline) weight loss, IT and sham surgeries caused an identical non-significant reduction in body weight (-10%). Results of the taste tests performed 3-6 weeks after the surgeries showed that unlike GBS, that reduced long-, and short-term sucrose preferences for higher concentrations of sucrose (>0.3M), IT surgeries had no effect on the same measures when the surgical group was compared with sham operated obese controls. These findings suggest that factors other than improved GI hormone release may be required for taste changes and body weight loss observed following GBS in this rat model. Further studies aiming to elucidate underlying mechanism may help identify novel pharmacological targets to reduce overeating and sweet cravings. Acknowledgements: Supported by NIH grant DK080899 to A.H.

#P48 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Habituation to the Pleasure Elicited by Sweetness in Lean and Obese Women

M. Yanina Pepino[1], Susana Finkbeiner[2], Julie A. Mennella[2]
[1]Washington University in St Louis St Louis, MO, USA, [2]Monell Chemical Senses Center Philadelphia, PA, USA

The current study bridges the gap between two theoretical approaches to the study of satiation processes: habituation and sensory specific satiety by examining whether hedonic responses to repetitive stimulation to an unswallowed sweet tasting solution follow habituation patterns. In addition, we examined whether these patterns differed between obese and lean women and verified that differences in hedonic responses were not due to adaptation to sweetness intensity. Twenty two obese and 32 lean women participated in the sensory study. A 24% v/v sucrose solution was repetitively presented every 2 minutes for 10 consecutive trials. Perceived hedonic value and sweet intensity were measured by using the general Magnitude Label Scale. Obese women respond with a slower rate of habituation to the pleasantness derived from savoring the sweet stimulus and reported they would feel more pleasure from eating something sweet than lean women. Noteworthy, obese and lean woman did not differ on their perception of sweetness intensity, which was kept relatively constant across trials. Therefore, 1) the decreased appetitive response to the sweet taste observed in lean women was not explained by adaptation processes at the or sensory level, and 2) the difference observed between obese and lean women was not due to differential perception of taste intensity or scale bias between the groups. In conclusion, obesity is associated with slower patterns of habituation and increased rewarding value for sweet tastes in women. These characteristics could contribute to slower satiation rates, prolongation of eating episodes and excessive sweet food consumption among the obese.
Acknowledgements: This project was funded, in part, by a grant from the Pennsylvania Department of Health. The Pennsylvania Department of Health specifically disclaims responsibility for any analyses, interpretations, or conclusions. Dr. Pepino is currently a KL2 fellow at Washington University in St. Louis (KL2 CTSA: KL2RR024994 and CTSA UL1RR024992).

#P49 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Is There a Link Between Otitis Media, Liking for Fat/Sugar Foods, and Obesity Among At-Risk Preschoolers?

Mastaneh Sharafi, Heather L. Harrington, Valerie B. Duffy
University of Connecticut Storrs, CT, USA

Chronic exposure to otitis media (OM) may influence food preference and obesity risk via altered oral sensations. We have shown that preschoolers with high OM exposure have lower fruit/vegetable preference and greater obesity risk than those with low exposure. Data from adults suggest that OM exposure associates with elevated obesity risk via an affinity for sweet and fatty foods. Here, we tested if the OM-fat/sweet preference relationship exists among racially-diverse, low-income preschoolers (231 boys, 253 girls; 4% underweight, 17% overweight, 13% obese). Reported OM exposure varied across nearly equal quartiles—low exposure (never, once) to high exposure (3-5 times, 6+ times) categories. Parents rated their children's liking of foods (high fat/added sugar, fruits/juice, vegetables) and pleasurable activities (bathing, dressing, brushing teeth). The low and high OM categories failed to differ significantly in average fat/sugar food liking. Yet, if ranking from most to least liked, fruits/juice achieved higher liking ranks for the low OM children, while fat/sugar foods achieved these ranks in the high OM children. The fat/sugar foods were ranked two-fold higher than the pleasurable activities among high OM children, but equally ranked among low OM children. These effects were most pronounced in boys, who were more likely to be in the high OM categories. Vegetables were ranked least liked for all children, yet the magnitude of dislike was greatest for the high OM children. Summary—Our findings were consistent with adult studies associating liking for fat/sugar foods with high OM exposure. We continue to support dietary preference as a mediator for the relation between OM exposure and obesity risk.
Acknowledgements: American Diabetes Association Foundation

#P50 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Deficits in encoding valence and intensity in Alzheimer's disease

Pauline Joussain, Catherine Rouby, Floriane Delphin, Anne Didier, Pierre Krolak-Salmon, Moustafa Bensafi
Centre de Neurosciences Lyon, France

Studies of olfaction in Alzheimer's disease (AD) mainly pointed deficits in detection and identification of odors (Djordjevic et al., 2008). As regards intensity and pleasantness judgments, one study concluded that Alzheimer patients did not differ from controls (Royet et al., 2001). Further studies however demonstrated that intensity of pleasant and unpleasant odors is integrated in the amygdala (Anderson et al., 2003; Winston et al. 2005). Moreover, atrophy of the amygdala has been demonstrated in AD (Horinek et al., 2007). The present study was thus designed to test the hypothesis that AD patients would exhibit a deficit in processing intensity of pleasant and unpleasant odors. To this end, 7 patients (mean age 84, 5 females, MMS score between 17 and 23) and their matched controls (MMS between 27 and 30) were tested. Participants were screened for odor detection and odor identification abilities (Thomas-Danguin et al, 2003) and were asked to rate intensity, pleasantness and edibility of 20 monomolecular odorants. Whereas no significant difference were observed in overall intensity and pleasantness ratings ($p>0.05$), a polynomial regression between those ratings revealed a quadratic relation for controls ($p<.001$) (perceived intensity was greater for pleasant and unpleasant odors compared to neutral odors) but not for AD patients ($p>0.05$). Moreover, calculating the Euclidian distances between all 20 odorants in the intensity/pleasantness perceptual space of both groups showed a significant reduction of these distances in the AD group (vs. the control group, $p<.0001$). These results suggest that the integration between intensity and hedonics, which was found at all ages and in many studies, is impaired in AD. Such deficit in these basic categorizations may impair discrimination between odors in AD.
Acknowledgements: CNRS and Région Rhône Alpes

#P51 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Chemosensory Changes in Olfactory Dysfunction Patients

Ling Yang[1], Yongxiang Wei[2], Wei Zhang[3], Di Yu[1], Jinfeng Zhang[3], Kunyan Li[2]
[1]The Center Lab of Beijing Tongren Hospital, Capital Medical University Beijing, China, [2]The Department of Otolaryngology-Head & Neck Surgery, Beijing Chaoyang Hospital, Capital Medical University Beijing, China, [3]The Department of Otolaryngology-Head & Neck Surgery, Beijing Tongren Hospital, Capital Medical University Beijing, China

Objective: To investigate the changes of chemosensory function in olfactory disorder patients. **Methods:** A total of 238 subjects participated in the study which were divided into three groups, healthy group, functional anosmia group and hyposmia group. T&T, olfactory event-related potentials (oERPs), trigeminal ERPs and triple drop method were used to examine the chemosensory function. **Results:** There were significantly different between healthy group and hyposmia/functional anosmia group in T&T. Measurement of oERPs revealed that functional anosmia patients had longer latencies and lower amplitudes of N1/P2 waves compared with healthy subjects. To compare with healthy subjects, trigeminal ERPs test showed that functional anosmia patients had longer latencies of N1/P2 waves. The results of gustatory examination showed that there was significant difference between functional anosmia group and healthy group. **Conclusions:** The present data indicate that olfactory dysfunction is associated with decreased trigeminal function. Moreove, functional anosmia is in relation to decreased taste function. Acknowledgements: China Natural Scientific Foundation under project No. 30973284, Beijing Natural Scientific Foundation under project No. 7102063, Capital Medical Development Foundation under project No. 2007-1034.

#P52 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

ERP Tasks that Combine Olfactory Function with Semantic Processing Best Classify Those At Risk for Alzheimer's Disease

Charlie D. Morgan[1], Joel Kowalewski[1], Jessica Bartholow[1], Claire Murphy[1,2]
[1]San Diego State University/Department of Psychology San Diego, CA, USA, [2]University of California Medical Center San Diego, CA, USA

Event-related potentials (ERPs) have proven useful in the study of Alzheimer's disease (AD) and those at risk for Alzheimer's disease. Various methods of eliciting ERPs and their utility in differentiating those at risk for AD were explored in a series of studies. Participants were healthy older adults ages 65 and over, all free of cognitive impairment as measured by the Dementia Rating Scale. All participants were screened for nasal health and genotyped for the presence of the ApoE e4 allele, a genetic risk factor for development of Alzheimer's disease. A number of methods of eliciting ERPs were utilized including an active olfactory task, a passive olfactory task, an odor identification task, and a semantic olfactory congruency task. The active and passive tasks included repeated presentation of one odor with either active responding or no responding to the stimulus. The odor identification task consisted of 14 odors presented and then identified from a multiple choice list. The semantic congruency task paired odor stimuli with visual pictures of odors, some trials where the odor was congruent with the picture (e.g. rose odor paired with rose picture) and others noncongruent (chocolate odor paired with popcorn picture). Binary logistic regression demonstrated good ApoE status classification rates for all tasks, with excellent classification rates for tasks that involved semantic processing. These findings suggest that combining olfactory ERP tasks with tasks involving semantic processing may help us to better understand the dementing processes associated with Alzheimer's disease. Acknowledgements: Supported by NIH Grant DC02064 to Claire Murphy. The authors would like to acknowledge the late Dr. Leon Thal and the ADRC for genotyping (P50AG05131), Krystin Corby, Roberto Zamora, and Richard Vail for their contributions.

#P53 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Olfactory Learning Deficits Are Associated With and May Precede Age-related Memory Loss

George Edwards[1], Karienn Montgomery[1], Cristina Banuelos[1], Sofia Beas[1], Barry Setlow[2], Jennifer Bizon[1]
[1]Dept. of Neuroscience, University of Florida Gainesville, FL, USA, [2]Dept. of Psychiatry, University of Florida Gainesville, FL, USA

Rodent models of cognitive aging routinely use spatial performance on the Morris water maze to characterize medial temporal lobe integrity. Water maze performance is dependent upon this system and, as in aged humans, individual differences in learning abilities are reliably observed among spatially-characterized aged rats. However, unlike human aging in which learning deficits rarely occur in isolation, few non-spatial learning deficits have been identified in association with spatial impairments among aged rats. Using a simultaneous two-choice discrimination task, we found that the same subset of male aged Fischer 344 rats was impaired both in the water maze and in their ability to discriminate between odors. These deficits were not due to anosmia and were specific to olfactory discrimination learning, as aged-cognitively impaired rats performed normally on an odor detection test and an analogous non-olfactory discrimination task, respectively. A second experiment determined the extent to which performance on the odor discrimination task could be improved by cognitive-enhancing drugs. Acute administration of either the cholinesterase inhibitor donepezil or the GABA(B) antagonist CGP55845 produced a significant improvement in odor discrimination learning in cognitively-impaired aged rats. Finally, middle-aged rats were assessed on both the water maze and olfactory discrimination learning tasks. While few individual differences in memory ability were observed among middle-aged subjects on the water maze, a wide range of individual differences was observed on the olfactory learning assessment. In combination, these data help to establish a rodent model in which age-related cognitive impairments can be predicted, and pharmacological and potential treatments for memory loss in aging investigated. Acknowledgements: R01AG029421 (to Jennifer Bizon)

#P54 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Olfactory Impairment and the 10-yr Incidence of Cognitive Impairment

Karen J Cruickshanks[1,2], Carla R Schubert[2], David M Nondahl[2], Ronald Klein[1,2], Barbara EK Klein[1,2], Rick Chappell[3]
[1]University of Wisconsin Department of Population Health Sciences Madison, WI, USA, [2]University of Wisconsin Department of Ophthalmology and Visual Sciences Madison, WI, USA, [3]University of Wisconsin Department of Biostatistics and Medical Informatics Madison, WI, USA

Olfactory impairment has been associated with the short-term risk of developing cognitive impairment but the long-term risk is unknown. In 1998-2000, olfactory impairment was measured with the San Diego Odor Identification Test (SDOIT) in the population-based longitudinal Epidemiology of Hearing Loss Study (EHLS). The Mini-Mental State Examination (MMSE) was obtained every five years beginning in 1998-2000. During the 2009-10 visit additional measures of cognitive function (Trail Making Test (TMTA and TMTB) and the Auditory Verbal Learning Test (AVLT)) were obtained. Cognitive impairment was defined as a MMSE score <24 or a proxy report of diagnosed dementia. Cox Proportional Hazard Models were used to assess associations with cumulative incidence. There were 2053 people with SDOIT results and MMSE≥24 in 1998-2000 (at risk for cognitive impairment). Olfactory impairment (SDOIT<6) was associated with the ten-year cumulative incidence of cognitive impairment (Hazard Ratio (HR)=3.18, 95% Confidence Interval (CI)=2.26,4.48), adjusting for age, sex, and education. Including additional covariates (occupation, smoking, and nasal polyps) did not alter this association (HR=3.32, 95%CI=2.33,4.75). The sensitivity was 46%, specificity 85%, positive predictive value (PPV) 24%, and negative predictive value 94%. Impaired olfaction also was associated with slower performance ten years later on the TMTA (9.8 sec longer, p<0.001) and TMTB (19 sec longer, p<0.001), but not with the AVLT (p=.25). These results suggest that olfactory impairment is an indicator of higher risk of cognitive impairment and dysfunction ten years later, which may reflect either shared risk factors or early brain changes, but the low PPV limits its usefulness as a screening tool.
Acknowledgements: R37AG11099 and U10EY06594

#P55 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Effects of olfactory training in patients with Parkinson's disease

Antje Haehner[1,2], Clara Tosch[1], Martin Wolz[2], Lisa Klingelhöfer[2], Christine Schneider[2], Thomas Hummel[1]
[1]University of Dresden Medical School, Dept. of Otorhinolaryngology Dresden, Germany, [2]University of Dresden Medical School, Dept. of Neurology Dresden, Germany

Decrease of olfactory function in patients with Parkinson's disease (PD) is a well-investigated fact. Studies indicate that pharmacological treatment of PD fails to restore olfactory function in PD patients. The aim of this investigation was whether patients with PD would benefit from ''training'' with odors in terms of an improvement of their general olfactory function. It was hypothesized that olfactory training should produce both an improved sensitivity towards the odors used in the training process and an overall increase of olfactory function. Methods: One group of PD patients with olfactory loss performed the training, whereas another group did not. Olfactory training was performed over a period of 12 weeks. Patients exposed themselves twice daily to four odors (phenyl ethyl alcohol: rose, eucalyptol: eucalyptus, citronellal: lemon, and eugenol: cloves). Olfactory testing was performed before and after training using the "Sniffin' Sticks'' (thresholds for phenyl ethyl alcohol, tests for odor discrimination, and odor identification) in addition to threshold tests for the odors used in the training process. Results: Compared to baseline, training PD patients experienced an increase in their olfactory function, which was observed for the Sniffin' Sticks test score and for thresholds for the odors used in the training process. In contrast, olfactory function was unchanged in PD patients who did not perform olfactory training. Conclusions: The present results indicate that olfactory training may increase olfactory sensitivity in PD patients.

#P56 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Identification of odors in patients with Parkinson's disease compared to patients with post-viral or post-traumatic smell disorders

Wakunyambo Maboshe[1], Thomas Hummel[2], Susann Bietenbeck[3], Birgit Herting[3], Alexander Storch[3], Heinz Reichmann[3], Antje Hähner[2,3]
[1]Cardiff School of Biosciences, Cardiff University Cardiff, Wales, [2]Smell & Taste Clinic, Dept. of Otorhinolaryngology, University of Dresden Medical School Dresden, Germany, [3]Dept. of Neurology, Technical University of Dresden Medical School Dresden, Germany

Background and Objective: Olfactory dysfunction as an early cardinal symptom of Parkinson's disease (PD) is observed in more than 90% of patients and can be helpful as a diagnostic tool. Previous observations showed that certain smells showed a particularly high sensitivity and specificity in these patients (Hawkes et al. 1993 or Daum et al. 2000). The present study examined whether odor identification in PD patients differs from that in patients with post-viral or post-traumatic olfactory loss. **Methods:** The "Sniffin' Sticks" test was applied on 20 consecutive PD patients with hyposmia or functional anosmia. The answers from PD patients to the 16 fragrances of the identification tests were to be "right" or "false" documents and, together with the results of 20 randomly selected gender and age matched patients with post-traumatic or post-viral olfactory disorder were used to compare olfactory disorder. **Results:** The majority of PD patients identified fragrances such as "apple" and "cinnamon" as incorrect whilst odors such as "Orange "and" garlic" were correctly identified. These results did not differ significantly from the error rates of patients with post-viral or post-traumatic olfactory disorder (Chi-square: p> 0.23). **Conclusion:** Odor identification in patients with PD, post-viral, or post-traumatic olfactory disorder is not significantly different. Rather, patients with olfactory dysfunction appear regardless of the cause, to multi-select a certain predetermined arrangement of fragrances and identify them with particular difficulty or impossibility.

We are currently expanding this study further by applying an extended Identification test to non-Parkinsonian hyposmic patients in order to compare the error rates of fragrances in these controls to our previously outlined findings in PD patients.

#P57 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Smell and Taste Function in Children with Cystic Fibrosis

Jessica E Armstrong[1,2], David G Laing[1,2], Maggie Aitken[2], Alistair Carrol[2], Fiona J Wilkes[3], Anthony L Jinks[3], Adam Jaffe[1,2]
[1]University of New South Wales Sydney, Australia, [2]Sydney Children's Hospital Sydney, Australia, [3]University of Western Sydney, School of Psychology Sydney, Australia

A major problem for patients with cystic fibrosis (CF) is the maintenance of adequate nutrition to maintain normal growth. The hypothesis that poor nutrition could be due to smell and/or taste dysfunction has been pursued in several studies with contradictory results. Accordingly, in an effort to determine the effects of chemosensory changes in CF patients, the present study investigated the relationship between BMI, FEV1 and smell and taste function in 42 CF and 42 healthy 5-18 yr olds. A three-choice 16-item odour identification test and a gustatory identification test involving five concentrations of sweet, sour, bitter and salty tastes were used to assess chemosensory function. Patients identified significantly fewer odorants than controls (89.8 vs 95.7% correct; $p < 0.001$). However, only a few patients were affected and their loss of olfactory function was not substantial and unlikely to affect their liking for foods. Taste identification was similar for the two groups (patients 92.6 vs controls 94.2% correct). There was no correlation between age and odour identification ability, but taste performance improved with age ($r = 0.39$, $p < 0.05$) suggesting cognition was the cause. No significant relationships existed between FEV1 and smell or taste function, the BMI of the groups were similar and there was no relationship between BMI and smell and taste function. The results indicate that the abnormal eating behaviour reported for many CF patients is not due to changes in chemosensory function which remains normal in most CF patients at least to 18 years of age. Acknowledgements: This study was partly funded by a grant from the Sydney Children's Hospital Foundation and J.E.A was supported by an Australian Postgraduate Award.

#P58 **POSTER SESSION I: TASTE PSYCHOPHYSICS; GUSTATORY RECEPTORS; CHEMOSENSATION AND DISEASE**

Comparison of pharyngeal chemosensitivity between patients with obstructive sleep apnea and healthy subjects

Clemens Heiser, Ingo Zimmermann, Karl Hörmann, J. Ulrich Sommer, Boris A. Stuck
Department of Otorhinolaryngology, Head and Neck Surgery, University Hospital Mannheim Mannheim, Germany

Signs of pharyngeal neurodegeneration have been detected in patients with obstructive sleep apnea (OSA). This degeneration is believed to be due to excessive soft tissue vibration which is typically associated with snoring and apneas. Along with this neurodegeneration, a decreased pharyngeal sensitivity has been described in OSA patients compared to healthy subjects. The decreased sensibility may play a role in the physiology and progression of this disease. Aim of the study was to investigate the chemosensitivity of the pharyngeal mucosa of OSA-patients compared to healthy controls. Healthy controls (c) and patients with OSA (age: 30 to 60 years) were included. Testing of oropharyngeal chemosensitivity was performed with subjective intensity rating (SIR, Visual Scale 0-10) of capsaicin, air puffs (6 l/min, presented with an olfactometer), and stimulation with CO_2 (60%) at the posterior pharyngeal wall. In addittion, a two point discrimination test at the soft palate, an intensity rating of capsaicin at the tongue, and a nasal lateralization test were performed. 26 patients with OSA and 18 healthy controls were included. No differences in the SIR of capsaicin on the tongue, in the nasal lateralization and taste strips were detected. The results demonstrated a decreased pharyngeal sensitivity to capsaicin (OSA: 6.8±2.3; c: 8.6±1.3) air puffs (OSA: 2.2±1.5; c: 4.2±1.8) and stimulation with CO_2 OSA: 4.3±3.1 c: 7.0±2.6) in patients with OSA. Two point discrimination at soft palate was reduced in the OSA group (OSA: 11.5±5mm; c: 4.7±2,4mm). The results demonstrate a reduced pharyngeal chemosensitivity in OSA-patients in addition to the reduced sensitivity of the soft palate. This underlines the hypothesis of a peripheral neurodegeneration in the context of this disease.

#P59 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

New Natural and Synthetic Perillaketone Derivatives: Isolation, Synthesis and *in vitro* Activity on the Somatosensory TRPA1 Receptor

Angela Bassoli[1], Gigliola Borgonovo[1], Gabriella Morini[2], Vincenzo Di Marzo[3], Luciano De Petrocellis[4]
[1]DISMA-University of Milano Milano, Italy, [2]University of Gastronomic Sciences Pollenzo (CN), Italy, [3]Endocannabinoid Research Group, Institute of Biomolecular Chemistry, CNR Pozzuoli (NA), Italy, [4]Endocannabinoid Research Group, Institute of Cybernetics, "Eduardo Caianiello", CNR Pozzuoli (NA), Italy

Perilla frutescens is a food plant widely diffused in all Asia. Different cultivars are characterised by compounds with a peculiar taste and chemestetic profile. Some of them, namely perillaldehyde and perillaketone, have been also previously demonstrated by us to be strongly active *in vitro* on TRPA1 receptor.[1] Some TRPA1-activating compounds are electrophiles able to undergo a Michael addition with nucleophilic thiol residues of cysteine on the receptor. Many of the known agonists of TRPA1 contain in fact electrophilic groups such as the isothiocyanates and unsaturated aldehydes as acrolein or cinnamaldehyde. Some of these compounds exhibit irritating or even noxious properties at high concentrations; interestingly, also perillaketone has some toxicity at pulmonary level in some animals, and its safety for human consumption is under discussion. We synthesised 17 derivatives of perillaketone by modifying the structure and position of alkyl chain on furane ring and/or adding substituted aryl groups; the compounds have been purified and submitted to *in vitro* tests with TRPA1 receptor. Most of them (11 over 17) are strongly active, with a potency in some cases higher than that of the natural lead. From the essential oils of some cultivars of *Perilla frutescens* experimentally grown in Italy we also isolated isoegomaketone, an unsaturated

perillaketone derivative. Also this natural compound proved to be specifically active on TRPA1. Therefore perillaketone and its natural and synthetic analogues are a very interesting new family of agonists for the TRPA1 ion channel and can be used to derive useful structure-activity relationship. [1] A. Bassoli, G. Borgonovo, S. Caimi, L. Scaglioni, G. Morini, A. Schiano Moriello, V. Di Marzo, L. De Petrocellis, J. Biorg. & Med. Chem., 2009, 17, 1636–1639. Acknowledgements: Ministry of Foreign Affair (MAE) Italy, Great Relevance project Italy-Korea 2010 "Bioactive compounds for the valorisation and promotion of traditional food" University of Milano University of Gastronomic Sciences.

#P60 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

A Common Mechanism for the Pungent Sensations of CO_2 and Weak Acids

Yuanyuan Y. Wang, Rui B. Chang, Emily R. Liman
Department of Biological Sciences, Section of Neurobiology, University of Southern California Los Angeles, CA, USA

Inhaled carbon dioxide (CO_2) and weak acids, such as acetic acid, elicit a variety of sensory responses in vertebrates including a painful sensation. The painful sensation has been attributed to the activation of nociceptors with cell bodies in the trigeminal ganglion that project to the nasal or oral cavities. The specific cell types that respond to these stimuli and the underlying mechanism of transduction are, however, unknown. Here we show that nociceptors that express the cinnamaldehyde-activated ion channel TRPA1 are specifically responsive to CO_2 and weak acids, and that a functional TRPA1 gene is required for these responses. CO_2 and weak acids strongly activate TRPA1 currents, as a consequence of acidification of the cell cytosol and direct gating of the channel by intracellular protons. Together our data show that TRPA1 is a general sensor for CO_2 and various weak acids that produce intracellular acidification and suggest that it functions within the pain pathway to mediate sensitivity to cellular acidosis. Acknowledgements: DC004564

#P61 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

TRPM8 and TRPA1 mediate the key somatosensory qualities of cooling agents

Tetsuya Dohara[1], Yuichi Furudono[1,2], Kentaro Takasaki[1], Hisanori Nagata[1], Takashi Inoue[1]
[1]Tobacco Science Research Center, Japan Tobacco INC. Yokohama, Japan, [2]Monell Chemical Senses Center Philadelphia, PA, USA

Menthol can elicit a thermal and painful sensation when topically applied to the skin or mucous membranes of the upper airways. Previous studies have shown that menthol activates at least two TRP channels, TRPM8 and TRPA1. TRPM8 is activated by cooling and chemical stimuli like icilin and TRPA1 is activated by pungent compounds such as mustard oil. Thus sensory qualities of menthol could be mediated by these two ion channels. Although cooling agents having a chemical structure based on or similar to Menthol are known to be TRPM8 agonist, little is known if they could activate TRPA1 and other somatosensory systems. The present study examined the effects of cooling agents structurally related menthol on TRPM8 and TRPA1, in addition to their sensory properties. In experiment 1, we observed the sensitivity of hTRPM8 or hTRPA1 transfected HEK293 cells to 9 cooling agents (*l*-Menthol, Isopulegol, Coolact1, Coolact5, Coolact10, Coolact38, WS3, WS23 and Frescolat-MGA) by measuring intracellular calcium. At the highest concentration tested, $[Ca^{2+}]_i$ increase induced by Isopulegol and Coolact38 were relatively small in comparison to other cooling agents in hTRPM8-HEK293 cells, while Menthol, Isopulegol and Coolact5 caused a larger $[Ca^{2+}]_i$ increase relative to other cooling agents in hTRPA1-HEK293 cells. In experiment 2, we presented aerosolized cooling agents to upper airways and asked 6 human subjects to rate the intensities of cooling and painful sensation on the labeled magnitude scale. Isopulegol and Coolact38 produced less than "moderate" intensity in cooling sensation, while Menthol, Isopulegol, Coolact5 and WS23 induced more than "moderate" intensity in painful sensation. These experiments illustrate that there was high relationship between TRPM8-/TRPA1-mediated [Ca2+]i increases and sensory qualities.

#P62 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Genetic dissection of nociception in *Drosophila melanogaster*

Madison L Shoaf, Wayne L Silver, Erik C Johnson
Wake Forest University Winston Salem, NC, USA

The detection of harmful chemical irritants is important for the avoidance of potential life threatening compounds. In vertebrates, the trigeminal nerve is an important anatomical site of chemical nociception, and the nerve directly responds to a variety of chemical compounds. A molecular target for many of these trigeminal stimulants is the TRPA1 channel. *Drosophila melanogaster* possess multiple homologs of mammalian TRPA1 channels, two of which are encoded by the *painless* and *dTRPA1* genes. Both of these channels are reported to be required for behavioral aversion to allyl isothiocyanate (AITC). Consequently, it is unclear whether these channels are acting independently or in combination. These channels are expressed in gustatory receptor neurons in taste sensilla. We are evaluating the expression patterns of *painless* and *dTRPA1* in the *Drosophila* central nervous system to determine if the two channels are colocalized in the same cells. Gene drivers were used to introduce GFP into cells expressing either *painless* or *dTRPA1*. Laser scanning confocal microscopy was utilized to image dissected brains from third instar larvae and adults and fluorescent cell numbers were counted. We are also determining if *painless* and *dTRPA1* are required for aversion to other known trigeminal irritants using a two-choice food preference assay. Our results will provide further insight into the specific role each of these channels play in chemical nociception.

#P63 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Fetal ethanol exposure attenuates the aversive oral effects of TrpV1 but not TrpA1 agonists

John I Glendinning[1], Yael Simons[1], Lisa Youngentob[2,3], Steve L Youngentob[2,3]
[1]Barnard College/Biological Sciences New York, NY, USA, [2]SUNY Upstate Medical University/Neuroscience and Physiology Syracuse, NY, USA, [3]SUNY Developmental Exposure Alcohol Research Center Syracuse & Binghamton, NY, USA

In humans, fetal ethanol exposure is highly predictive of adolescent ethanol use and abuse. Prior work in our labs indicated that fetal ethanol exposure results in stimulus-induced chemosensory plasticity in the taste and olfactory systems of adolescent (P30) rats. In particular, we found that increased ethanol acceptability could be attributed, in part, to an attenuated aversion to ethanol's quinine-like taste quality. Here, we asked whether fetal ethanol exposure also alters the oral trigeminal response of adolescent rats to ethanol. We focused on two excitatory ligand-gated ion channels, TrpV1 and TrpA1, which are expressed in oral trigeminal neurons and mediate the aversive orosensory response to many chemical irritants. To target TrpV1, we used capsaicin; to target TrpA1, we used allyl isothiocyanate (AITC). We assessed the aversive oral effects of ethanol, capsaicin and AITC by measuring short-term licking responses to a range of concentrations of each chemical. Experimental rats were exposed to ethanol *in utero* via the dam's diet, whereas control rats were given chow ad libitum. We found that fetal ethanol exposure attenuated the oral aversiveness of ethanol and capsaicin, but not AITC. Moreover, the reduced aversiveness of ethanol was directly related to reduced aversiveness of the TrpV1-mediated orosensory input. We propose that fetal ethanol exposure not only makes ethanol smell and taste better, but it also attenuates ethanol's capsaicin-like burning sensations. Acknowledgements: This work was supported by NIH-NIAAA Grants AA014871 and AA017823

#P64 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Soy-derived glycopeptides induce inward current in TRPV1-expressing cells by whole-cell patch-clamp recording

Mee-Ra Rhyu[1], Bo Hyun Lee[1], Yong Ho Kim[2], Ah Young Song[1], Hee Jin Son[1], Seog Bae Oh[2], Vijay Lyall[3], Eun Young Kim[1]
[1]Functional Food Technology Research Group, Korea Food Research Institute Seongnam-Si, South Korea, [2]Department of Physiology, School of Dentistry, Seoul National University Seoul, South Korea, [3]Physiology, Virginia Commonwealth University Richmond, VA, USA

In previous presentations we have shown that naturally occurring glycopeptides (FII) derived from a mature Korean soy sauce modulate salt taste on human, mouse behavior and amiloride-insensitive NaCl chorda tympani taste nerve responses by interacting with TRPV1 variant salt taste receptor (TRPV1t). In this presentation, we performed whole-cell patch-clamp recordings in TRPV1 expressing cells to test the interaction with the peptides and TRPV1 receptor. FII was further separated into LHa, LHb, LHc, LHd, LHe1, LHe2, and LHe3 by Sephadex 20 column chromatography and tested on TRPV1-transfected Human Embryonic Kidney (HEK293) cells by whole-cell patch clamp recording in the extracellular Ca^{2+}-free condition at -60 mV. 0.2% LHd induced inward currents around 20 pA in TRPV1 expressed cells, 0.2% LHe1 evoked 260 pA but others did not. LHe1 (from 0.00002% to 0.2%) caused inward currents in a dose-dependent manner (EC_{50} = 0.0027%). The characteristics of receptor which responded by LHe1, current-voltage (I-V) relationship was tested from -120 mV to 80 mV. Capsazepine (10 M), a specific antagonist of TRPV1, completely eliminated LHe1-evoked inward currents. These results indicated that LHe1 has agonistic activities and a site specific response to TRPV1. These data further support to the suggestion that the food-derived glycopeptides interact with the TRPV1 variant cation channel in taste receptor cells. Acknowledgements: Supported by a Korea Food Research Institute (KFRI) grant E0105101.

#P65 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Activation of the trigeminal system by odorous substances

Matthias Luebbert[1,2], Jessica Kyereme[1], Markus Rothermel[3], Klaus Peter Hoffmann[1], Hanns Hatt[1]
[1]Faculty of Biology and Biotechnology, Ruhr University Bochum Bochum, Germany, [2]Ruhr University Research School Bochum, Germany, [3]The Brain Institute, University of Utah Salt Lake City, UT, USA

Mammalian chemosensation is predominantly mediated via the olfactory and the trigeminal system (TS). Both systems are linked to each other and nearly all olfactory stimuli induce trigeminal sensations upon passing a certain concentration. The molecular basics of this phenomenon are only rudimentarily investigated and understood. In order to identify some of the molecular players underlying this activation, we performed whole cell voltage-clamp recordings using neurons from dissociated trigeminal ganglia (TG) of rats (P2-P5). These were stimulated with different odorous substances. We could identify two different populations of neurons. In the first population, application of helional led to an increase in membrane conductance, whereas the same substance inhibited background-currents within the second population. To transfer our findings from an *in vitro* to an *in vivo* situation, we used voltage sensitive dyes to monitor neuronal activity at the level of the TG in anesthetized adult male whistar rats. We observed a diffuse pattern of neuronal activity spreading over the whole TG upon stimulating the animal with helional and the structurally related odorant vanillin. Members of the TRP-family might be potentially targeted by the tested substances. Therefore, we performed voltage-clamp recordings using CHO cells heterologously expressing rTRPV1 to directly test the effect of the used odorants on rTRPV1. At room temperature, application of helional, vanillin, and heliotropylacetone elicited capsazepine sensitive currents with a strong outward rectification, whose amplitudes increased upon elevating the bath temperature. Here, we identified several so far unknown modulators of the TRPV1-channel which might play an important role during the perception of odorants via the trigeminal system.

#P66 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

TRPV3 agonists induce a temporally desensitizing pattern of oral irritation and affect lingual temperature sensitivity

Amanda H Klein, Mirela Iodi Carstens, Earl Carstens
University of California Davis, Neurobiology, Physiology & Behavior Davis, CA, USA

Eugenol and carvacrol are oils extracted from the spices clove and oregano, respectively. Both are agonists of TRPV3 which is implicated in transduction of warmth and possibly heat pain. We presently investigated the temporal dynamics of lingual irritation elicited by eugenol and carvacrol, and their effects on thermosensitivity, using a half-tongue method in human subjects. Either eugenol (600 mM) or carvacrol (50 mM) was applied unilaterally, with vehicle applied to the other side. Eugenol elicited irritation that decreased significantly across repeated applications delivered at 1-min interstimulus intervals (ANOVA, $p<0.05$, n=17). Similar results were obtained with 50 mM carvacrol. After a 10 min rest period, the same agent was applied bilaterally. For eugenol and carvacrol, a significant proportion of subjects chose the vehicle-treated side to have stronger irritation (binomial test, $p<0.05$), and assigned significantly higher intensity ratings (paired t-test, $p<0.05$), indicating self-desensitization. We also investigated the effects of these agonists on heat pain. One side of the tongue received either eugenol or carvacrol and the other vehicle. Zero, 0.5, 5 and 10 min later, a 49°C heat stimulus was applied to the tongue bilaterally. Significant proportions of subjects chose the eugenol- or carvacrol-treated side as more painful, and assigned significantly higher intensity ratings to that side, after 0 and 1.5 min, but not at later intervals, indicating a brief heat hyperalgesia ($p<0.05$, n=30). TRPV3 agonists induce oral irritation that rapidly desensitizes, accompanied by a short-lasting heat hyperalgesia. The latter effect may involve peripheral sensitization of heat-sensitive TRPV1-expressing nociceptors, and/or central sensitization of thermal nociceptive transmission. Acknowledgements: NIH (DE013685 and AR057194)

#P67 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Specificity of Chemical Irritant Tolerance in Birds

Kayla L. Davis[1], David J. Anderson[2], Wayne L. Silver[3]
[1]Wake Forest University/ Biology Winston-Salem, NC, USA,
[2]Wake Forest University/ Biology Winston-Salem, NC, USA,
[3]Wake Forest University/ Biology Winston-Salem, NC, USA

The paradox of the *Capsicum* pepper- that capsaicin (CAP), a trigeminal irritant inherent to the chili fruits, discourages consumption by mammalian seed predators without repelling seed dispersing birds- leads to an inquiry of whether birds also have a tolerance for other trigeminal irritants. We used allyl isothiocyanate (AITC), which stimulates the Transient Receptor Potential A1 (TRP-A1) channel of the trigeminal system and capsaicin (CAP), which stimulates the Transient Receptor Potential V1 (TRP-V1), to assess the specificity of the evolved response of the bird trigeminal system. While the avian tolerance of CAP is well documented, no information exists about the ability of birds to detect AITC. In the present study, house sparrows (*Passer domesticus*) were used to address the question of chemical irritant specificity in the bird trigeminal system. We experimentally compared the birds' food consumption and aversive response to bird chow coated in 100mM AITC solution, 100mM CAP solution, 100mM methyl anthranilate (MA) solution, and 100mM phenethyl alcohol (PEA) solution versus un-enhanced bird chow. Our results show that the birds' food consumption may have been slightly altered by the addition of CAP and MA but unaffected by the addition of AITC and PEA. Marginally significant differences were detected in number of head shaking events between MA and control, and significant differences were observed for food rejection events between MA and control. These data suggest that birds can detect CAP and MA but not AITC and PEA; however, the effects of CAP on aversive response remain unclear. Acknowledgements: Wake Forest University Undergraduate Research Fellowship

#P68 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

The Effect of Menthol Vapor on Sensitivity to Chemical Irritation

Paul M Wise, Charles J Wysocki
Monell Chemical Senses Center Philadelphia, PA, USA

Among other effects, menthol added to cigarettes may modulate sensory response to cigarette smoke, either by masking "harshness" or contributing to a desirable "impact." However, harshness and impact have been imprecisely defined and assessed using subjective measures. Thus, the current experiments used an objective measure of sensitivity to chemical irritation in the nose to test the hypothesis that menthol vapor modulates sensitivity to chemical irritation in the airways. Nasal irritation thresholds were measured for two model compounds (acetic acid and mustard oil) using nasal lateralization. In this technique, participants simultaneously sniff clean air in one nostril and chemical vapor in the other, and attempt to identify the stimulated nostril. People are unable to lateralize based on smell alone, but can do so when chemicals are strong enough to feel. In one condition, participants were pre-treated by sniffing menthol vapor. In a control condition, participants were pre-treated by sniffing an odorless blank (within-subjects design). Pre-treatment with menthol vapor decreased sensitivity to nasal irritation from acetic acid (participants required higher concentrations to lateralize), but increased sensitivity to nasal irritation from mustard oil (lower concentrations were required). The current experiments provide objective evidence that menthol vapor can modulate sensitivity to chemical irritation in the upper airways in humans. Cigarette smoke is a complex mixture of chemicals and particulates, and further work will be needed to determine exactly how menthol modulates smoking sensation.

#P69 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Individual differences in irritation from Ibuprofen covary with Olive Oil but not Capsaicin

Samantha M Bennett, John E Hayes
The Pennsylvania State University/Department of Food Science University Park, PA, USA

Ibuprofen and oleocanthal, a natural irritant in extra virgin olive oil, are unique among chemesthetic stimuli. Unlike most irritants, they are locus specific, triggering irritation mostly in the throat, which is predominately a tickle rather than a burn. Ibuprofen and oleocanthal also share structurally similar motifs, have similar anti-inflammatory properties, and elicit variable irritation across people. Collectively, this is seen as evidence they share a common receptor that may be unique to the throat, a view buttressed by data that oleocanthal intensity is unrelated to carbon dioxide irritation. Subsequent work suggests CO2 irritation occurs via TRPA1 but not TRPV1, so the role of TRPV1 remains unclear. Here, we compare irritation elicited by ibuprofen, extra virgin olive oil and capsaicin, the prototypical TRPV1 agonist, in 30+ participants. Intensity for burn, stinging/pricking, itch, tingle, warm/hot, numb and tickle was collected in replicate over 180s with the generalized labeled magnitude scale (gLMS). From the time intensity profiles, maximum intensity (Imax) and area under the curve (AUC) were extracted for each individual. Across participants, tickle predominated for olive oil and ibuprofen versus burn for capsaicin, although multiple qualities were used for all stimuli. For both AUC and Imax, olive oil and ibuprofen were highly correlated, whereas ibuprofen and capsaicin irritation were not. Unexpectedly, olive oil and capsaicin were also correlated. In summary, these data support the hypothesis that ibuprofen and oleocanthal share a common receptor, and this mechanism is likely TRPV1 independent, due to the absence of a relationship between the irritation from ibuprofen and capsaicin. The olive oil-capsaicin correlation suggests unknown TRPV1 agonists may be present in olive oil.

#P70 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Responsiveness to capsaicin in regular spicy food users versus non-users

Mary-Jon Ludy, Richard D. Mattes
Purdue University West Lafayette, IN, USA

Some studies suggest that capsaicin diminishes orexigenic sensations, but reports are conflicting. One methodological issue potentially accounting for inconsistencies may be differing characteristics of study populations. The purpose of this randomized crossover trial was to contrast responsiveness to capsaicin's appetitive effects in regular spicy food users (n=13) and non-users (n=12). Self-reported appetitive sensations and *ad libitum* challenge meal intake were measured after 1 g red pepper (RP) (1.995 mg capsaicin) and no RP test loads. Subjects were characterized by sensory, physiological, personality, and cultural attributes. Preoccupation with food and desire to consume fatty, salty, and sweet foods were reduced more in non-users than users after RP test loads, but did not vary after no RP test loads. Energy intake was lower after test loads with RP than no RP in non-users, but not in users. These observations suggest that individuals may become desensitized to capsaicin's appetitive effects with long-term spicy food use. Differences between users and non-users were primarily related to sensory and cultural attributes (i.e., users reported consuming spicy foods from an earlier age, rated spicy foods as more palatable, perceived the burn of spicy stimuli to peak at a higher concentration, and could better discriminate this burn than non-users). Users and non-users exhibited comparable responsiveness to noxious pressure pain, oral tactile sensitivity, and auditory sensitivity, varying only in responsiveness to oral thermal heat (i.e., users were more sensitive to increases than non-users). Personality traits did not vary between users and non-users. These findings are of public health interest, given that spicy food consumption is reported to confer weight management and food safety benefits. Acknowledgements: This work was supported by the McCormick Science Institute (MSI) and the National Institutes of Health (NIH) under Ruth L. Kirschstein National Research Service Award (5T32DK076540). Its contents are solely the responsibility of the authors and do not necessarily represent the official views of the MSI or the NIH.

#P71 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Sweet taste and taste nerve lesion modify trigeminal capsaicin perception in adult human subjects

Nicole Schoebel[1], Amir Minovi[2], Hanns Hatt[1]
[1]Department of Cell Physiology, Ruhr-University Bochum Bochum, Germany, [2]Department of Otorhinolaryngology, St. Elisabeth Hospital Bochum, Germany, [3]Department of Cell Physiology, Ruhr-University Bochum Bochum, Germany

Activation of the taste system delivers information about the palatability and nutritional value of food and may also be accompanied by a reduction of pain-related behaviors. The ingestion of mother's milk and different sugars exerted antinociceptive effects in neonatal rats and human infants in previous studies. The effect of sucrose involves the stimulation of taste receptors and is independent of post-ingestive effects. In concert with that, eating-induced analgesia emerges quickly and is of short duration. As another interesting feature, eating-induced analgesia requires the ingestion of hedonic food. In summary, the analgesic effect of eating is well established in neonates, however, it is still a matter of controversy whether it is present in adults. We investigated if trigeminal pain perception is modulated by the taste system in adult humans. The effects of different tastants on the perception of pain induced by acute lingual capsaicin application were studied in a group of 28 healthy subjects. Sampling hedonic sweet taste significantly decreased the mean perceived intensitiy of capsaicin-induced pain whereas aversive bitter taste did not. Anatomical and functional studies suggest that eating-induced analgesia is based on interactions between brain areas associated with taste-processing and pain-regulation. Thus, we studied the effect of peripheral taste system damage on lingual trigeminal perception in 16 subjects with documented anterior hemiageusia. In accordance with the functional model of eating-induced analgesia these subjects were significantly more sensitive to capsaicin on the contralateral compared to the ipsilateral anterior lingual side. Based on our findings, we propose that eating-induced analgesia is present not only in human infants but also in adults.

#P72 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Lateralization of trigeminal stimuli

Thomas Meusel[1,2], Güpfert Mark[2], Birgit Westermann[3], Thomas Hummel[4], Antje Welge-Lüssen[2]
[1]Department of Otorhinolaryngology, Head and Neck Surgery, University Hospital Erlangen Erlangen, Germany, [2]Department of Otorhinolaryngology, University Hospital Basel Basel, Switzerland, [3]Department of Neurosurgery, University Hospital Basel Basel, Switzerland, [4]Smell & Taste Clinic, Department of Otorhinolaryngology, University of Dresden Medical School Dresden, Germany

Objectives In contrast to pure olfactory stimuli intranasally applied trigeminal stimuli can typically be allocated to the side of stimulation. Moreover, intranasal trigeminal sensitivity in an- or hyposmic subjects is reduced. However, so far the temporal resolution of almost simultaneously applied intranasal trigeminal stimuli is unknown. The aim of our study was to examine this temporal resolution. **Methods:** Two groups were examined, each consisting of 30 subjects matched in age and sex. One group consisted of normosmic subjects, the other group of posttraumatic an- or hyposmic patients. Olfactory function was tested psychophysically using the Sniffin Sticks test battery. Bilateral trigeminal stimulation was carried out using a birhinal olfactometer OM8b. The trigeminal stimulus used was CO 60% v/v, the interstimulus interval ranged from 28-32 s, stimulus duration was 200 ms. Time lags tested between right and left side of stimulation were at 40, 80, 120, 160 and 200 ms. Subjects raised their left or right hand to indicate the side on which the stimulus was perceived first. **Results:** Normosmic as well as anosmic subjects were unable to localize the side of trigeminal stimulation correctly at minimal time lags, i.e. at time lags between 40 and 120 ms both groups could only localize the trigeminal stimuli by chance. While 68.5% of normosmic subjects were able to identify the side of stimulation at a time lag of 140 ms correctly, anosmic subjects reached a comparable incidence only at 200 ms. **Conclusions:** The time lag at which intranasal administered trigeminal stimuli can be perceived was estimated between 120 and 200 ms. The reduced trigeminal sensitivity in patients with anosmia or hyposmia leads to an increased time lag in the perception of intranasally, almost simultaneously, applied stimuli.

#P73 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Comparison of perceptual odor adaptation onset time courses for trigeminal and olfactory odorants

Wendy M. Yoder[1], Seth Currlin[2], Allison LaRue[2], Kyle Stratis[2], Kristina M. Fernandez[2], Sweta Pattinaik[2], Alex Molina[2], Jennifer Nguyen[2], Rolondo S Liboy[2], David W. Smith[1,3,4]
[1]Behavioral and Cognitive Neuroscience, Dept. of Psychology, Univ. of Florida Gainesville, FL, USA, [2]Dept. of Psychology, University of Florida Gainesville, FL, USA, [3]Center for Smell and Taste, Univ. of Florida Gainesville, FL, USA, [4]Dept. of Otolaryngology-HNS, Univ. of Florida School of Medicine Gainesville, FL, USA

Adaptation is a critical, time-dependent process that serves to attenuate the neural response to long-duration stimulation, in order to increase the salience of new stimuli. Previous studies suggest that adaptation produced by trigeminal odorants is less rapid, compared with adaptation resulting from olfactory odorants. Recent data from our laboratory demonstrate that the onset time course of odor adaptation can be estimated by use of a simultaneous odorant stimulus paradigm, where the delay from the onset of a relatively long-duration adapting odorant to the onset of a brief target odorant is varied (i.e., at different temporal points along the adaptation time course; Smith et al., Chem Senses. 35:717-25, 2010). Using this technique, we compared the estimated onset time courses of trigeminal odor adaptation to acetic acid with the adaptation time course to an olfactory odorant (vanilla extract) utilizing an automated, liquid dilution olfactometer. Thresholds were estimated in a group of college-aged student volunteers (N=18; 8 females) for a target odorant presented during a simultaneous adapting odorant, at varying adapting to target onset delays. The adapting odorant concentration was fixed at twice threshold and the order of onset delays presented was random. Using this new technique, the results showed that the onset time course of adaptation for the vanilla odorant was relatively faster compared with that for the acetic acid. The asymptotic levels of adaptation, however, were similar for the two odorants. These data agree well with previous studies and suggest that adaptation fro trigeminal odorants is relatively slower than that produced by olfactory odorants.
Acknowledgements: Funded in part by the Office of Research and Graduate Programs and the Institute for Food and Agricultural Sciences, University of Florida

#P74 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Effects of Artificial Sweeteners on Pain Threshold and Tolerance

Kristin McCombs, Bryan Raudenbush, Mark Sappington
Wheeling Jesuit University Wheeling, WV, USA

Previous research has shown that sweet substance consumption can increase pain tolerance and decrease pain ratings. The present study assessed the ability of artificial sweeteners to produce the same results. Participants completed a cold pressor task (hand immersion in 3 degree Celsius water) under six conditions (xylitol, sucralose, aspartame, saccharin, stevia, and a non-sweetener control condition). Tolerance was greatest in the stevia condition, such that participants held their hand in the cold pressor device an average of 30 seconds longer as compared to other conditions, $F(5,135)=2.553$, $p=.031$. Xylitol was rated as the most pleasant artificial sweetener while stevia was rated as the least pleasant artificial sweetener, $F(4, 92)=25.792$, $p=.000$. Similar results were found for sweetness intensity ratings, with xylitol being the most intense and stevia being the least intense, $F(4, 92)= 5.928$, $p=.000$. Results indicate that some artificial sweeteners can influence pain tolerance in a similar way to natural sweeteners. Implications for such research include seeking ways to use such artificial sweeteners for distraction to pain and as an adjunct to pain management.

#P75 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Solitary chemosensory cells play a critical role in limiting access of toxicants and maintaining the normal function of the VNO

Kurt Krosnowski, Janell S. Payano Sosa, Weihong Lin
University of Maryland, Baltimore, MD, USA

The mouse vomeronasal organ (VNO) detects pheromones and other semiochemicals to regulate innate social and sexual behaviors. The VNO also mediates interspecies defensive behaviors (Papes et al 2010). Complex stimuli, such as body secretions are drawn into the lumen of the VNO via the anterior opening and entry duct. Some complex stimuli may contain irritants and contaminants harmful to the VNO. Mechanisms regulating chemical access to the VNO are largely unknown. In the VNO, the majority of the transient receptor potential channel M5 (TRPM5) expressing solitary chemosensory cells (SCCs) is localized in the anterior vomeronasal duct. Our recent research has shown that SCCs of the VNO respond to a broad range of stimuli including high concentration odorants and bitter compounds (Ogura et al 2010). Here we tested the role of the SCCs in protecting the VNO and how this protection might affect VNO mediated behaviors. We exposed the VNO of WT and TRPM5 knockout (KO) mice to several toxicants via the external naris over one to two weeks. When exposed in this fashion the toxicant access to the VNO is limited in WT animals but in the TRPM5KO animals. Following the toxicant exposure regimen, the mice were exposed to a predator odor and tested for defensive responses. We observed diminished defensive behaviors in the TRPM5KO mice exposed to the toxicants, while wild type animals treated under the same experimental conditions exhibit normal reactions. Our results indicate that TRPM5-expressing SCCs play a critical role in limiting access of toxicants and maintaining the normal function of the VNO.
Acknowledgements: Supported by NIH/NIDCD 009269 and ARRA administrative supplement to WL.

#P76 **WITHDRAWN**

#P77 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Oral Experience with Amniotic Fluid Alters Motor Behavior and Chemosensory Responsiveness in the Perinatal Rat

Valerie Mendez-Gallardo[1], Scott R Robinson[2]
[1]University of Iowa/Department of Psychology Iowa City, IA, USA, [2]Idaho State University/Department of Psychology Pocatello, ID, USA

Amniotic fluid (AF) is a biologically important stimulus that may play an influential role in directing and modulating the behavior of fetal and newborn mammals. In the rat fetus, our laboratory has demonstrated that oral exposure to AF reduces responding to novel chemosensory stimuli and alters the temporal patterning of spontaneous motor activity. Rat fetuses prepared for *in vivo* behavioral testing on E20 of gestation received a 20 µl intraoral infusion of AF. Fetuses showed a significantly reduced facial wiping response to lemon odor. AF begins to alter fetal behavior within 15 s after oral exposure, but effects of AF persist for only 3-4 min. The behavioral effects of AF were blocked by pretreating subjects with naloxone or nor-binaltorphimine, confirming that AF evoked activity at kappa receptors of the fetal opioid system. Fetuses exposed to AF after preparation with an esophageal/tracheal ligature continue to express behavioral effects, suggesting that AF acts through oral exposure alone, either by permitting transport of a neuroactive substance directly to the fetal CNS, or by evoking a chemosensory response that results in activation of the opioid system. After birth, AF odor continues to influence behavior of infant rats. Pups tested one day after birth (P1) exhibited differential behavioral responses to AF, including: (a) behavioral activation upon exposure to AF odor, (b) crawling locomotion oriented toward the source of AF odor, (c) enhanced oral grasping of an artificial nipple in the presence of AF odor, and (d) classical conditioning to a novel conditioned stimulus paired with AF as the unconditioned stimulus. Together, these findings suggest an important role for AF in modulating CNS activity and promoting behavioral continuity during fetal and neonatal development. Acknowledgements: Supported by NIH grant HD 33862 and HSSRC grant from Idaho State University to SRR.

#P78 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Effects of perinatal flavour exposure on stress-related parameters in piglets after weaning

Marije Oostindjer, J. Elizabeth Bolhuis, Kristina Simon, Henry van den Brand, Bas Kemp
Wageningen University, Adaptation Physiology Group Wageningen, Netherlands

Previously we found positive effects of perinatal flavour exposure to anethol (anise flavour) through the maternal diet on food intake, growth and behaviour of piglets in the first two weeks postweaning when exposed to anise-flavoured feed. Piglets did not, however, prefer the anise-flavoured feed, suggesting that the positive effects of perinatal flavour learning could not be attributed to reduced food neophobia, but were mediated by reduced stress due to the presence of a familiar flavour, and this was investigated in the current study. Sows were offered an anethol-flavoured diet during late gestation (d98-115) and lactation (d2-d24, Flavour treatment), or a control diet throughout gestation and lactation (Control treatment). Piglets were weaned on day 25 and were given either two feeders with control food and a permeable container with anethol-flavoured food (Air treatment) or one feeder with anethol-flavoured food, one with control food and a container with control food (Feed treatment), resulting in a 2x2 design. Treatments did not affect food intake, anethol preference or growth. Flavour-piglets showed more play behaviour, indicative of low stress, especially in the Air-treatment (1.14%, vs. others: 0.76% of observations, $p=0.02$).Parameters indicative of high stress were higher in Control-piglets: chewing on pen mates (Control: 0.41%, Flavour: 0.30% of observations, $p=0.05$) and vocalising on day 1 postweaning (Control: 39%, Flavour: 32% of observations, $p=0.02$). Piglets in the Air-treatment were less reluctant to touch a novel object in the pen on day 2 postweaning than Feed-piglets. Salivary cortisol levels will also be reported. Perinatal flavour learning decreased stress in newly weaned piglets, yet it seems not

necessary to provide the flavour in the food to obtain such effects. Acknowledgements: This work was supported by NWO-STW (grant 07722), with co-financers Lucta S.A., Product Boards for Livestock and Meat (PVV), Product Board Animal Feed (PDV), Nutreco Nederland B.V., and Verbakel B.V.

#P79 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Taste of Love: a Role for the Gustatory Sensory System in Mating Behaviors

Yehuda Ben-Shahar
Washington University/Biology St. Louis, MO, USA

Sexual behaviors are complex and require sensory inputs from multiple modalities. The majority of the signals, their cognate receptors, and the cells that mediate them are still poorly understood. We use the genetic model *Drosophila melanogaster* to investigate how animals encode and process socially related signals. Here we focus on the role of the gustatory system in mediating sex-related signals in insects. We have identified a novel ligand-gated ion channel (*aguesic*, *agu*) that plays a role in chemosensory functions underlying mating behaviors in *Drosophila*. *agu* is expressed in a subpopulation of chemosensory bristles on the legs and wings that are distinct from those expressing feeding-related gustatory receptors. Moreover, *agu* is not expressed in the labellum, the primary organ involved in taste, and disrupting *agu* or inhibiting activity of *agu*-expressing neurons does not alter gustatory responses. Instead, *agu* is sexually dimorphic, and co-expressed with the sex-determination gene *fruitless* in adult legs. Consistent with this pattern, blocking *agu*-positive neurons or mutating the *agu* gene delays the initiation and reduced the intensity of male courtship. These data indicate that *agu* and the cells expressing it are an essential component of the peripheral sensory system that determines sexual behavior in *Drosophila*. In addition, our results indicate the presence of at least two types of chemosensory bristles on appendages, some specialized to influence mating and some for feeding. Acknowledgements: NIH/NIDCD

#P80 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Molecular and neuronal mechanisms that generate sexually dimorphic behavior

Sandeepa Dey, Lisa Stowers
The Scripps Research Institute/ Department of Cell Biology
La Jolla, CA, USA

The sense of olfaction is vital for survival and enables individuals to locate food, mating partners, threat and predators. Mammals often display gender dimorphic responses to odor cues. Thus, for instance in mice, a male odor will elicit aggressive behavior when detected by a male and no aggression when detected by a female. How does the same set of stimulants elicit such widely different responses in the two sexes? The neural mechanisms that underlie these distinct response patterns are unknown. One hypothesis proposes that male and female sensory neurons equally detect odor cure which activate anatomically and functionally different neural circuits. Recent findings in drosophila and mice support this view[1-5]. An alternative hypothesis is that detection of olfactory cues by peripheral sensory neurons is itself gender specific. There is little precedent for this model, specifically in mammals, where visual and auditory cues are gender invariant. Chamero et al[6] have recently purified individual pheromone odorants that promote intermale aggression, a stereotypic gender dimorphic behavior in the mouse. These ligands now enable us to specifically activate aggression promoting neural circuits to study and compare the underlying mechanisms that generate sexually dimorphic behavior. We are investigating the neural and molecular basis that may generate sexual differences in detection and processing of olfactory cues. We anticipate our results will provide new insight to mechanisms underlying neural activity that generates stereotypic social behavior.

#P81 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

A Functional Main Olfactory System is Necessary and Sufficient for the Initiation and Maintenance of Maternal Behavior in Mus musculus

Kyle M. Roddick, Heather M. Schellinck
Dalhousie University Halifax, NS, Canada

It is now evident that both the main olfactory system (MOS) and the accessory olfactory system (AOS) may be activated by general and pheromonal odorants. Moreover, it has been demonstrated that the MOS is required to activate the AOS to initiate male aggression and puberty acceleration in mice. It is not clear, however, if the main olfactory system may act independently or whether action of both systems are required for specific behavioral responses to occur. We report here the results of a double dissociation study designed to examine how olfactory cues modulate maternal behaviour in mice. Ablation of the main olfactory epithelium with $ZnSO_4$ was found to significantly reduce pup survival. In addition, ANOVA revealed that pup growth, nest quantity and quality, nursing behavior, and pup retrieval were significantly impaired in $ZnSO_4$ treated mice compared with animals in which the AOS had been deactivated by surgical removal of the vomeronasal organ. Mice with both a nonfunctional MOS and a nonfunctional AOS also had significant deficits in all maternal behaviors; control mice with sham inactivation of both systems showed no significant deficits. These results suggest that the main olfactory system is both necessary and sufficient for the expression of maternal care whereas the accessory olfactory system does not appear to be essential for such behavior. Acknowledgements: Natural Sciences and Engineering Research of Canada Discovery Grant to HMS

#P82 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Sea Lamprey Neural Responses to Pheromone Exposure

Anne M. Scott, Yu-Wen Chung-Davidson, Huiyong Wang, Weiming Li
Michigan State University/Fisheries & Wildlife East Lansing, MI, USA

Sexually mature male sea lampreys (*Petromyzon marinus*) release a pheromone called 3-keto-petromyzonol sulfate (7α, 12α, 24-trihydroxy-5α-cholan-3-one 24-sulfate or 3kPZS) that stimulates the olfactory receptor neurons and downstream neural circuits. In behavioral testing, 3kPZS triggers responses of varying amplitude in females depending on their maturity. We hypothesized that 3kPZS olfactory stimulation alters brain excitatory neurotransmission in sea lampreys in a sexually dimorphic manner. To test this hypothesis, we examined the effects of pheromone exposure or in combination with AMPA antagonist (CNQX) treatment, on transcripts of Jun and glutamate $AMPA_2$ receptor genes, as well as glutamate concentrations in sea lampreys. Preovulatory female and prespermiating male sea lampreys were injected intraperitoneally (1ml/Kg) with saline or 10μM CNQX, followed by immediate exposure to the vehicle (1 ml 0.91ppm methanol) or 10^{-10}M 3kPZS for 2h. Messenger RNA concentrations were analyzed by real-time quantitative PCR and neurotransmitters were measured by a Liquid Chromatography-Tandem Mass Spectrometry method. Exposure to 3kPZS decreased Jun mRNA transcripts in both the forebrain and hindbrain of prespermiating males but not in preovulatory females ($p<0.05$, one-way ANOVA). In CNQX treated animals, 3kPZS exposure decreased $mGluR_2$ mRNA in the forebrain of prespermiating males but not in preovulatory females ($p<0.05$, one-way ANOVA). The hindbrains of prespermiating males contained lower glutamate concentrations than preovulatory females after treatment ($p<0.05$, one-way ANOVA). These results demonstrate sexually dimorphic neural responses to pheromone exposure which may be associated wtih the sexually dimorphic behavioral responses in sea lampreys.
Acknowledgements: This study was supported by NIH grant 5R24GM083982 and Great Lakes Fishery Commission.

#P83 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Olfactory Imprinting and Discriminating Ability of Sockeye Salmon

Hiroshi Ueda
Hokkaido University Sapporo, Japan

The amazing homing abilities of salmon to their natal streams among thousands of river for spawning have been investigated mainly focusing on their olfactory functions. However, there are still many unknowns how to imprint and discriminate their natal stream odorants during downstream migration in juveniles and upstream migration in adults, respectively. Olfactory imprinting and discriminating ability of lacustrine sockeye salmon (*Oncorhynchus nerka*) were examined using a single amino acid (L-proline; Pro). The electro-olfactogram responses to one-year-old lacustrine sockeye salmon exposed to Pro from March to June for 2 weeks were significantly greater than those of non-exposed control fish, but not those of test fish exposed in July. When Pro and control water were added to the water inlets of a two-choice test tank during the spawning season 2 years after the test water exposure, 80% of maturing and matured test fish exposed from March to June showed a preference for Pro, whereas those exposed in July did not. Moreover, to investigate odor information processing of the natal stream, the blood oxygenation level-dependent (BOLD) functional magnetic resonance imaging (fMRI) was applied to investigate the response to the natal stream water in the olfactory bulb and telencephalon of lacustrine sockeye salmon. The strong responses to the natal stream water were mainly observed in the lateral area of dorsal telencephalon, which are homologous to the medial pallium (hippocampus) in terrestrial vertebrates. These recent findings are discussed in relation to physiological mechanisms of the amazing imprinting and homing migration in salmon.
Acknowledgements: The present study was supported in part by Grant-in-Aid for Scientific Research from the Ministry of Education, Culture, Sports, Science and Technology (MEXT), Japan, from the Japan Society for the promotion of Science (JSPS), from the Mitsubishi Foundation, from the Mitsui & Co. Ltd, and from the Hokkaido University.

#P84 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Individual odortypes in mouse urine are not disrupted by addition of exogenous chemicals

Jae Kwak[1], Talia Martin[1], Maryanne C. Opiekun[1], Claude C. Grigsby[2], George Preti[1,3], Kunio Yamazaki[1], Gary Beauchamp[1]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]Air Force Research Laboratory Dayton, OH, USA, [3]University of Pennsylvania/Dermatology Philadelphia, PA, USA

Genetically determined body odors that distinguish one individual from another are termed odortypes. The most thoroughly studied odortypes, highly expressed in urine, are those specified by genes of the major histocompatibility complex (MHC) but other odortypes originate from variations in other loci (background genes) in the rest of the genome. From the perspective of ecology and evolution, we expect that these genetically-determined odortypes would be resistant to alteration by environmental or other kinds of variation. These expectations have been verified in studies that have shown that trained mice continue to recognize MHC odortypes in the face of changes in urine volatiles induced by diet change, certain diseases, or even removal of urinary proteins. To put this expectation to a more rigorous test, we asked whether odortypes would continue to be recognized when the pattern of volatiles was substantially altered by addition of exogenous chemicals. We used two chemicals to manipulate urine: guanidine hydrochloride (a protein denaturant) and butylated hydroxytoluene (a ligand of urinary proteins). Mice trained to discriminate between unadulterated urine odors of mice that differed only in MHC type or of mice derived from different genetic backgrounds generalized the discrimination to the altered urine samples. Thus, the discrimination of individual odor signatures is robust even in the face of massive changes in volatile profiles. It remains to be determined what chemically constitutes these profoundly conserved individual odor signatures.

#P85 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Differential binding affinities between volatile ligands and urinary proteins due to background genetic variation in mice

Jae Kwak[1], Claude C. Grigsby[2], Jesusa Josue[1], Mateen M. Rizki[3], George Preti[1,4], Kunio Yamazaki[1], Gary K. Beauchamp[1]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]Air Force Research Laboratory Dayton, OH, USA, [3]Wright State University/Computer Science and Engineering Dayton, OH, USA, [4]University of Pennsylvania/Dermatology Philadelphia, PA, USA

Two different structural classes of chemical compounds present in mouse urine have been extensively investigated as possible chemical signals: volatiles and the major urinary proteins (MUPs). These classes of compounds interact closely since MUPs have a hydrophobic pocket that sequesters volatile ligands. While qualitative and/or quantitative differences in each chemical class have been reported in different inbred mouse strains, previous studies have examined only one of the classes at a time. No study has compared these two sets simultaneously, particularly binding affinities between ligands and proteins in urines of different strains. We hypothesized that differential binding affinities between these chemical classes may be observed among different strains due to their qualitative and/or quantitative differences caused by genetic variation. To test this hypothesis, we compared the release of ligands in male urines of three different inbred strains (C57BL/6J, BALB/b and AKR) before and after denaturation of urinary proteins, mainly MUPs. As expected, a unique volatile profile with quantitative differences in volatile compounds was observed in the intact urine of each strain. Upon denaturation, these profiles changed dramatically. Several dozen volatile ligands were released from urine after denaturation and their binding affinities remarkably differed between strains. These data demonstrate that the binding affinities in volatile ligands and urinary proteins differ between strains, adding another level of complexity to chemical communication in mice.

#P86 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Ultra-high olfactory sensitivity for the sperm-attractant aromatic aldehyde bourgeonal in CD-1 mice

Linda Larsson, Matthias Laska
Linköping University Linköping, Sweden

Recent studies have shown that certain aromatic aldehydes are ligands for olfactory receptors expressed both in olfactory sensory neurons and in mammalian sperm cells. Using a conditioning paradigm, the olfactory sensitivity of eight CD-1 mice for seven aromatic aldehydes was investigated and compared to that of four spider monkeys and 20 human subjects tested in parallel. With all seven stimuli, the mice discriminated concentrations ≤0.01 ppm (parts per million) from the odorless solvent, and with bourgeonal the animals were even able to detect concentrations as low as 0.1 ppq (parts per quadrillion) which constitutes the lowest olfactory detection threshold value reported in this species so far. Spider monkeys and human subjects were found to be less sensitive than the mice but similar to each other in their olfactory sensitivity and detected concentrations <1 ppm with all seven aromatic aldehydes. With several stimuli single individuals of both primate species even discriminated concentrations <1 ppb from the solvent. Across-odorant patterns of sensitivity correlated significantly between humans and spider monkeys, but not between mice and humans or spider monkeys, respectively. No significant correlation between presence/absence of an oxygen-containing moiety attached to the benzene ring or presence/absence of an additional alkyl group next to the functional aldehyde group and olfactory sensitivity was found in any of the three species. However, the presence of a tertiary butyl group in para-position (relative to the functional aldehyde group) combined with a lack of an additional alkyl group next to the functional aldehyde group may be responsible for the finding that all three species were most sensitive to bourgeonal.

#P87 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

L-Felinine as a Potential Reproductive Inhibitor in Rodents

Vera V. Voznessenskaya, Artyom B. Klinov, Tatiana V. Malanina
A.N.Severtzov Institute of Ecology & Evolution Moscow, Russia

Felinine is a unique sulfur-containing amino acid found in the urine of domestic cats and select members of the Felidae family (Rutherfurd et al.,2002). In our earlier studies we showed that exposures of mice *Mus musculus* to urine from feral cats *Felis catus* under semi-natural conditions significantly affected survivorship of offspring. Manipulations with the diet of predator and non-predator urine donors revealed the key role of sulfur-containing compounds.In current study we examined the influence of the precursor of potential Felidae family pheromone felinine on reproductive output in mice and rats. We used three basic approaches: behavioral, endocrinological and immunohistochemical. We recorded number of newborn pups, sex ratio, weight of pups at weaning. Corticosterone metabolites were monitored non-invasively (Touma et al.,2004). Fos positive cells were recorded in response to stimulation with felinine in the main olfactory and accessory olfactory bulbs and in vomeronasal receptor tissue. Cotton balls soacked with felinine(0.05% w/v, 0.05 ml, US Biologicals) were placed directly into home cages of mice and rats each other day during all period of gestation. Control animals were exposed to tap water. Exposure to felinine affected sex ratio in mice (n=40, $p<0.001$) and rats (n=36, $p<0.001$) in favour of males. In mice felinine also affected litter size (n=40, $p<0.05$). By the day of weaning in control groups of animals average weight of pups was significantly ($p<0.001$) higher then in the exposed to felinine. Both, mice and rats showed seasonal sensitivity to felinine. We observed long lasting elevation of corticosterone under felinine exposured in mice (n= 10, $p<0.001$) but not in rats. The data obtained indicate that felinine is a potential chemical signal in mice. Acknowledgements: Supported by Russian Foundation for Basic Research 10-04-1599 to VVV

#P88 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

(Z)-5-tetradecen-1-ol produced in the male preputial gland as a natural ligand for an odorant receptor in mice

Keiichi Yoshikawa[1], Hiroaki Nakagawa[1], Naoki Mori[2], Hidenori Watanabe[2], Kazushige Touhara[1,2]
[1]Department of Integrated Biosciences, The University of Tokyo Chiba, Japan, [2]Department of Applied Biological Chemistry, The University of Tokyo Tokyo, Japan

Olfactory signals play a major role in regulating a variety of biological and physiological functions in individual organism. To understand intraspecies communication via the olfactory system, it is important to identify natural substances that are used to convey individual information. Although several odorant receptors (ORs) have been matched with cognate odorous compound, the natural ligands, which are produced and received within the same species, are largely unknown. Here, we searched for a natural ligand for a mouse OR from several tissue extracts. Using a heterologous OR expression system and assay-guided fractionation, we identified (Z)-5-tetradecen-1-ol as an OR ligand from the male preputial gland, an exocrine gland that is located in front of the genitals. (Z)-5-tetradecen-1-ol is an unsaturated aliphatic alcohol that has not been identified in mammals but has a structure similar to some insect sex pheromones. Given that the preputial gland is thought to synthesize scents related to reproductive behavior, our results suggest a possibility that some ORs sense (Z)-5-tetradecen-1-ol released from the preputial gland as male information.

#P89 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

The Florida Manatee, *Trichechus manatus latirostris*, Chemosensory System: Histological and Behavioral Assessment of the Reproductive Use of Taste and Smell

Meghan L. Bills[1], Julie D. Sheldon[2], Kelly M. Evans[2], Don A. Samuelson[1], Iskande V. Larkin[1]
[1]University of Florida/College of Veterinary Medicine/Aquatic Animal Health Program Gainesville, FL, USA, [2]University of Florida/Department of Animal Sciences Gainesville, FL, USA

The use of taste and smell in reproduction is well documented in terrestrial mammals and small aquatic vertebrates but has never been fully examined in aquatic mammals. Anecdotal evidence indicates that the fully aquatic, endangered Florida manatee, *Trichechus manatus latirostris*, is capable of sensing a female in estrus. This project seeks to define the chemical sensing abilities of the manatee including a characterization of its receptive and transmission anatomy and behavioral assessment of male reaction to female urine. Behavioral assays of three captive male manatees indicate that males are able to perceive and react to female estrus urine. Through gross and histological examination of suspected sites of chemoreception a thorough analysis of the manatee's anatomical capabilities has been possible. To date; eight males and six females of varying ages have been assessed using gross documentation, histological processing, and transmission electron microscopy. The examination of the mouth and nasal passages has corroborated the presence of olfactory epithelium and taste buds of the tongue. Previously undescribed in the manatee, the soft palate also appears to have taste buds and there are large apocrine glands located at the recto-anal junction. These glands are similar to the circumanal scent glands of the dog. These anal glands are present in fetal, juvenile and adult male and female manatees. The glands contain lipids and mucin as demonstrated histochemically by oil red o and PAS stains. It is apparent that the manatee has chemosensory structures similar to those used in pheromone transmission and reception by terrestrial mammals. Through continued anatomic analysis and additional behavioral assays the specific structures used by male manatees to detect females in estrus can be determined. Acknowledgements: Funding is provided through the Florida Fish and Wildlife Conservation Commission, Whitney Marine Laboratory and the University of Florida Aquatic Animal Health Program, the University of Florida College of Veterinary Medicine and Sigma Xi-Grant in Aid of Research.

#P90 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Ring-tailed Lemurs (*Lemur catta*) Preferentially Scent Mark Certain Types of Vegetation

Julie C. Hagelin[1], Jen C. Crick[1], Alison Jolly[2]
[1]Swarthmore College, Biology Swarthmore, PA, USA, [2]University of Sussex, Biology and Environmental Science Brighton, United Kingdom

Ring-tailed lemurs (*Lemur catta*) from Madagascar rely heavily on social chemosignals. Males mark plant stems with their forearm by simultaneously wiping a scent gland and gouging plant bark with a keratinized spur. Both sexes also mark with anogenital glands. We studied the kinds of plants that lemurs scent mark via two methods. (1) We quantified plant morphology by measuring 100 scent marked stems and compared each to its nearest, unmarked neighbor. (2) We surveyed all plants at our study site (n = 1911) in order to calculate the relative abundance of each major plant genus and compare it to the proportion marked by lemurs. Lemur-marked stems were often forked or Y-shaped (df = 1, $\chi^2 = 27.5$, $P<0.0001$) and smaller in diameter (3.0cm ± 3.4) than unmarked neighbors (14.8cm ± 15.1; df = 99, paired-$t = 7.57$, $P<0.0001$). Markings were typically deposited at a mean height of 42.3cm ± 21.6 on vertical stems. Stems growing at acute angles also made up 32% of all marked plants, compared to only 5% of unmarked neighbors (df = 1, $\chi^2 = 19.9$, $P<0.0001$). Lemurs overwhelmingly marked the genus *Uncarina* most often (45.3% of all marked plants; 84.5% of all *Uncarina* sampled), yet this genus made up only 4.8% of the plants at our site (df = 1, $\chi^2 = 1223.3$, $P<0.0001$). Interestingly, *Uncarina* is aromatic, known for its foul-smelling sap, which could synergistically interact with male scent, as spur gouging exposes green tissue. *Uncarina*'s small, forked stems provided multiple hand grips with which a male could "anchor" itself with one arm while marking with the other. Small, angled stems also appeared to be a good "fit" for anogenital marking. Captive facilities may benefit by providing lemurs with plants of appropriate stem shapes, sizes, and possibly odors, so as to promote scent marking behaviors observed in the wild. Acknowledgements: Swarthmore College Field Funds

#P91 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

"Stink Flirting" in Ring-tailed Lemurs (*Lemur catta*): Male Olfactory Displays Operate as Costly Signals Impacting Female Choice and Male Mating Success

Amber D Walker-Bolton, Caroline Ross
Roehampton University/Department of Life Sciences London, United Kingdom

Ring-tailed lemurs (Lemur catta) communicate primarily via chemical signals and the visual and auditory displays associated with scent communication. Males engage in tail anointing and wafting displays towards both male and female targets. We examined the function of these displays from male senders to female receivers in a mating context. Data were collected on two groups of wild ring-tailed lemurs at Berenty Reserve, Madagascar. Male tail anointing and wafting displays were shown to operate as costly signals of male genetic quality to pre-oestrous and oestrous females in three ways. First, tail anointing displays placed a male at risk, as males were targeted for aggression at a higher rate in the 40 seconds after tail anointing (X + SE = 0.93+1.083 g, N = 120) than they were during matched controls (X + SE = 0.08+0.433 g, N = 120). Second, females exhibited preferential mate choice for males who directed tail anointing and wafting displays towards them, even after controlling for male rank (anoint: $T_{rw, xy, z}$=0.31, P=0.01, waft: $T_{rw, xy, z}$=0.25, P=0.02). Finally, male mating success (copulations) correlated with the performance of tail anointing and wafting displays when male rank was controlled for (anoint: d.f. = 12, r = 0.53, P=0.5, waft: d.f. =12, r=0.55, P =0.04). This study provides the first evidence that male tail anointing and wafting displays impact mating outcomes in this species. These findings show how male ring-tailed lemurs use a species-specific behavioural display to bypass the bulletin board effect inherent in scent marking. These olfactory displays act as honest signals of genetic fitness while allowing a means for the efficient chemical transfer of genetic quality information from male to female. Acknowledgements: This work was supported by a Roehampton University scholarship, a L.S.B. Leakey Trust grant and a partial award from the Roehampton University Sacred Heart Scholarship Fund.

#P92 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Convergence in the Orosensory Nucleus of the Solitary Tract: An investigation using confocal microscopy, electron microscopy and statistical modeling

James A. Corson, Alev Erisir
University of Virginia/ Department of Psychology Charlottesville, VA, USA

Despite fairly extensive knowledge regarding the inputs and outputs of the orosensory nucleus of the solitary tract (NTS), the intrinsic circuitry remains relatively understudied, knowledge of which is essential to understanding orosensory processing in central gustatory circuits, as well as to the interpretation of plastic changes resulting from the perturbation of sensory information. In order to confirm the efficiency and reliability of high-magnification confocal analysis in identifying putative synaptic junctions, we utilized electron microscopy and statistical modeling. We reprocessed tissue previously scanned on the confocal microscope for electron microscopy to measure the distances between confocally-identified axonal and dendritic processes. Confocal and electron microscopic distances were linearly correlated with confocal measurements displaying -0.35 m offset, revealing that at least two voxels of overlap was necessary for two processes to be accurately identified as apposing. Monte Carlo statistical modeling revealed that labeled voxels are present within our confocal apposition zones at a rate much greater than chance. Furthermore, the presence of a contact is minimally correlated with labeled voxel density, all of which suggest biological specificity rather than stochastic co-localization. We observed a convergence of the chorda tympani and glossopharyngeal nerves onto individual projection neurons. Often, like-inputs were clustered together onto individual dendritic branch segments, demonstrating an input compartmentalization that may relate to signal strengthening. This is the first anatomical analysis of such convergent innervation and thus will be foundational for future studies of the functional microcircuit in the orosensory NTS. Acknowledgements: NIH 1RO1DC10183

#P93 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Ultrastructural Morphology and Synaptic Organization of Parabrachial Input to the Rat Gustatory Thalamus

Stephen L Holtz, James A Corson, Anqi Fu, Alev Erisir
University of Virginia/ Dept Psychology Charlottesville, VA, USA

Ventroposterior parvocellular nucleus (VPpc) is the thalamic relay of gustatory sensation from the tongue to insular cortex via nucleus tractus solitarii (NTS) and parabrachial nucleus (PBN). Unlike other sensory thalamic nuclei, explorations of synaptic circuitry in gustatory thalamus have been rudimentary. We examined morphological properties of synaptic circuitry in VPpc, starting with identified PBN input. Anterograde tracer injected into the gustatory-responsive waist region of PBN (n=10) led to prominent fiber labeling in the ipsilateral VPpc, within a thin strip extending 400 m AP x 500 m ML, located dorso-medial to medial lemniscus. At the ultrastructural level, PBN axons were observed to be myelinated and bear very large terminal boutons that form asymmetric, adherent and perforated synapses onto large dendrites and dendritic appendages. Labeled terminal morphology resembled RL-type terminals bearing Type 1 synapses found in other sensory thalamic nuclei. PBN boutons were often encased in glia; however glomeruli and triadic arrangements, characteristic features of other sensory thalamic nuclei, were not encountered. Dense core vesicles were detectable in lightly stained terminals. Also typical of primary inputs to thalamic nuclei, PBN terminals were more than 5 times larger than unlabeled terminals, while they constituted about 5-10% of all synapses. PBN terminals wrapped around regions where dendrites emerged from soma, and formed multiple synapses onto single cells. Our results will allow differential comparison of the anatomical basis of functional processing in thalamic sensory nuclei, and may reveal unique properties of rodent thalamic gustatory processing. Acknowledgements: NIH-NIDCD 1R01AC10183 (AE), UVa Double Hoo Undergraduate Award (SH & JC)

#P94 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Transgenic labeling of the gustatory neural pathway originating from phospholipase C-β2-expressing taste receptor cells in medaka fish

Takashi Ieki[1], Shinji Okada[1], Yoshiko Aihara[1], Makoto Ohmoto[1], Keiko Abe[1], Akihito Yasuoka[2], Takumi Misaka[1]
[1]Department of Applied Biological Chemistry, Graduate School of Agricultural and Life Sciences, The University of Tokyo Tokyo, Japan, [2]Department of Biological Engineering, Maebashi Institute of Technology Maebashi, Japan

As already shown in our study, the fish ortholog of phospholipase C- 2 (*plc*-β2) is expressed in a subset of taste receptor cells (TRCs) that transmit favorable and aversive tastes signals to the central nervous system. Although fish and mammals share some common signaling mechanisms in the peripheral gustatory system, the information regarding the similarities and differences between their central neural pathways is insufficient. To analyze the neural pathways originating from *plc*-β2-expressing cells in fish, we generated transgenic medaka fish expressing the trans-neuronal tracer, wheat germ agglutinin (WGA), under the control of medaka *plc*-β2 gene regulatory region. In the taste buds of the transgenic fish, faithful and robust expression of WGA mRNA was observed in *plc*-β2-expressing TRCs. Immunohistochemical examination with anti-WGA antibody revealed that WGA protein was transported in a subset of neurons in the sensory ganglia of facial, glossopharyngeal and vagal nerves. At the medullary level, WGA signals were observed in the facial and vagal lobes, both of which are anatomically homologous to the solitary nucleus in mammals. The signals also existed in the reticular formation and vagal motor nucleus, which mediate swallowing reflex. Interestingly enough, a significant number of WGA-positive neurons were observed in more central regions homologous to the higher order gustatory relay nuclei in mammals, such as the thalamus and endbrain. In considering the absence of WGA signals in these regions of t1r3-WGA transgenic mice, this study is the first to report the labeling of the higher order gustatory neurons connected specific taste bud cells, providing information about a common neural architecture of the gustatory transduction pathway in vertebrates.
Acknowledgements: This research was supported by the Japan Society for the Promotion of Science (#09016357, #20688015, #20780090, #21658046, #19658053, #20380183), Grant for Umami research and the Iijima Foundation for the Promotion of Food Science and Technology.

#P95 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Topographic Organization of Endogenous Opiates in the Rostral Nucleus of the Solitary Tract: Functional Implications

Nicole R Kinzeler[1], Yuchio Yanagawa[2], Susan P Travers[1]
[1]Oral Biology, Ohio State University Columbus, OH, USA, [2]Department of Genetic and Behavioral Neuroscience, Gunma University Graduate School of Medicine Maebashi, Japan

The effects of opiate receptor ligands injected into the rostral nucleus of the solitary tract (rNST) suggest that endogenous opiates modulate taste processing and oromotor integration. Fibers expressing enkephalins (ENK) (mixed mu/delta agonists) and endomorphins (END) (pure mu agonists) are both present in rNST but their relationship to functionally distinct regions and cell types has not been described. To address these questions, we used multiple fluorescent immunohistochemistry for ENK and END and retrograde tracing from the parabrachial nucleus (PBN) in a mouse line where EGFP is expressed under the control of the GAD67 promoter (Tamamaki '03). Results reveal a strong topography for ENK and END fibers. Both were densely distributed in the medial subdivision of rNST with a secondary terminal field in the ventral subdivision. ENK labeling was more robust and extended weakly into the rostral central subnucleus. In contrast to other EGFP-GAD mouse lines (Travers '07; Wang & Bradley '10), rNST GFP soma in this knock-in strain were distributed throughout the nucleus with the striking exception of the medial rNST where ENK/END staining was strongest. PBN projection neurons were most numerous in the rostral central subnucleus, lateral to the densest ENK, but interspersed with a few, varicose ENK fibers. More ENK fibers intermingled with the sparser population of ventrally-located PBN projection cells. Interestingly, previous studies have shown that preganglionic parasympathetic neurons and NST-reticular projection neurons are prominent in the medial and ventral NST, precisely where ENK/END fibers are densest. This suggests an intimate relationship between endogenous MOR ligands and NST neurons controlling oromotor and autonomic output, a hypothesis that will be assessed by future experiments. Acknowledgements: Supported by DC60020944

#P96 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Anatomical convergence between lingual and dental afferents within the gustatory NTS and trigeminal ganglion in the rat

Aurelie Vandenbeuch[1,2,4], Adeline Braud[1,2], Fouzia Zerari-Mailly[2,3], Yves Boucher[1,2]
[1]UFR Odontology, University of Paris Denis Diderot Paris, France, [2]CRicm UMRS 975 Paris, France, [3]UFR Biologie, University of Paris Denis Diderot Paris, France, [4]Department of Otolaryngology and Rocky Mountain Taste and Smell center, University of Colorado Denver Denver, CO, USA

A recent clinical study revealed an increase in taste thresholds with dental deafferentation (Boucher *et al.,* 2006), suggesting interactions between dental and lingual afferents. In order to investigate the anatomical connections between these two systems in the brainstem, we used a triple-labeling approach on rats. NTS neurons were retrogradely labeled with fluorogold from gustatory solitary-parabrachial neurons (PBN) and the trigeminal inferior alveolar nerve (IAN) and the chorda tympani (CT) or the lingual nerve (LN) were antero-retrogradely labelled with BDAs or fluororuby. Our results show that NST second order gustatory neurons receive input from both by IAN afferents and CT or LN afferents. Moreover, electrophysiological recordings of NTS neurons showed a modulation of the taste response by electrical stimulation of the IAN. Taken together, our results provide an anatomical and functional basis to support trigeminal dental and gustatory interactions in the brainstem. Furthermore, we also observed neuronal cellular bodies in the trigeminal ganglion double-labeled from LN and IAN afferents suggesting a possible integration of trigeminal lingual and dental information at the ganglion level.

#P97 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Primary Cilia in the Rostral Nucleus of the Solitary Tract (rNST)

Min Wang[1], Robert M. Bradley[1,2], Charlotte M. Mistretta[1]
[1]Department of Biologic and Materials Sciences, School of Dentistry, University of Michigan Ann Arbor, MI, USA,
[2]Department of Molecular and Integrative Physiology, School of Medicine, University of Michigan Ann Arbor, MI, USA

Neurons in several brain areas posses a single, non-motile, primary cilium shown to have significant roles in brain development, neurogenesis and signaling. We report that primary cilia are present on neurons of the rNST, the first relay in the central taste pathway. Cilia were identified in embryonic, postnatal and adult mice and rats using adenylyl cyclase III (ACIII) antibody, a specific marker of primary cilia. We observed that about 50% of rNST cells have a single cilium extending from the cell surface in adult mouse and rat. Cilia were on cells in all subdivisions of the rNST. There was no significant difference in number of cells with cilia among the subdivisions. In embryonic mouse, cilia were first identified at day E16 in rNST. The average length of the cilia increased during embryonic development and postnatally to adulthood. GABAergic interneurons in rNST also had cilia, identified in 60% of GAD65-GFP neurons and 65% of GAD67-GFP neurons. Cilia are aberrantly formed in Oak Ridge Polycystic Kidney (ORPK) mice and we therefore used these mice to assess cilia defects in rNST. ORPK mice had shorter and less numerous cilia in rNST neurons than wild type mice. Furthermore, investigators have shown that central neuron cilia often express somatostatin receptors. We found that somatostatin receptor 3, but not other somatostatin receptors, co-localized with ACIII in rNST; this suggests that rNST cilia may function in a modulatory circuit receiving descending input from more rostral brain areas. Acknowledgements: NIH Grants DC000288 and DC009982

#P98 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Differential neural representation of oral ethanol by central taste-sensitive neurons in selectively bred ethanol-preferring (P) and Wistar rats

Christian H Lemon[1], David M Wilson[1], Susan M Brasser[2]
[1]St. Louis University School of Medicine St. Louis, MO, USA,
[2]San Diego State University San Diego, CA, USA

In randomly bred rats, orally applied ethanol stimulates neural substrates that process sweet taste. To study associations between ethanol's oral sensory signal and genetically mediated ethanol preference, we made electrophysiological recordings from taste-sensitive nucleus tractus solitarius neurons in anesthetized selectively bred ethanol-preferring (P) rats and Wistar (W) control rats. 50 P and 39 W cells were sampled. Stimuli (26 total) included ethanol (3, 5, 10, 15, 25 and 40%), sucrose (0.01, 0.03, 0.1, 0.3, 0.5 and 1 M) and other sweet, salt, acidic and bitter stimuli. *k*-means ($k = 2$) applied to sucrose responses identified cells showing high (S_1) or relatively low (S_0) responses (spikes) to sucrose. A 3-way interaction was found among a neuron's sensitivity to sucrose, its response to ethanol and rat line ($P = 0.01$). Ethanol produced concentration-dependent responses in S_1 neurons that were larger than those in S_0 cells (P's $<10^{-5}$). Although ethanol responses by S_1 cells did not differ between line ($P > 0.05$), S_0 cells showed larger responses to ethanol in the P line ($P < 0.01$). Compared with prototype stimuli ([in M] 0.5 sucrose, 0.1 NaCl, 0.01 HCl and 0.01 quinine), activity to 40% ethanol by W cells most robustly correlated with that to sucrose ($r = +0.73$). In P neurons, 40% ethanol evoked activity that was less correlated with sucrose ($r = +0.41$) than in W cells and most correlated with NaCl ($r = +0.58$). Multidimensional scaling of neural response patterns and magnitudes to all stimuli showed that in W neurons responses to ethanol and sweet stimuli were selectively similar, but in P neurons activity to ethanol was not easily related to any existing taste quality class. Ethanol evokes a unique oral sensory signal in P rats genetically selected to prefer alcohol. Acknowledgements: AA015741 (SMB) and AA015512 (IARC)

#P99 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Anti-lick Cells in the Nucleus of the Solitary Tract of the Freely-licking Rat

Andre T. Roussin[1], Jonathan D. Victor[2], Patricia M. Di Lorenzo[1]
[1]Binghamton University, Psychology Binghamton, NY, USA,
[2]Weill Cornell Medical College, Neurology and Neuroscience New York, NY, USA

It is well known that many cells in the rostral nucleus of the solitary tract (NTS) in anesthetized rats respond to taste stimuli bathed over the tongue. However, most investigators also find cells that do not respond to taste stimuli alongside taste-responsive cells. The function of these non-taste-responsive cells is unknown. In the present study, we recorded from the NTS of awake rats via a chronically-implanted bundle of microwires. Electrophysiological responses to taste stimuli were then recorded while the rats were actively licking, unrestrained, in an experimental chamber. In addition to previously reported taste-responsive cells, we also recorded from many cells (n = 20 thus far) in the NTS that conspicuously ceased firing when the animal was licking. More detailed analyses of these "anti-lick" cells revealed that most of these cells show an increase in firing rate that peaks just before the initiation of a lick bout (defined as a series of licks with inter-lick intervals <1 sec) and then drops precipitously when the lick bout is launched. In some cases the firing rate of the anti-lick cell trails off during the first couple of licks. During the lick bout, these cells either do not fire at all or fire only intermittently. Just after the lick bout ends, there is often an abrupt surge in the firing rate before the cell resumes a relatively rapid inter-bout firing pattern. These pre- and post-bout surges in firing rate suggest that the activity of these cells contains a signal that a lick bout is about to begin or end. On a population level, the cessation of firing of the anti-lick cells as the taste-responsive cells enter a phase of sensory acquisition while the rat is licking suggests the existence of a reciprocal relationship between taste-responsive and anti-lick cells. Acknowledgements: Supported by NIDCD grant DC006914 to PMD.

#P100 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Electrical Stimulation of the Central Amygdala Activates Neurons in the Gustatory Brainstem and Alters Taste Reactivity Behaviors in Conscious Rats

Christopher A. Riley, Trevor W. Tobin, Michael S. King
Stetson University DeLand, FL, USA

A role for the central amygdala (CeA) in the control of taste reactivity (TR) behaviors was investigated by electrically stimulating the CeA in conscious rats while monitoring TR behaviors and then mapping active brainstem neurons using Fos immunoreactivity (Fos-IR). Following surgery to implant an electrode into the CeA and intra-oral cannulas, and a recovery/adaptation period, 48 male Wistar rats were videotaped for 5 min during intra-oral infusion (0.233 ml/min) of either dH_2O (W), 0.1 M NaCl (N), 0.1 M sucrose (S), 0.03 M HCl (H), 0.003 M quinine HCl (Q) or nothing. In half of the rats in each group, the CeA was stimulated (40 Hz, 0.4 ms, 0.1-0.2 mA) during intra-oral infusion. One hour after behaviors were videotaped, the rats were sacrificed, perfused, and their brains removed, sectioned, and processed for Fos-IR. With no intra-oral infusion, CeA stimulation increased ingestive TR behaviors (23x) and increased the number of Fos-IR neurons (2x) in the waist area of the parabrachial nucleus (PBN, $p<0.05$). During infusion of Q, CeA stimulation caused a doubling of both ingestive TR behaviors and Fos-IR neurons in the waist area of the PBN ($p<0.05$). Stimulation of the CeA during intra-oral infusion of N increased aversive TR behaviors (20x, $p<0.05$) with a concomitant increase in the number of Fos-IR neurons in the rNST (2.1x), the PBN (12x), and the reticular formation (RF, 2.3x). Although CeA stimulation during intra-oral infusion of W, S, and H did not change TR behaviors, there were increases in the number of Fos-IR neurons in the rNST (for S) and the PBN and RF (for W). These data suggest that descending projections from the CeA alter TR behaviors by influencing the processing of gustatory input within the rNST and PBN as well as the generation of oromotor output in the reticular formation. Acknowledgements: Supported by NIH grant DC007854 to M.S.K. and a grant from Stetson's Summer Undergraduate Research Experience Program to C.A.R

#P101 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Effects of BLA and VTA stimulation on gustatory cortical dynamics

Haixin Liu[1,2], Alfredo Fontanini[1]
[1]Department of Neurobiology & Behavior, SUNY Stony Brook Stony Brook, NY, USA, [2]Program in Neuroscience, SUNY Stony Brook Stony Brook, NY, USA

The gustatory cortex (GC) is embedded in a network of areas involved in the processing of emotions and rewards. Anatomical evidence shows that GC receives direct projections from both basolateral amygdala (BLA) and ventral tegmental area (VTA). These inputs are believed to be responsible for transferring to GC information related to hedonic value, salience, expectation and motivation. However, how BLA and VTA influence spontaneous firing activity and enrich taste coding in GC neurons is not understood. Here, we show the effects of BLA and VTA stimulation on GC spontaneous activity and on taste processing in freely moving rats. Rats were implanted with movable bundles of recording wires in GC, unipolar stimulating electrodes in BLA and VTA, and intraoral cannulae for taste deliveries. Electrical stimulation (single 0.1Hz or train 200 Hz, 50 msec; 0.1 msec biphasic, 300uA ~ 500uA) to each of the two areas was tested alone or paired with passive taste stimulation (dT= 0 or -500msec). We found that BLA and VTA stimulation evoked a combination of excitatory and/or inhibitory effects in different cells with different time courses. Further, data were obtained comparing the effects of electrical stimulation on taste processing with coding of self-administered and passively delivered tastes. Preliminary analysis of these results shows that the effects on the time course of taste responses vary depending on the timing difference between electrical stimulation and gustatory stimulation. All together, our data provide a description of the dynamic role of reward-processing areas in modulating gustatory temporal codes. Acknowledgements: NIDCD Grant R01-DC010389 and Klingenstein Foundation Fellowship

#P102 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Effects of gustatory thalamic inactivation on processing of bottom-up signals in gustatory cortex

Chad L Samuelsen, Alfredo Fontanini
SUNY Stony Brook Department of Neurobiology and Behavior Stony Brook, NY, USA

Taste processing requires the integration of gustatory information from limbic, cortical and thalamic networks. This network of reciprocally connected areas begins in the central nervous system at the level of the brain stem in the nucleus of the solitary tract, which sends ipsilateral projections to the parabrachial nucleus (PBN). From PBN, taste information diverges along two routes: (1) a ventral forebrain, primarily amygdala path and (2) a thalamo-cortical pathway via the parvicellular portion of ventroposteromedial nucleus of the thalamus (i.e. gustatory thalamus - GTh). While these pathways have been well-described anatomically, the contribution of each of them to gustatory cortical coding is currently unknown. As GTh is a primary region involved taste processing, having both feedforward and feedback connections with PBN and GC, we hypothesize its role in conveying information about the chemical identity of tastes. Head restrained behaving rats, implanted with movable electrode bundles and intraoral cannulae (IOC), were passively delivered tastes while neural ensembles in GC were recorded both prior to and immediately following inactivation of GTh with the GABA agonist Muscimol. Temporary inactivation allows for the comparison of GC neuronal ensembles just prior and following GTh disruption. Preliminary data suggest that inactivation of GTh, modulates baseline firing rates, alters specific taste responses and uncovers novel taste responses. These preliminary data will be interpreted in relation to the effects of amygdalar inactivation and suggests that effective amygdalar and thalamic input is required for cortical processing of sensory quality of passively delivered tastes. Acknowledgements: NIDCD Grant R01-DC010389 and Klingenstein Foundation Fellowship

#P103 **POSTER SESSION II: TRIGEMINAL AND CHEMESTHESIS; CHEMICAL SIGNALS AND ANIMAL BEHAVIOR; TASTE: CNS**

Specific Expectation Modulates Taste Coding in Gustatory Cortex

Matthew P.H. Gardner[1,2], Alfredo Fontanini[1,2]
[1]Stony Brook University Department of Neurobiology and Behavior Stony Brook, NY, USA, [2]Stony Brook University Program in Neuroscience Stony Brook, NY, USA

We recently found that expectation of the availability of tastes alters stimulus processing in gustatory cortex (GC). Specifically, our results indicated that expectation accelerates taste coding by more than 100 ms and that this effect results from GC activation by a general anticipatory cue. Here we extend those studies by investigating how expectation of a specific gustatory stimulus alters activity and taste processing in GC. Rats were trained on an auditory go/no-go self-administration task in which they had to associate an auditory cue with sucrose and another cue to quinine. Multi-electrode bundles were chronically implanted to record from GC and from the basolateral amygdala, an area projecting to GC and known to be involved in processing of anticipatory cues. Preliminary findings show that GC neurons can in fact selectively respond to either one of the taste-specific cues and that these responses can disappear upon extinction. To address how these cue-induced responses affect subsequent taste processing, we compared responses to expected tastes with responses to unexpected or erroneously cued tastes. In erroneously cued trials quinine was self-delivered following a sucrose anticipating tone. Within these quinine "catch" trials, a subset of neurons show initial quinine taste processing more similar to sucrose deliveries than to quinine deliveries. These findings suggest that specific expectation may elicit taste-specific coding in GC prior to the actual taste delivery and that this anticipatory processing might play an important role in taste processing. Acknowledgements: NIDCD Grant R01-DC010389 and Klingenstein Foundation Fellowship

#P104 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Is the olfactory bulb developing like the ROB (rest of brain)?

Willi Bennegger[1,2], Elke Weiler[1]
[1]Faculty of Medicine, Institute of Physiology, Department of Neurophysiology, Ruhr-University, Universitaetsstr. 150 44801 Bochum, Germany, [2]Maria-von-Linden-Schule, Heckentalstr. 86 89518 Heidenheim, Germany

The olfactory bulb is a phylogenetical old structure, comprising originally a major part of the brain, and decreasing proportionally during evolution. Ontogeny usually resembles phylogeny, so we were interested, if the portion of the olfactory bulb (OB) decreases during development. Therefore we investigated the OB during postnatal development in the American mink (Neovison vison var. atratus), a species born very altricial, eyes and ears closed, with a body weight less than the adult brain weight, thus a major part of development occurs postnatally. A total of 65 males ranging from newborn (postnatal day 0, P0) to 1.5 years were morphometrically analyzed. The volume of one olfactory bulb in newborns is 1.85 ± 0.04 mm^3, increasing continuously (P15-30: 42.27 ± 3.01 mm^3; P60-90: 94.64 ± 3.84 mm^3) more than 80-fold to adult values (152.00 ± 9.14 mm^3). In contrast, the brain weight increases postnatally not even 40-fold from P0 (0.33 ± 0.06 g) up to only P70, when maximal values of 13.01 ± 0.95 g are reached, and decreasing afterwards (adults: 11.30 ± 0.52 g). Thus, the proportion of both olfactory bulbs on the total brain (bulb/brain ratio) increases postnatally dramatically from an initial value of 1.08 ± 0.09 % in newborns, to about 1.6 % during brain overshoot and further to 2.64 ± 0.11 % in adults ($p<0.001$). Thus the OB shows a different developmental pattern compared to ROB, with no neuronal overshoot however with a continuous increase in absolute size and of portion on the total brain, indicating the increasing importance during postnatal life (in newborns: nutrition and social odors; in juveniles: additionally prey and predator odors; in adults: rivals, area marking and sexual cues) requiring increasing information processing and structure, resulting in a gain of olfactory function. Acknowledgements: DFG (SFB 509 /TP C4) FORUM F208/00 M122/13 (2000)

#P105 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Multiple differentiation pathways specify mouse olfactory bulb dopaminergic neurons

John W Cave[1,2], Kasturi Bakerjee[2], Harriet Baker[1,2]
[1]Weill Cornell Medical College New York, NY, USA, [2]Burke Medical Research Institute White Plains, NY, USA

Olfactory bulb (OB) interneurons exhibit a diverse number of neurochemical phenotypes that are essential for processing and transmitting olfactory sensory information, but the molecular and genetic mechanisms that specify the differentiation of these phenotypes are not determined. We have examined co-expression of tyrosine hydroxylase (TH), the rate limiting enzyme in dopamine biosynthesis, and the transcription factor ER81, which our previous studies showed is important for TH expression in the OB. Based on size, dopaminergic OB interneurons have two distinct sub-populations. This study found that loss of functional ER81 disproportionately reduces that number of small TH-expressing cells in the OB. Based on these findings, we anticipated that the larger dopaminergic neurons would not co-express TH and ER81 in the wild-type OB. However, our analysis revealed that the TH-expressing cells lacking ER81 were a sub-population of the smaller sized neurons. Together, these findings suggest that there are at least three distinct subsets of OB dopaminergic neurons: 1) small neurons that co-express TH and ER81 that are lost in ER81 mutant mice; 2) small neurons that do not co-express ER81 and remain in the OB of ER81 mutant mice; 3) large neurons that co-express TH and ER81 and also remain in ER81 mutant mice. These findings suggest that multiple differentiation pathways specify OB dopaminergic neurons. We suggest that these pathways are regulated by partially overlapping combinatorial codes of transcription factor proteins that are a reflection of the distinct temporal and spatial origins for each subset of OB dopaminergic neurons. Acknowledgements: NIH DC008955

#P106 POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR

Age-Related Changes in Dopamine Receptor Expression in Olfactory Cortical Areas

Kurt R. Illig, Anna K. Zimmerman
University of St. Thomas Biology Department and Program in Neuroscience Saint Paul, MN, USA

Anatomical and physiological evidence suggests that dopamine (DA) input to higher order olfactory structures may play an important role in associating environmental stimuli with behavioral responses. Previous studies in rats have shown that the orbitofrontal cortex (OFC), piriform cortex (PC) and amygdala may form a network that is responsible for such associations. Moreover, involvement of this circuitry may contribute to age-related differences in learning olfactory-guided tasks, and in differences in susceptibility to addiction following exposure to stimulant drugs that affect DA. However, the development of DAergic circuitry in the OFC and PC remains largely unexplored. In this study, we examined the density, structure and organization of immunohistochemical labeling for tyrosine hydroxylase (TH), DA D_1 and D_2 receptors within each of these areas in young, adolescent and aged rats. Subregions of cortex displayed unique patterns of labeling, and several differences could be observed across the lifespan. For example, TH-positive fibers in old rats exhibited a more uniform morphology and higher degree of organization in the OFC than in young rats, especially in the lateral orbital cortex. In addition, age-related changes in the number and appearance of DA receptor subtypes were found. The patterns of age-related differences in DAergic components of higher-order olfactory areas suggest that the circuitry remains malleable into adulthood. Such late plasticity in DAergic innervation may underlie age-related differences in olfactory-guided learning. Further, these results raise the possibility that changes in DA in the OFC, amygdala and piriform cortex may contribute to age-related differences in susceptibility to drug addiction. Acknowledgements: AKZ was supported by a Collaborative Inquiry Grant and a Young Scholars Grant from the University of St. Thomas

#P107 POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR

Odor-induced plasticity in the olfactory bulb of adult mice

Nicolas Busquet, Josephine Todrank, Giora Heth, Diego Restrepo
University of Colorado Denver, CO, USA

Glomeruli in the olfactory bulb are formed by the coalescence of axons from olfactory sensory neurons (OSNs) expressing the same receptor in the olfactory epithelium. Glomeruli vary in size depending on the number of OSNs projecting to them.
A previous study in our laboratory has shown dramatic effects of odor exposure *in utero* and/or during nursing on glomerular size in juvenile mice (P21). The increase in size of the activated glomeruli was accompanied by an induced preference for the familiar odor. Shaping the sensory system in response to the olfactory environment could have evolutionary benefits, such as heightened sensitivity to odors of available, palatable foods. In this study we explored the effects of odor exposure on glomerular size on genetically modified adult mice (P63) co-expressing GFP in the OSNs with the M71 receptor. Mice were exposed through their diet to the target odorant, acetophenone, at different moments (*in utero*, during nursing, after weaning or 3 weeks before perfusion) and for different lengths of time (from continuous exposure to never). Measuring the volume of GFP-tagged glomeruli (estimated from areas of serial 20 µm sections) revealed significant effects of odor exposure on glomerular size. This large-scale study also revealed interesting differences between males and females, and between lateral and medial glomeruli. These results suggest that olfactory perception is actively shaped by olfactory experience, throughout development and into adulthood.

#P108 POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR

Physiological Changes in Olfactory Sensory Function Induced by Unilateral Olfactory Deprivation in the Adult Mouse

Marley D. Kass, Daniel J. Turkel, Joseph Pottackal, Tom Rubinstein, John P. McGann
Rutgers University Psychology Department New Brunswick, NJ, USA

Olfactory sensory deprivation has been shown to induce alterations in the synaptic physiology of olfactory receptor neurons in the developing brain (Tyler et al. *J Neurosci* 2007). While deprivation has been shown to alter the neurochemistry of the adult olfactory bulb, the physiological consequences are poorly understood. Here we used optical imaging techniques to visualize odorant-evoked transmitter release in adult mice that underwent unilateral olfactory deprivation via removable noseplugs. Mice expressing synaptopHluorin (spH) from the OMP locus (Bozza et al. *Neuron* 2004) were deprived for 2, 3, or 4 weeks. Deprivation efficacy was demonstrated by a decrease in periglomerular tyrosine hydroxylase in the olfactory bulb ipsilateral to the occlusion. Twenty-four hours after plug removal spH signals were recorded bilaterally from the dorsal olfactory bulbs in response to presentations of a panel of 4 odorants at up to 3 concentrations. In control mice, responses to these odorants are symmetrical between left and right olfactory bulbs. However, in deprived mice odorant-evoked transmitter release from the olfactory nerve was observed in significantly fewer glomeruli in the olfactory bulb ipsilateral to the reopened naris compared to the control naris. This was not a consequence of reduced airflow on the reopened side because blocking the control naris to force all airflow through the reopened side did not change the response. We did not observe a significant change in the odorant-selectivity of the glomerular responses that were observed on the reopened side, but even at saturating concentrations of odorant the number of glomeruli responding on the reopened side never equaled that on the control side. These results suggest that sensory environment can strongly influence primary olfactory processing. Acknowledgements: This work was funded by grant number DC009442 from NIDCD to JPM.

#P109 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Chronic odorant exposure can alter olfactory sensory neuron function in adult mice *in vivo*

Andrew H. Moberly, Marley D. Kass, John P. McGann
Rutgers University Psychology Department Piscataway, NJ, USA

Chronic exposure to odorants has been shown to alter the neurophysiology of the olfactory epithelium and bulb, but the functional effects of these changes are poorly understood. To explore this plasticity, we used within-subjects optical methods to visualize odorant-evoked neurotransmitter release from the olfactory nerve before and after a one week exposure to an odorized environment. Mice expressing the fluorescent exocytosis indicator synaptopHluorin from the OMP locus (Bozza et al. *Neuron* 2004) had a cranial window implanted overlying the olfactory bulbs and underwent an initial imaging session in which spH signals were recorded bilaterally from the dorsal olfactory bulbs in response to presentations of a panel of four odorants. Mice then spent the next week in either their open shoebox-style home cage or a "den-like" exposure chamber with reduced airflow compared to the home cage. Mice in the exposure chambers were exposed to their own odorant plus methyl valerate (MV), butyl acetate (BA), or vehicle control added to the airflow (4 h duty cycle). In mice that spent the exposure week in their home cage, we found no significant changes in the number of glomeruli receiving odorant-evoked synaptic input from the olfactory nerve or the amplitude of that input. In mice that spent a week in the exposure chamber, the amplitude of glomerular input in response to non-exposed odorants was significantly reduced in all three groups. The number of glomeruli receiving input was significantly reduced in the two groups that were exposed to MV or BA. For the two groups in which an odorant was explicitly added to the exposure chamber, we observed equivalent suppression of responses to the exposed odorant and non-exposed odorants. The results suggest global regulation of primary olfactory responses. Acknowledgements: Funded by grant number DC009442 from NIDCD.

#P110 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Role of functional expression of TRPM5 in maintaining the survival of canonical olfactory sensory neurons expressing nonfunctional CNGA2

David A Dunston, Wangmei Luo, Muna Merdato, Weihong Lin
University of Maryland, Baltimore County Baltimore, MD, USA

Most olfactory sensory neurons (OSNs) utilize the cyclic nucleotide gated channel subunit A2 (CNGA2) in signal transduction. Due to X inactivation of CNGA2, female mice heterozygous for the CNGA2 knockout (KO) make two distinct OSN populations with one population expressing nonfunctional CNGA2 and GFP (CNGA2 KO GFP+/-) (Zhao and Reed 2001). Our previous work showed that surviving OSNs with nonfunctional CNGA2 can be mature and are located in the same area as transient receptor potential channel M5 (TRPM5) expressing OSNs (Dunston et al AChems 2010, Lin et al 2007). We examined the importance of odor enrichment and functional expression of TRPM5 for the survival of OSNs with nonfunctional CNGA2 with or without TRPM5 KO (CNGA2 KO GFP +/- / TRPM5 KO) and CNGA2 KO GFP +/- respectively. Odor enriched mice were group housed and had soiled bedding from a mating pair periodically added, and odor non-enriched mice were singly housed without additional odor stimulation. We monitored changes in randomly selected glomeruli in the olfactory bulb, particularly GFP signals. CNGA2 KO GFP +/- / TRPM5 KO mice had fewer strong GFP glomeruli than CNGA2 KO GFP +/- mice in odor enrichment groups, indicating functional TRPM5 promotes the survival of OSNs with nonfunctional CNGA2. The number of strong GFP glomeruli in CNGA2 KO GFP +/- mice was lower in odor enriched than odor non-enriched groups, indicating odor enrichment decreases the survival of OSNs with nonfunctional CNGA2. Soiled bedding activated some strong GFP glomeruli, as indicated by expression of c-fos, a neuronal activation marker in periglomerular cells. The PLC pathway that utilizes TRPM5 is a likely candidate to activate OSNs with nonfunctional CNGA2, thereby prolonging their survival.

#P111 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Development and activity dependence of the interglomerular circuit

Celine Plachez, Stephanie Parrish-Aungst, Emi Kiyokage, Michael T. Shipley, Adam C. Puche
Department of Anatomy and Neurobiology, University of Maryland, School of Medicine Baltimore, MD, USA

Sensory experience is critical for brain development and function. A striking example is in the main olfactory bulb (MOB) where reduction of odorant sensory signals profoundly down-regulates dopamine (DA) in glomerular neurons. MOB DA neurons co-express glutamic acid decarboxylase 67 (GAD67) and thus are dopaminergic and GABAergic. These DA-GABAergic neurons are short axon (SA) cells that form an extensive interglomerular circuit. ZnSO4 lesion and nares occlusion, which eliminate/reduce sensory input, reduced both tyrosine hydroxylase (TH) and GAD67 in SA cells. By contrast, GAD65 expression in periglomerular neurons was independent of sensory activity. Thus, sensory input regulates the enzymes responsible for neurotransmitter synthesis by SA but not PG cells. In some circuits activity also determines neural connectivity. We asked if sensory deprivation influences interglomerular connections by mapping the projections of SA cells labeled by small glomerular DiI injections following ZnSO4 lesion or nares occlusion. At 4 months post lesion/occlusion SA cells project approximately half the distances observed in normal MOB. Thus, sensory input regulates SA cell connections as well as their neurotransmitter enzymes. Activity-dependence in adult mice suggested that interglomerular connections might also be influenced by sensory experience during development. To test this we analyzed the distribution of cells -labeled DiI at postnatal days 0, 4, 8, and 15. At birth, interglomerular projections are short spanning only several glomeruli. With increasing age SA cells project greater

distances up to their adult patterns. Thus the formation and maintenance of the circuitry that regulates interglomerular neural processing is determined by sensory experience. Acknowledgements: NIH DCCD005676, DCCD19015

#P112 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Sensory preconditioning leads to separate odor memories in newborn rabbits

Gérard Coureaud[1], Audrey Tourat[1], Guillaume Ferreira[2]
[1]CSGA, UMR 6265 CNRS/INRA/Université de Bourgogne/Agrosup Dijon Dijon, France, [2]Laboratoire NutriNeuro, UMR 1286 INRA/Université Bordeaux 2 Bordeaux, France

In newborn rabbits, a biological odor signal emitted by all lactating females, the mammary pheromone (MP), triggers a stereotyped behaviour allowing the rapid localization and oral seizing of the nipples. The MP acts also as a reinforcer allowing the efficient acquisition of novel odorants. Here, we evaluated whether a MP-learned odorant (A) becomes by itself able to promote the acquisition of another odorant (B) and whether memory of odorant B depends on associative chain with A (BàA) or is independent on odorant A. For this purpose we used a chain-like associative paradigm, the sensory preconditioning, in which the neutral pairing of A and B (exposure to AB) was followed by the reinforcement of A and a final test of behavioral response to B. We first showed that after unreinforced exposure to AB on postnatal day 1 (d1), the MP-acquisition of odorant A on d2 led to similar responses to odorants B, A and to the MP on d3. Control experiments confirmed the associative nature and selectivity of the preconditioning: simultaneous, but not sequential exposure to AB on d1, or A-MP on d2, supported the acquisition of the odorants, and exposure to AB then CD mixtures on d1 followed by MP-A induced learning on d2, led to responses to A and B but not to C and D. Next, following A-B and A-MP associations on d1 and d2, respectively, amnesic treatment after reactivation of A on d3 abolished the response to A on d4, but left the response to B intact. The reverse was also true: amnesia of B on d3 did not disrupt the souvenir of A on d4. Our results indicate that chemosensory preconditioning functions in the neonatal brain and that, rapidly after memory formation based on chain-like associations, the maintenance of neonatal odor memories relies on an independent more than a sequential organization. Acknowledgements: Supported by MEMOLAP grant from Agence Nationale de la Recherche (ANR) 2010-JCJC.

#P113 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Aversive olfactory conditioning influences neurogenesis in the adult mouse olfactory bulb

Florence Kermen[1,2], Joëlle Sacquet[1,2], Nathalie Mandairon[1,2], Anne Didier[1,2]
[1]Lyon University Lyon, France, [2]Centre National de Recherche Scientifique Lyon, France

The olfactory information coming from the sensory neurons in the nasal cavity is processed in the olfactory bulb (OB). This structure undergoes a constant renewal of its interneurons during adult life, which function remains unknown. Previous works showed that an associative olfactory conditioning, during which mice learned to associate a positive reward to an odorant, increases survival of newly formed neurons in the OB. However, it is unknown whether these changes in neurogenesis contribute to the encoding of the acquired positive significance of the odor. To address this issue, we currently investigate the influence of an appetitive versus an aversive olfactory learning on bulbar neurogenesis. Adult mice learn during 5 days to associate an odor (+Limonene or –Carvone) to a sweet or a bitter bit of cereal. A week after conditioning, mice are tested for long term retention of the association and euthanized to assess survival and spatial distribution of new neurons in the granule cell layer of the OB. Preliminary data suggest that both appetitive and aversive learning elicit an increase in neurogenesis in the OB. However, the spatial distribution of new neurons in the granule cell layer of the OB is different depending on the hedonic value acquired by the odor. Thus, this would suggest that neuronal bulbar turnover could contribute to the encoding of odor hedonic value.

#P114 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

NT4 is More Potent than BDNF at Promoting, Attracting, and Suppressing Embryonic Geniculate Neurite Outgrowth

Mathew J Biehl, Natalia Hoshino, Son Ton, M William Rochlin
Loyola U Chicago, Biology Chicago, IL, USA

NT4 and BDNF are TrkB ligands that support the survival of geniculate ganglion (GG) neurons and may be involved in innervation of fungiform papillae. Curiously, whereas overexpression of NT4 in the lingual epithelium stifles chorda tympani branching and innervation of the epithelium; overexpression of BDNF enhances branching and undirected exploration of the epithelium (Lopez and Krimm, Dev. Biol, 292:457). To gain insights into the basis of these differences, we characterized the influence of NT4 and BDNF on GG neurites in collagen I gels. NT4 is ~20 fold more potent than BDNF and both factors exhibit concentration optima, i.e., intermediate concentrations promote maximal neurite extension (0.25-1 ng/ml, NT4; 10-50 ng/ml, BDNF) and high concentrations suppress it (10 ng/ml, NT4; 200 ng/ml, BDNF). At high concentrations, outgrowth was nearly eliminated with E13-E18 GG, but little suppression was observed with E12 or postnatal GG. At targeting stages (E15-18) the concentration of NT4 that fully suppresses neurite extension is near the threshold concentration for

stimulating neurite outgrowth by BDNF. This may explain, in part, the different phenotypes observed in overexpression studies in vivo. BDNF is implicated as a chemoattractant for GG axons during targeting. Using beads soaked in low concentrations of NT4, we find that it too chemoattracts GG neurites at targeting stages. At E12-13, NT4 and BDNF promote significantly longer outgrowth than at later embryonic stages. Chemoattraction by NT4 soaked beads is evident as early as E13, but not at E12; whereas BDNF chemoattracts from E12-E18. NT4 and BDNF exert a trophic, but not tropic effect on postnatal GG neurites. The mechanisms underlying the potency difference of NT4 and BDNF and the stage dependence of the responses remain to be determined. Acknowledgements: Supported by NIH 1 R15 DC009043-01.

#P115 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Expression of *Bdnf* is downregulated in the gustatory system during postnatal mouse development

Tao Huang, Robin F. Krimm
Dept. Anatomical Sciences and Neurobiology, University of Louisville, School of Medicine Louisville, KY, USA

Neurotrophins have multiple functions during gustatory development such as controlling taste bud development and maintenance, neuronal survival and gustatory innervation. During embryonic development, expression patterns of BDNF and NT4 in taste system correlate with their functions. However, taste bud maturation and taste function are not completed until postnatal development. To determine if neurotrophins are involved in these processes, time-course expression of *Bdnf, Nt4* and *TrkB* in geniculate ganglion and fungiform taste buds were examined at birth, postnatal day 10 (P10), P20 and P60, using β-gal staining in BDNF-LacZ mice and real-time RT-PCR. In the geniculate ganglion, *Bdnf* expression decreased from birth to P10 by 52% ($p<0.01$), and the lower expression levels of *Bdnf* were maintained through P60. The expression of TrkB, receptor for BDNF, was not different from birth to adult. In fungiform papillae, the distribution of BDNF was not limited to taste buds. *Bdnf* expressed in gustatory tissue was greatly reduced (83%) after P10, compared with that at birth ($p<0.05$), but its expression in non-gustatory lingual epithelium was not different across postnatal ages. There was very little (if any) *TrkB* and *Nt4* expression in the postnatal fungiform papillae. Thus, *Bdnf* expression is downregulated in both fungiform taste buds and geniculate ganglion from birth to P10. As taste buds undergo proliferation, differentiation and functional maturation after birth and gustatory innervation patterns are maturing, BDNF within postnatal taste buds may influence these developmental events. Similarly, other postnatal events like NTS terminal field development and the relationship between taste bud size and the number of innervating neurons may be regulated by BDNF expressed in the postnatal gustatory system. Acknowledgements: NIH DC007176

#P116 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Detection of brain derived neurotrophic factor in the gustatory system after chorda tympani nerve transection

Lingbin Meng[1], Chengsan Sun[1], Tao Huang[1], David L Hill[1], Robin Krimm[1]
[1]*University of Louisville Louisville, KY, USA,* [2]*University of Virginia, Virginia, VA, USA*

Brain derived neurotrophic factor (BDNF) is required for the survival and maintenance of taste buds and gustatory nerve fibers during development. Therefore, BDNF may also regulate nerve degeneration or regeneration in gustatory system. In this experiment, we examined BDNF expression using rt-PCR in the adult lingual epithelium and geniculate ganglion from the cut and uncut sides, 2 days and 2, 4, and 8 weeks following unilateral chorda tympani nerve section of adult mice. BDNF is normally expressed in taste buds of the lingual epithelium and therefore could be reduced or lost as taste buds degenerate following nerve section. However, we found that BDNF does not change expression levels in either the lingual epithelium or the geniculate ganglion after nerve cut. This indicates that BDNF is maintained in the ganglion and the lingual epithelium after nerve cut and could influence nerve regeneration. Consistent with a loss of taste buds, expression of keratin 8 (which is the marker for taste buds) is reduced 2 weeks and one month following nerve section on the cut side but not the uncut side ($p<0.01$). Surprisingly, BDNF increases in the geniculate ganglion of uncut side two weeks following nerve section ($p<0.01$). Lastly, we found that expression levels for BDNF are much higher than for keratin 8 in the lingual epithelium. One possible explanation is that BDNF expression is not limited to the taste bud following nerve section. Consistent with this possibility, preliminary data indicate that in control mice, epithelium from taste buds, fungiform papillae, and filiform papilla each express BDNF as measured with rt-PCR. We are currently examining BDNF expression separately in each of these regions following nerve cut using rt-PCR and in BDNF-lacZ mice. Acknowledgements: Supported by DC006938.

#P117 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Neuropilin-2 and Calbindin Expression in Developing Solitary Tract and Rostral Nucleus of Solitary Tract of Rat Embryonic Brainstem

Miwon Kim[1], Charlotte M Mistretta[1], Robert M Bradley[1,2]
[1]*Department of Biologic and Materials Sciences, School of Dentistry, University of Michigan Ann Arbor, MI, USA,* [2]*Department of Physiology, School of Medicine, University of Michigan Ann Arbor, MI, USA*

Taste receptors in the oropharynx and larynx send sensory information to the rostral nucleus of the solitary tract (rNST) via the facial, glossopharyngeal and vagus nerves. These nerves enter the brainstem to form the solitary tract (ST) and project to neurons in the rNST. The neuropilins, a family of transmembrane proteins, are important in development of the facial, glossopharyngeal and vagus nerves and have roles in axonal

growth and fasciculation. We therefore explored the role of neuropilin-2 (Npn-2) in formation of the ST in embryonic rats from E13 to E18 using immunohistochemistry. Antibody to calbindin was used to label rNST neurons. Npn-2 label in the early ST is already apparent at E13, in a narrow band of fibers running rostral-caudal, near the lateral border of the brainstem. At E14 large collateral, tuft-like collections of Npn-2 positive processes extend medially from the ST. By E16 the ST consists of a broad band of fascicles and medially directed collateral branches. Calbindin expression is apparent at E13 in a group of cells medial to the ST that has increased in numbers by E14. By E16 calbindin labeled cells were localized close to the IVth ventricle. At E18 calbindin-positive neurons were abundantly expressed medial to the mature ST. These preliminary results describe the developmental time course of ST projections and their relation to developing rNST neurons. Further, results identify Npn-2 as a potential molecular determinant involved in formation of the ST and rNST. Acknowledgements: SUPPORT: NIDCD, NIH Grant DC009982.

#P118 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Decreased terminal field volume in the mouse NTS after unilateral chorda tympani nerve cut

Chengsan Sun, David L. Hill
University of Virginia/Psychology Charlottesville, VA, USA

There has been relatively little attention devoted to the central consequences of gustatory nerve damage. Degenerating neuronal profiles were seen in the NTS post nerve cut in hamster; however, changes in terminal field volumes were not quantified. Our lab found large-scale decreases (~60%) in the terminal field volume of the chorda tympani nerve after nerve section (CTX) in rat. To better understand the cellular/molecular mechanisms underlying these injury-induced changes in terminal fields at adulthood, we chose to use the mouse as an experimental model because of the advantages in manipulating genetic products. Here, we establish that unilateral CTX at adulthood produces a significant loss of terminal field volume and density in a mouse strain often used as the background for genetic manipulations, the C57Bl/6J mouse. Following a unilateral CTX in the neck of mice, we labeled the chorda tympani nerve in the tympanic bulla with biotinylated dextran (3 kD) at various periods post CTX. We found no effects on the mean terminal field volumes from the intact, contralateral side. The terminal field volume of the chorda tympani nerve was significantly decreased in the NTS after 30 and 60 days post nerve section by 38% and 43%, respectively. To begin addressing candidate mechanisms that may underlie these events, the amount of Brain Derived Neurotrophic Factor (BDNF) mRNA in the NTS was also studied with quantitative real-time PCR. The BDNF mRNA in NTS ipsilateral to the CTX was different from the intact side at early stages post CTX (2-14 days post CTX). These effects in terminal field volume and BDNF provide us with a new and exciting direction to further examine the plasticity of the gustatory system in genetic modified adult mice induced by taste nerve damage. Acknowledgements: R01 DC00407 and R01DC006938

#P119 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Changes to Chorda Tympani Nerve Terminal Field Following Greater Superficial Petrosal and Glossopharyngeal Nerve Section and Regeneration

Sara L. Dudgeon, David L. Hill
University of Virginia Charlottesville, VA, USA

The rostral nucleus of the solitary tract contains overlapping terminal fields of three gustatory nerves: the chorda tympani (CT), the greater superficial petrosal (GSP) and the glossopharyngeal (IX) nerves. These three nerves all undergo a progressive decrease in terminal field volumes throughout postnatal development, leading to decreases in overlapping fields. Previously, we found that sectioning the GSP and IX nerves at any age, including adulthood, and thus removing their input to the NTS results in an expanded CT nerve terminal field prior to regeneration of the sectioned nerves. In fact, the expanded CT nerve terminal field is roughly four times larger than what we would expect to find in an adult control rat, and is similar in volume and distribution to the terminal field of an intact, immature animal (post-natal day 15). We believe this expansion is due the CT nerve no longer being subject to competitive influences from the GSP and IX nerves. In order to address this further, we explored changes to the CT nerve terminal field following regeneration of the GSP and IX nerves. Additionally, we examined the terminal field morphology of the regenerated nerves along with that of the CT nerve terminal field to understand the dynamic relationship between these three nerves. Preliminary data show that following the regeneration of the GSP and IX nerves the CT terminal field maintains its expanded volume compared to controls. However, the volume is less than that seen prior to GSP and IX regeneration. Acknowledgements: Supported by NIH Grants R01 DC00407 and R01 DC006938

#P120 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Impaired regeneration of taste buds in aged rats

Lianying He, Lynnette P McCluskey
Georgia Health Sciences University / Institute of Molecular Medicine and Genetics Augusta, GA, USA

Neural regeneration typically lags several days in senescent compared to young animals. Aging impacts the peripheral taste system more severely. Neural taste responses can be recorded from the regenerated chorda tympani nerve (CT) of young rats by 45 days after injury. In aged F344 rats, however, the regenerated CT fails to respond to tastants until at least day 85 post-sectioning, which is the end of the life span. We determined whether impaired regeneration of taste buds might contribute to functional deficits. Cytokeratin (CK) 19 immunopositive taste buds were analyzed 45-60 days after CT injury in three month old and two year old rats. The number of CK19+ taste buds on the regenerated side of the tongue was ~80% less in aged vs. young adult rats. Most of the taste buds that did regenerate were the same size in young and old animals. Our results indicate that

aging prevents taste buds from regenerating efficiently. Aged taste cell precursors may be particularly dependent upon trophic support or more vulnerable to trauma. Acknowledgements: NIH DC005811

#P121 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Postnatal *GLI2* activation suggests roles for Shh signaling in maintaining tongue filiform and fungiform papillae and taste buds

C.M. Mistretta[1], H.-X. Liu[1], M. Gratchtchouk[2], A.A. Dlugosz[2, 3]
[1]Department of Biologic and Materials Sciences, School of Dentistry, University of Michigan Ann Arbor, MI, USA, [2]Department of Dermatology, Medical School, University of Michigan Ann Arbor, MI, USA, [3]Department of Cell and Developmental Biology, Medical School, University of Michigan Ann Arbor, MI, USA

Sonic hedgehog (Shh) regulates taste papilla development. The gene encoding Gli2, the transcriptional activator in Shh signaling, is expressed throughout basal cells of tongue epithelium, whereas Shh is tightly restricted to embryonic apical fungiform papilla epithelium and postnatally, to taste bud cells. We are testing effects of *GLI2* activation to understand how Shh signaling affects sustained differentiation of postnatal tongue organs. Transgenic mice are used, with conditional activation of an activator form of *GLI2* in basal cells of skin and tongue epithelium. Tongues were analyzed from 2-3 month postnatal mice with doxycycline-regulated *GLI2* activation for 7-12 days, and from controls with no doxycycline. In *GLI2*-activated tongues, the basal cell marker K5 was seen in basal cells and in suprabasal layers. From Ki67 immunoreactions there was an observed increase in proliferating cells within the basal cell layer and also, proliferation was noted in suprabasal layers throughout lingual epithelium, suggesting an expansion of the basal cell compartment. Spines were lost from filiform papillae, where apical epithelial cells were proliferating, and the papillae had dedifferentiated in *GLI2*-activated tongues. Fungiform papillae were decreased by up to half of numbers in control tongue, thereby reducing taste buds also. In remaining fungiform papillae, small taste buds were present with cells that were positive for the taste cell marker, K8. Furthermore, sustained papillae and taste buds were innervated, demonstrated with neurofilament immunoreactions. Results from a conditional activation of *GLI2* for only 1-2 weeks demonstrate profound importance of balanced hedgehog signaling in postnatal tongue integrity and maintenance of lingual taste organs. Acknowledgements: NIH NIDCD Grant DC000456 (CMM) and NIAMS AR045973 (AAD)

#P122 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Distinct longevities of the cell types in adult taste buds

Isabel Perea-Martinez[1], Takatoshi Nagai[2], Stephen D Roper[1,3], Nirupa Chaudhari[1,3]
[1]Dept of Physiology & Biophysics, University of Miami Miller School of Medicine Miami, FL, USA, [2]Dept of Biology, Keio University School of Medicine Yokohama, Japan, [3]Program in Neurosciences, University of Miami Miller School of Medicine Miami, FL, USA

As with all epithelia, cells in taste buds are continually renewed throughout adult life. Previous estimates using ^{3}H-thymidine or BrdU labeling suggested that taste cells have an average longevity of 10-14 days. Our recent perspective on the distinctly different roles of Types I, II and III cells raises the question of whether these cell types are renewed at a similar rate. We have examined this question with a pulse of the thymidine analog, 5-ethynyl-2'-deoxyuridine (EdU) and detection by click chemistry. EdU is incorporated during DNA synthesis and progeny nuclei remain labeled through 1-2 mitoses. We birth-dated cells of lingual epithelium by injecting EdU into adult PLC 2-GFP transgenic mice and sacrificing them 1, 2, 3, 5, 7, 10, 15, 20, 25, 30, 35 or 40 days later. We immunostained vallate cryosections for GFP (Type II cells), 5HT (Type III cells), KCNQ1 (all mature cells of taste buds) and stained for EdU (cells born on day 0). EdU-labeled non-taste epithelial nuclei were detected within 1 day of injection and persisted <10 days. Using high-resolution confocal microscopy, we scored EdU+ nuclei in Type II or Type III taste bud cells. EdU-labeled nuclei in Type II cells appeared within 3 days post-injection and were rare by 25 days. In contrast, EdU-labeled Type III cells appeared at day 5, and their numbers remained relatively constant until day 40. EdU-labeled taste bud nuclei that were in non-Type II, non-Type III cells were prominent from day 1 and surprisingly, persisted for up to 30 days. This last category is primarily mature Type I cells, but may also include labeled cells that remain undifferentiated for an extended period, and subsequently give rise to one of the defined cell types. Our data suggest that the neuron-like synapse-forming cells of taste buds turn over the slowest. Acknowledgements: Supported by NIH/NIDCD grant R01DC006308 (NC).

#P123 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Solitary Chemosensory Cells Turnover in Tracheal Epithelium, in vivo and in vitro models

CJ Saunders[1], Susan D Reynolds[2], Thomas E Finger[1]
[1]Rocky Mtn Taste & Smell Ctr, Neurosci Prog, Univ Colo Denver Med Sch Aurora, CO, USA, [2]Dept of Pediatrics, National Jewish Health Denver, CO, USA

Solitary chemosensory cells (SCCs) in the epithelia of the nose and trachea utilize the canonical taste transduction pathway ("bitter" (T2R) receptors, G -gustducin and TRPM5), to detect a variety of substances including some bitter molecules. Morphologically, SCCs are flask shaped and resemble type II taste

receptor cells and pulmonary neuroendocrine cells (PNECs). Tracheal SCCs are, however, not immunoreactive for CGRP, a marker for PNECs. In the present study, we examined turnover of tracheal SCCs in vivo and in vitro. Tracheal epithelial cells were recovered from TRPM5-GFP mice and allowed to proliferate on Transwell membranes until confluent. In vitro, SCCs appear only after the removal of growth factors and establishment of an air-liquid interface (ALI day 0). During the first 14 days at ALI, the number of SCCs increased significantly reaching just over 1% of the total population. While the number of differentiated SCCs increased rapidly, the total density of cells in culture did not change significantly. Therefore SCC differentiation and maturation is a relatively late event in vitro. In keeping with this, BrdU pulses during the first 7 days at ALI labeled more SCCs then BrdU pulses before ALI indicating terminal differentiation of SCCs occurs only after ALI. To study SCC turnover *in vivo*, we injected TRPM5-GFP mice transgenic mice with 5-bromo-2'-deoxyuridine (BrdU) to label dividing cells. 10 days after injection, scattered BrdU labeled cells are present in the tracheal epithelium but no SCCs were labeled. Also, no SCCs were immunoreactive for Ki67, a marker of proliferative cells. These data are consistent with the hypothesis that once differentiated, the tracheal SCCs have a longer half-life than similar cells in the nasal cavity. Acknowledgements: Supported by NIDCD, NHLBI

#P124 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Potential contribution of P0-expressing neural crest derived cells to developing gustatory papillae and taste buds

H.-X. Liu, Y. Komatsu, Y. Mishina, C.M. Mistretta
Department of Biologic and Materials Sciences, School of Dentistry, University of Michigan Ann Arbor, MI, USA

Taste bud cells have been described as arising from the local tongue epithelium, in contrast to receptor cell types derived from neurogenic ectoderm including the neural tube, neural crest and ectodermal placodes. In continuing studies of neural crest cell contribution to taste organs, we labeled neural crest derived cells (NCDCs) using a P0-Cre mouse line that expresses Cre recombinase in a neural crest-specific manner in combination with Cre reporter mouse lines. The distribution of Cre-driven reporter labeled cells is progressively associated with taste papillae from embryonic (E) day 11.5 to postnatal (P) day 10. Labeled cells are throughout the mesenchymal core of papillae, in the basal lamina region underlying early taste buds, and in mesenchyme just under the lingual epithelium. Labeled cells also are seen in the epithelium, scattered separately at E11.5 -12.5 and in clusters within and between papillae at E13.5-16.5. At E18.5-P10, intensely labeled cells are observed in early taste buds. At P10, most taste buds contain labeled cells. No Cre immunoreactivity was observed in the Cre-driven reporter labeled cells at the stages examined (E11.5, P1), excluding the possibility of ectopic Cre activity labeling of epithelial and mesenchymal cells. The distribution of P0-Cre marked cells in taste papillae and early taste buds suggests a potential neural crest contribution to papilla epithelium and taste bud cells. Also, progressively dense distribution of P0-expressing NCDCs in the mesenchymal core of taste papillae and mesenchyme just under the lingual epithelium suggests important roles of NCDCs in cell interactions among epithelium, mesenchyme and taste buds. Together our data suggest important roles of NCDCs in the development of taste papillae and taste buds. Acknowledgements: NIDCD, NIH Grant DC009055 to HXL

#P125 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Transient expression of Oxytocin Receptor in the lineage of Glial-like cells of mouse taste buds

Gennady Dvoryanchikov[1], Nirupa Chaudhari[1,2]
[1]Department of Physiology and Biophysics, University of Miami Miller School of Medicine Miami, FL, USA, [2]Program in Neurosciences, University of Miami Miller School of Medicine Miami, FL, USA

Adult taste buds turnover continuously, and are replenished by cells from local lingual epithelium. Epithelial cells that express the transcription factor, Sox2, are located adjacent to taste buds, and generate both taste cells and keratinocytes. Do such Sox2-expressing cells produce all the component cell types of adult taste buds? Oxytocin Receptor (OXTR) is prominently expressed on the periphery of taste buds and in some Type I/Glial-like cells. Here, we explored whether the OXTR-expressing cells are immature Sox2-expressing taste cells. We visualized OXTR using knock-in OXTR-YFP mice and immunostained for Sox2 and markers of mature taste cell types. In vallate papillae, all OXTR-YFP positive cells inside and on the periphery of taste buds were found to be Sox2 positive (120 of 121 inside; 42 of 42 on the periphery). The same result was also apparent in palate, foliate and fungiform taste buds. Nearly all Sox2 nuclei (286 of 289) belong to non-Receptor (PLC 2+), non-Presynaptic (5HT+) cells. Because ~ half of all nuclei inside taste buds are Sox2-positive and Type I cells constitute ~ half of taste bud cells, our results suggest that the Sox2-expressing bipotential progenitors of the lingual epithelium principally give rise to Type I cells, not Type II or III. We confirmed these observations using an independent method, single-cell RT-PCR. Interestingly, Keratin 8, which is used as a marker of mature taste cells, is prominent in Type II and III cells; Sox2-positive cells and OXTR-YFP cells were poorly or not immunoreactive for Keratin 8. Taken together, our data indicate that Sox2 and OXTR expressing cells generate Type I/Glial-like cells. We cannot exclude the alternative possibility that Sox2 is rapidly downregulated, becoming undetectable as cells commit to Type II or III differentiation. Acknowledgements: Supported by NIH/NIDCD R01DC6308 (NC)

#P126 POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR

Dependence of intra-nasal odorant concentrations on sniff behaviour

Jonathan Beauchamp[1], Mandy Scheibe[2], Thomas Hummel[2], Andrea Buettner[1,3]
[1]Sensory Analytics, Fraunhofer Institute for Process Engineering and Packaging (IVV) Freising, Germany, [2]Department of Otorhinolaryngology, University of Dresden Dresden, Germany, [3]Department of Chemistry and Pharmacy - Emil Fischer Center, University of Erlangen-Nuremberg Nuremberg, Germany

It has been known for the last three decades that optimum odour perception is achieved with just a single sniff, as was demonstrated by the early experiments of Laing. Thus, the act of sniffing is itself an intrinsic aspect of the olfactory percept. Nevertheless, despite the importance attributed to this instinctive and frequently used mechanism, there is still a great deal of uncertainty as to what is actually happening to odorant molecules inside the nose when sniffing is being performed. To provide a first assessment of the relationship between sniffing and odorant delivery to the olfactory epithelium, we have performed direct intra-nasal odorant concentration measurements at the nostril and olfactory cleft using proton-transfer-reaction mass spectrometry (PTR-MS) during different sniffing procedures. In particular, absolute intra-nasal odorant intensities were monitored in real-time according to inhalation performance of subjects. In this feasibility study using 2,3-butanedione as the target odour molecule it could be shown that odorant concentrations at the olfactory cleft are indeed highest during a single sniff, and lowest when inspiration is rapid or forced. These analytical data were corroborated by the perceived intensities of individual panellists.

#P127 POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR

A graded olfactory contrast between nasal passages enables stereo human olfaction

Jennifer Chen[1], Wen Zhou[2], Denise Chen[1]
[1]Psychology Department, Rice University Houston, TX, USA, [2]Institute of Psychology, Chinese Academy of Sciences Beijing, China

The three dimensional world is encoded through pairs of human sensory organs. Each in a pair receives slightly different input from the other. For sight and sound, such gradient is well documented to enable spatial localization. Findings regarding human olfaction have largely been negative with mononaral odorant presentations. Here we demonstrate that humans are capable of olfactory localization when simultaneously presented with odorants that differ in quality and detectability, one to each nostril, hence creating a graded olfactory contrast. The degree of localization varies depending on the contrast between the two nasal inputs. Our findings shed new light on the mechanism of olfactory perception.

#P128 POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR

The Use of a Modified Glatzel Mirror for the Evaluation of Human Nasal Airflow

Caitlin C. Estes, David E. Hornung, Alan Searleman
St. Lawrence University Canton, NY, USA

The objectives of this study were to evaluate the effectiveness of the Glatzel mirror as a tool for measuring airflow in the left and right nostrils of human subjects and to then use this tool to quantify changes in nasal airflow after the application of a nasal dilator. Left-handed and right-handed male and female subjects were tested. To increase the reliability of the plume measurements, a linear scale was fixed to the reflecting surface of the mirror. The subjects breathed onto the modified mirror once every minute for 3 minutes, and the left and right nostril expiration plumes were measured each time. A Breathe Rite nasal dilator was applied after the first 3 minutes, and readings were subsequently taken every minute for 15 minutes. As expected, the plume increased in size immediately after the application of the dilator. However, over the next 15 minutes, the plume size returned to pre-dilator levels, likely reflecting a decrease in lung tidal volume as blood carbon dioxide levels were auto-regulated. Grouping the subjects by handedness indicated that, on average, left-handed subjects experienced a greater increase in the size of the left nostril plume as compared to the size of the right plume. No such differences were observed in right-handed subjects. The data from the left handed subjects are consistent with previously published research suggesting a relationship between handedness and nasal dominance. The data from the present study demonstrate that the modified Glatzel mirror is a sensitive and reliable technique for monitoring nasal airflow. The increase in plume size after dilator application is consistent with previous research suggesting nasal dilators improve olfactory ability by decreasing nasal resistance.

#P129 POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR

The Relationship between Nasal Airflow and Olfactory Perception

Kai Zhao[1], David E. Hornung[2], Donald A. Leopold [3]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]St. Lawrence University Canton, NY, USA, [3]University of Nebraska Medical Center Omaha, NE, USA

We attempted to better understand the relationship between nasal airflow and olfactory perception by perturbing the nasal airway with a nasal dilator while quantitatively documenting the changes in the flow and odorant absorption patterns to the olfactory region using Computational fluid dynamics (CFD) modeling techniques. Two adult subjects had axial CT scans just before and 4 hours after the application of a nasal dilator. After each CT scan, subjects rated the intensity of 8 odorants and one subject had his PEA and butanol olfactory thresholds measured. For both subjects, the physical expansion of the nasal valve area by the dilator was clearly visible on the CT scans. While the physical effects of the nasal dilator were mainly confined to the nasal valve area, a perturbation in airflow patterns persisted into the bony

areas of the nasal cavity. In addition, nasal cycle related unilateral inferior and middle turbinates size increases were observed in one subject. Taking both the effect of the nasal dilator and the nasal cycle into account, CFD models predicted a post dilator decrease in the respiratory peak air velocity bilaterally. However the changes in the overall nasal resistance and odorant absorption rate in the olfactory region were not consistent across both nostrils. In both subjects the uninasal decrease in nasal airflow and odorant absorption was accompanied by an increase in intensity ratings and a decrease in both the butanol and PEA thresholds. This unexpected result suggests olfactory perception is more complicated than a simple direct relationship between flow or absorption rate and olfactory ability. In addition, this study provides, for the first time, some measurements of the effect the nasal cycle may have on nasal airflow and olfactory ability.

#P130 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Direct behavioral evidence for retronasal olfaction in rats

Shree Hari Gautam[1,2], Justus V Verhagen[1,2]
[1]John B Pierce Laboratory New Haven, CT, USA, [2]Department of Neurobiology, Yale School of Medicine New Haven, CT, USA

The neuroscience of perception of flavor is becoming increasingly important to understand feeding behavior and associated diseases such as obesity. Yet, flavor research has mainly depended on human subjects due to the lack of an animal model. A crucial step towards establishing an animal model of flavor research is to establish whether the animal uses the retronasal mode of olfaction, a crucial element of flavor perception. Although indirect evidence of retronasal olfaction in rats exists, it still remains obscure. We designed two different behavioral odor-discrimination and -detection paradigms to test directly whether rats are capable of using retronasal olfaction. In both of these paradigms tasteless aqueous solutions of odorants were licked from vacuum-cleaned lick spout by moderately water-deprived rats. Orthonasal smell was avoided by employing a combination of fans and vacuums. In one paradigm, free-moving rats were asked to discriminate between two orally presented odorants to receive water rewards from one of the two spatially associated spouts (two-alternate forced choice). The other paradigm employed head-fixed rats trained for a retronasal go no-go odor discrimination task. Various odorants, such as, amyl acetate vs. benzaldehyde, 2-hexanone vs. vinyl cyclohexane, were discriminated successfully. Moreover, the tasteless odorant amyl acetate was reliably discriminated against pure distilled water. The accuracy of their performance was well above chance level with relatively higher accuracy in the head-fixed rats. The results from both of these odor-discrimination tasks suggest that rats are capable of smelling retronasally. This direct behavioral evidence establishes the rat as a useful animal model for flavor research.
Acknowledgements: This work is supported by NIH/NIDCD grant. R01DC009994.

#P131 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

On Human Olfactory Sensitivity Across Odorants and Across Subjects

J. Enrique Cometto-Muniz[1], Adriana Tzigantcheva[1], Michael H. Abraham[2]
[1]Chemosensory Perception Laboratory, Dept. of Surgery (Otolaryngology), University of California, San Diego La Jolla, CA, USA, [2]Dept. of Chemistry, University College London London, United Kingdom

We present concentration-detection (i.e., psychometric) odor functions for 28 odorants tested on subgroups from a pool of 132 normosmic, nonsmoker participants (64 female), most of them between 18 and 40 years old (n=123), a few between 41 and 59 years old (n=9, 2 female). Odorants included n-alcohols (n=4), acetate esters (n=4), 2-ketones (n=4), alkylbenzenes (n=5), aldehydes (n=6), and carboxylic acids (n=5). Vapors were presented by dynamic olfactometry. We used a three-alternative forced-choice procedure against carbon-filtered air blanks, and an ascending concentration approach. During subject testing, gas chromatography (flame ionization detector) served to quantify delivered concentrations. The results from 12 subjects (8 female) tested on 10 or more odorants showed strong across-subject agreement regarding the least potent (highest odor detection thresholds) and the most potent (lowest thresholds) odorants. Odor functions from 6 subjects (3 female) tested on a common set of 6 odorants, agreed fully on which were the least and the most potent odorants, and agreed partially on the odorants of intermediate potency. Across these 6 subjects, the odor threshold range between the least and the most potent odorant covered 2 to 3 orders of magnitude. For each of the 6 odorants in common, the ratio between the least and the most sensitive of the 6 subjects was at or below 1 order of magnitude. All odorants had thresholds below 1 part per million by volume (ppm). Notably, carboxylic acids with 4 or more carbons in the chain length, and the aldehydes were the most potent olfactory stimuli with thresholds often below the part per billion (ppb) level. Acknowledgements: Supported by grant R01 DC 002741 form the NIDCD, NIH.

#P132 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Effects of Background Stimuli on Odor Detection Thresholds

Ashley N Phares, Marion E Frank, Thomas P Hettinger
University of Connecticut School of Dental Medicine and Taste and Smell Center Farmington, CT, USA

The ability of humans to distinguish odors is diminished when stimuli are presented in multi-component mixtures (Livermore & Laing, 1996; Goyert et al, 2007). In the present study, we use psychometric functions (Cometto-Muñiz et al, 2008) to study odor mixture suppression. Solutions containing 1, 2 and 3 component combinations of vanillin, l-menthol, and phenethyl alcohol were presented to 18 subjects. Increasing concentrations of vanillin, 0.0001 mM to 1 mM in ½ log steps, were used to determine the detection threshold (ODT) for the vanilla odor. For each concentration, 5 presentations of vanillin were

randomized among 5 water controls. The concentration at which correct identification was 75% (midway between chance, 50% and perfect performance, 100%) was defined as threshold. The data were fitted to a 4-parameter sigmoidal function by Sigmaplot and changes in thresholds evaluated by ANOVA. Addition of near-threshold background stimuli to mixtures affected detection of the vanillin component. With the exception of vanillin + l-menthol, the vanillin ODT within mixtures increased by 1 log unit (vanillin + phenethyl alcohol, p=0.0001; vanillin + phenethyl alcohol + l-menthol, p=0.0001). Thus, the threshold for detection of the vanilla odor increased in the presence of a background containing the rose odor, which we suggest results from competitive mutual suppression between olfactory-bulb glomeruli responding to different stimuli. The mint background odor did not affect the vanilla threshold, which may relate to the intensity or quality of menthol. Menthol elicits a cool sensation through stimulation of the trigeminal nerve. It remains to be tested whether stimuli with a trigeminal component behave differently from pure olfactory stimuli in mixtures. Acknowledgements: Supported by the University of Connecticut School of Dental Medicine.

#P133 **WITHDRAWN**

#P134 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Features of Multicomponent Odour Mixtures Leading to Blending Effect in Humans

Charlotte Sinding, Gérard Coureaud, Claire Chabanet, Adeline Chambault, Noelle Béno, Benoist Schaal, Thierry Thomas-Danguin
Centre des Sciences du Goût et de l'Alimentation (CSGA), CNRS/UB/INRA/AgroSup Dijon 21 000 Dijon, France

Multicomponent odour mixtures have been shown to be mainly configurally perceived. Nevertheless, the factors inducing a blending effect in mixtures remain unclear. To evaluate whether this blending-induced configural perception depends on chemical composition, we used a 6 components mixture (RC) known to produce a blending effect in human adults and in rabbit pups. First, we tested the hypothesis that RC is perceived as different from its components using an odour categorization protocol followed by a perceptual map building. 73 untrained human subjects carried out a free-sorting task on 7 stimuli: RC mixture and its 6 components. Data were analysed through a non-metric MDS and a bootstrap analysis to create a map of dissimilarity with confidence ellipses. Results indicated that RC was not located at the barycentre of the perceptual map; supporting the idea that the mixture carries a specific odour-quality distinct from those of its components. Secondly, we investigated some features that could lead to the configural perception of the mixture, in particular the number of components and their proportions. 73 subjects performed a free-sorting task on 7 stimuli: RC, 2 mixtures similar to RC but in which one component was removed, and 4 mixtures including all the 6 components but in modified proportions. Results showed that RC was significantly isolated from all the other mixtures except for the two including the lowest variations in components proportion. This suggests that blending properties in RC are broken when one component is missing or when there is a large modification in the components proportions. In sum, the blending properties of a multicomponent mixture would result from a specific association of components promoting the perception of a new mixture-specific configural odour. Acknowledgements: Supported by grants from the Burgundy Region, EU-ERDF, IFR 92 and French MESR.

#P135 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Can Probability Summation Account for Gustatory-Olfactory Flavor-Mixture Detection?

Lawrence E. Marks[1,2,3], Maria G. Veldhuizen[1,4], Timothy G. Shepard[1], Adam Y. Shavit[1,2]
[1]John B. Pierce Laboratory New Haven, CT, USA, [2]School of Public Health, Yale University New Haven, CT, USA, [3]Department of Psychology, Yale University New Haven, CT, USA, [4]Department of Psychiatry, Yale University School of Medicine New Haven, CT, USA

Chemosensory stimuli – such as odorants and flavorants – typically contain many components. Often, it is easier to detect these multicomponent stimuli than it is to detect any one component presented alone, detection improving as the number of stimulus components increases. Detection may improve because chemosensory systems integrate the sensory effects of the components. Alternatively, detection may improve probabilistically in the absence of neural integration, even when the components are detected by independent channels (probability summation). Models of probability summation assert or assume that (1) the sensory effect of each stimulus component (e.g., gustatory and olfactory components of a flavorant) is stochastically independent of the sensory effects of other components; (2) the detection of each component is independent of the detection of the others; (3) a separate decision is made whether each component is detected; and (4) the behavioral response depends solely on the set of separate decisions. Models of probability summation traditionally assume high thresholds for the detection of each component, with sensory noise being absent or inconsequential. The core assumptions of probability summation may also be adapted, however, to the framework of signal-detection theory, which recognizes how noise limits sensory detection. The predictions that we derive differ markedly when separate-decision, probability-summation models are formulated within high-threshold and signal-detection frameworks. Detection of gustatory and olfactory flavorants, presented separately and together, suggest that neither high-threshold nor signal-detection models of probability summation can readily account for the results, which may instead reflect sensory-neural integration with the flavor system.
Acknowledgements: Supported by NIH grants DC006688 and DC009021

#P136 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Vapor-phase Long-Chain 18-Carbon Fatty Acids Are Not Discriminated From Blanks Oral-Cavity-Only

Naji A. Wajid[1], Bruce P. Halpern[2]
[1]Cornell University/Biological Sciences Ithaca, NY, USA, [2]Cornell University/Psychology & NBB Ithaca, NY, USA

Oral-cavity-only discrimination between vapor-phase linoleic, oleic, and stearic fatty acids versus blanks was studied, using undiluted fatty acids. A previous study (Bolton and Halpern, Chem Senses 35: 229-38, 2010) had found that vapor-phase 40% oleic acid, 50% linoleic acid, and undiluted stearic acid could not be discriminated from blanks when presented oral-cavity-only, although they could be discriminated from blanks when presented retronasally or orthonasally. METHODS: Undiluted linoleic, oleic, or stearic fatty acids were presented in vapor-phase to 15 participants. For each fatty acid, participants received 5 odorant delivery containers (ODC) on 2 trials, with 4 of the ODC holding one fatty acid and the 5th ODC, a blank (equal volume mineral oil for linoleic and oleic acids, equal weight NaCl for stearic acid). In addition, for a positive control, on 2 trials participants received vapor-phase peppermint flavor odorant versus mineral oil. For all trials, the task was to select the one different ODC. RESULTS: The 1 different ODC was selected on 20% of stearic acid trials, 43% of oleic acid trials, 23% of linoleic trials, and on 90% of peppermint odorant trials. An ANOVA found ≥1 differences between these distributions, $p<0.0001$; pairwise Wilcoxons found no differences between fatty acids. Discrimination between fatty acids and blanks, i.e., selecting the 1 different ODC on both trials (chance $p = 0.04$) was done by 0% of participants for stearic acid, 7% for linoleic acid, 27% for oleic acid, and 80% of participants on peppermint odorant trials. CONCLUSIONS: The long-chain fatty acids linoleic, oleic, and stearic are not discriminated from blanks when presented in vapor-phase oral-cavity-only, suggesting that the oral cavity trigeminal system is not responsive to these fatty acids. Acknowledgements: Support from a Susan Linn Sage Professorship.

#P137 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Vapor-Phase Long-Chain 18-Carbon Fatty Acids can be Discriminated Retronasally

Omar Kallas[1], Bruce P Halpern[2]
[1]Weill Cornell Medical College Doha, Qatar, [2]Cornell University/Departments of Psychology and Neurobiology and Behavior Ithaca, NY, USA

Retronasal discrimination between vapor-phase linoleic, oleic, and stearic fatty acids was studied, using concentrations previously shown to be detectable retronasally (Bolton and Halpern, Chem. Senses 35: 229-38, 2010). The concentrations used were undiluted stearic acid, 40% oleic acid, and 50% linoleic acid. These long-chain, 18-carbon fatty acids were presented in vapor-phase retronasally to 40 participants who received 5 odorant delivery containers (ODC) on 2 trials, with 4 of the ODC holding one fatty acid and the 5th ODC, a different fatty acid. In addition, for a 'negative control', on 2 trials participants received very dilute linoleic acid (0.005%) versus mineral oil. For all trials, the task was to select the one different ODC. RESULTS: The one different ODC was selected on 83% of the stearic acid versus linoleic acid trials, 75% of the stearic acid versus oleic acid trials, 58% of the linoleic acid versus oleic acid trials, and 12% of the 'negative control' 0.005% linoleic acid versus mineral oil trials. An ANOVA found 1 or more differences between these distributions, $p <0.0001$. Discrimination between fatty acids, i.e., selecting the one different ODC on both trials (chance $p = 0.04$) was done by 70% of the participants between stearic and linoleic acids, 65% of the participants between stearic and oleic acids, and by 38% of the participants between linoleic and oleic acids. No participants discriminated on 'negative control' trials. CONCLUSIONS: Vapor-phase stearic acid is discriminated from linoleic and oleic acids when smelled retronasally. Vapor-phase oleic and linoleic acids could be discriminated retronasally from each other by some participants; using undiluted oleic and linoleic acids might produce greater discrimination between them. Acknowledgements: Support from a Susan Linn Sage Professorship at Cornell-Ithaca.

#P138 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Odor training influences olfactory perception in specific anosmia

Selda Olgun[1], Laura Müller[1], Ilona Croy[1], Günter Gisselmann[2], Hanns Hatt[2], Thomas Hummel[1]
[1]Smell and Taste Clinic, Department of Otorhinolaryngology, University of Dresden Medical School Dresden, Germany, [2]Department of Cellular Physiology, University of Bochum Bochum, Germany

The detection threshold of individual odors is highly variable between individual. Specific odors are not detected (or only detected at very high concentrations) in certain group of peoples (i.e. specific anosmia). Recently, it was reported that olfactory function was increased in patients who performed olfactory training compared to baseline. Therefore, this study aimed to examine whether olfactory training can increase sensitivity to specific odors in normosmic people who at first were unable to detect the odors. To screen specific anosmia, we used ten different odors (e.g. isovaleric acid and muscone, etc.) for 1000 normosmic peoples. For 56 people with specific anosmias we performed the "Sniffin' Sticks" test (TDI score) and the EEG-derived olfactory event-related potentials for PEA and H_2S. In addition, we performed an odor detection task using four odors: three control odors and one individual specific odor could not be detected before. All participants were performed by olfactory training using the four odors over 14-16 weeks. After olfactory training, they were asked to perform the "Sniffin' Sticks" test and the odor detection task again. Compared to baseline (i.e. before training), the trained participants increased in their olfactory function, which was observed in general for the "Sniffin' Sticks" test score, and specifically for the detection thresholds of the odors for which subjects exhibited a specific anosmia. In conclusion, the present results demonstrate that "olfactory training" may increase odor perception in specific anosmia. Acknowledgements: Acknowledgement: This project was funded by SPP 1392 "Olfaktorik" of the Deutsche Forschungsgemeinschaft (DFG)

#P139 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Effects of Congruent vs. Incongruent Scent Administration During a Scent Dependent and Information Dependent Learning Task

Justin Schmitt, Bryan Raudenbush
Wheeling Jesuit University Wheeling, WV, USA

A connection between scent and memory has long been recognized. Scent dependent learning exists when the same scent is present in both the learning and assessment phase, which leads to greater performance. The present study assessed scent dependent learning interactions between scent congruent vs. incongruent information. Prior to participation, participants completed the Profile of Mood States (POMS). They then watched a 50 min. video on coffee history under one of three ambient scent conditions (none, coffee, cherry). Following the video, a questionnaire related to the video information was completed under one of three ambient scent conditions (none, coffee, cherry). Following the questionnaire, participants again completed the POMS, in addition to the NASA-TLX to determine perceived workload and task performance. Between-subjects ANOVAs were conducted controlling for coffee preference and consumption. Scent dependent learning was validated, such that performance was better when the same scent was in both the learning and recall situations. Recall was greater than control when the scent in both the learning and recall situations matched the information presented (i.e. coffee). Recall was greater than control when coffee scent was present in the recall situation, regardless of whether it was presented in the learning condition. Thus, scent dependent learning interacts with the type of information being presented, and can provide greatest performance with congruent testing information, even in the absence of that scent being presented in the learning condition.

#P140 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Potential Mechanisms of Odor Referral

Juyun Lim, Maxwell Johnson
Oregon State University/Department of Food Science and Technology Corvallis, OR, USA

Referral of retronasal odors to the mouth is a fundamental phenomenon of flavor perception, yet little is known about the sensory mechanisms that underlie it. The present study investigated the effects of taste and tactile stimulation on retronasal odor referral. Ss reported the perceived locations of odors as they inhaled food odors through the mouth alone or in the presence of either water or tastes in the mouth. When perceived alone, vanilla and soy sauce odor were localized 55:26:19% and 60:22:18% to nose, oral cavity, and tongue, respectively. The localization of odors alone was not significantly different from when water was also presented in the mouth. However, the presence of sucrose and NaCl, but not other tastes, significantly increased (^{2}, $p<.05$) localization of vanilla and soy sauce odor, respectively, to the tongue. These data suggested that retronasal odor can be referred to the mouth in the absence of taste or touch, but that congruent and potentially nutritive tastes increase odor referral to the tongue. In a follow-up study, Ss performed the same tasks under more natural tasting conditions where taste, odor and tactile stimulation were combined in a gelatin. The results showed that when perceived alone, citral and chicken odor were localized 48:28:24% and 48:33:19% to nose, oral cavity, and tongue, respectively. Again, congruent tastes (e.g. sucrose for citral; MSG for chicken odor) significantly enhanced odor referral to the mouth (^{2}, $p<.05$). Interestingly, the presence of congruent tactile stimulation (i.e. an appropriate food texture) improved the ability of congruent taste to evoke odor referral to the mouth. These findings suggest that odor referral is maximized when congruent flavor dimensions are combined to trigger 'flavor objects' that represent known or potential foods. Acknowledgements: This work was supported by Oregon State University start-up funds.

#P141 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Crossmodal olfactory-visual integration in humans

Jessica Albrecht[1,2], Valentin A. Schriever[3], Eva C. Alden[1], Johan N. Lundstrom[1,4,5]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]University of Aachen/Department of Clinical and Interventional Neuroradiology Aachen, Germany, [3]University of Göttingen/Department of Neurophysiology and Cellular Biophysics Göttingen, Germany, [4]University of Pennsylvania/Department of Psychology Philadelphia, PA, USA, [5]Karolinska Institute/Department of Clinical Neuroscience Stockholm, Sweden

In our everyday lives we are faced with multisensory rather than pure unisensory experiences. It has been shown that stimulation of more than one sensory modality enhances behavioral variables and modulates cognitive brain processes. The largest perceptual effects occur when the stimuli are congruent in their spatial and temporal presentation, but also a congruent semantic context is of importance. During a first experiment we assessed the effects of congruent and incongruent visual stimuli on olfactory sensitivity and perceptual ratings and in a second experiment we obtained electrophysiological (ERP) data during olfactory-visual stimulation. In Experiment 1, we used a within-subjects 2 x 3 design to measure olfactory thresholds for two odors while subjects viewed the image of either an empty screen, or an image congruent or incongruent to the odor. In addition, we acquired olfactory intensity and pleasantness ratings for each of the six conditions. In Experiment 2, we measured visual and olfactory ERPs related to each of the six conditions. Experiment 1 demonstrated that odor detection threshold and ratings of odor intensity were not modulated by visual congruency; however, congruent visual stimuli significantly enhanced odor pleasantness ratings for both odors in comparison to either blank or incongruent visual stimulation. ERP data from Experiment 2 is currently being evaluated and will be presented. Results from Experiment 1 suggest that peripheral olfactory functions are not modulated by visual stimuli, whereas higher-order cognitive olfactory processes are influenced by a congruent visual stimulus. Acknowledgements: Supported by start-up funds from the Monell Center awarded to JNL and a DAAD postdoctoral fellowship D/08/40252 awarded to JA.

#P142 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

The Eyes Tend to Follow the Nose: Olfaction Guides Visual Attention

Kepu Chen, Shan Chen, Bin Zhou, Wen Zhou
Institute of Psychology, Chinese Academy of Sciences Beijing, China

Attention is intrinsic to our perceptual representations of sensory inputs and is typically depicted as a spotlight moving over a saliency map which, in the case of visual attention, topographically encodes strengths of visual features and feedback modulations over the visual scene. The current study introduces smells to two well established attentional paradigms – the dot-probe paradigm and the visual search paradigm—to assess whether olfactory inputs modify the deployment of visual attention. We find that a smell automatically attracts attention to the congruent visual image and facilitates visual search of that image in a manner independent of cognitive control. We thus propose that smell quality acts as an object feature whose presence adds to the perceptual saliency of the corresponding object, thereby guiding the spotlight of visual attention. Our discoveries provide the first empirical evidence that a non-visual sensory attribute is weighted in the saliency map of visual inputs and suggest such feature integration operates at a relative early stage of perceptual processing. Acknowledgements: Supported by the Knowledge Innovation Program of the Chinese Academy of Sciences Grants No. KSCX2-YW-R-250 and 09CX192019

#P143 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Nostril-specific olfactory modulation of binocular rivalry

Wen Zhou[1], Jennifer Chen[2], Xiaomeng Zhang[1], Li Wang[1], Yi Jiang[1], Denise Chen[2]
[1]Institute of Psychology, Chinese Academy of Sciences Beijing, China, [2]Psychology Department, Rice University Houston, TX, USA

It is known that olfaction and vision can work in tandem to represent object identities. What is yet unclear is the stage of the sensory processing hierarchy at which the two types of inputs converge. Here we study this issue through a well-established visual phenomenon termed binocular rivalry. We show that smelling an odor from one nostril significantly enhances the dominance time of the congruent visual image in the contralateral visual field, relative to that in the ipsilateral visual field. Moreover, such lateralization-based enhancement extends to category-selective regions so that when two images of words and human body, respectively, are engaged in rivalry in the center visual field, smelling natural human body odor from the right nostril increases the dominance time of the body image as compared with smelling it from the left nostril. These results, taking advantage of the anatomical and functional lateralization in the olfactory and visual systems, provide strong evidence for an object-based early convergence of olfactory and visual inputs in feed-forward sensory processings. Acknowledgements: Knowledge Innovation Program of the Chinese Academy of Sciences grants KSCX2-YW-R-250, KSCX2-YW-R-248, and 09CX192019

#P144 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Background sounds influence performance of odor discrimination task

Mandy Scheibe, Volker Gudziol, Antje Hähner, Han-Seok Seo
Smell & Taste Clinic, Department of Otorhinolaryngology, University of Dresden Medical School Dresden, Germany

We often smell odors while being exposed to various background sounds. One example is an experience of crowd party where people smell many different odors including food odors, body odors, and ambient fragrances while hearing background music or listening to a conversation. In addition, we often perceive food odors via ortho- and/or retronasal route during eating or drinking. Despite of its high occurrence, little is known about an association between olfactory and auditory stimuli in humans. This study aimed to investigate whether background sound can modulate participants' performance in an odor discrimination task. Participants were asked to perform the odor discrimination task while listening to either background noise (e.g. verbal or non-verbal noise) or no additional sound (i.e. "silent" condition). Participants' performance in odor discrimination task was significantly more deteriorated in the presence of background noise than in the silent condition. Rather, the disrupting effect of verbal noise on the task performance was significantly higher than that of non-verbal noise. In conclusion, our findings provide new empirical evidence that background sound influences the performance in an odor discrimination task.

#P145 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

The Scratch and Sniff "Wheel": A New Smell Identification Test for Children

E. Leslie Cameron[1], Richard L. Doty[2]
[1]Carthage College, Department of Psychology Kenosha, WI, USA, [2]University of Pennsylvania, Smell & Taste Center Philadelphia, PA, USA

Olfactory identification ability is commonly assessed with "scratch and sniff" smell tests such as the University of Pennsylvania Smell Identification Test (UPSIT) or its shorter versions, such as the 12-item Brief Smell Identification Test (B-SIT). Most such tests employ booklets for stimulus presentation. The current pilot study assessed the test performance of children and young adults on, and acceptability of, a newly developed game-like rotating "wheel" microencapsulated odor test specifically designed for grade school classrooms. The test, which is comprised of a rotating disk within an outside folder, contained 11 microencapsulated odorants and both pictures and verbal labels in a 4-item multiple-choice alternative format. METHOD: Ninety-three (50 female) children from a public elementary school (6-7 and 10-11 year olds) and young adults (17-18 year old college students) participated. Each was instructed according to the direction on the test and told how the game was to be played. RESULTS: Testing was very rapid (less than 5 minutes per participant) and enjoyable for the participants. No student evidenced any problem understanding the rotating disk test format. An ANOVA revealed a main effect of age (odor identification performance increased with age), but no effects of gender nor its interaction with age. CONCLUSION: The rotating disk format for presenting odors was found to be acceptable to both children and adults. Like analogous booklet tests, this test was sensitive to age. Because of the game-like format, this test will likely be of value in the self-administered testing of young children.

#P146 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Automated odor presentation for odor identification testing

Valentin A Schriever[1], Samanta Viana[2], Thomas Hummel[3]
[1]University of Goettingen Department of Neurophysiology and Cellular Biophysics Goettingen, Germany, [2]Faculdada de Medicina da Universidada de Lisboa Lisboa, Spain, [3]Smell & Tast Clinic Department of Otorhinolaryngology University of Dresden Medical School Dresden, Germany

Odor identification tests are widely used instruments for the assessment of specific aspects of olfactory function. There are many test in use like the UPSIT or the "Sniffin' Sticks" testing battery. So far most tests are administered by an experimenter. Therefore, they are time consuming and costly. We evaluated an automated odor presentation odor identification test using the olfactometer aerome® ScentController (aerome, Cologne, Germany). Seventy volunteers participated in the study. Participants performed two ten item odor identification tests in one session: One test was the self-administered test using the olfactometer, the other test was administered by an experimenter using the "Sniffin' Sticks". Twenty participants repeated testing on a different day to obtain the re-test reliability of either technique. Participants reached significantly higher scores on the "Sniffin' Sticks" odor identification test than on the olfactometer based test. This effect was driven by individual odors. No difference in odor identification scores was found after exclusion of these odors from the analysis. Both methods showed no significant difference in scores obtained during the first and second session, indicating that results were comparable between sessions. In conclusion, the automated odor presentation method was shown to be a valid option for a self administered odor identification test. We would like to thank aerome GmbH Ambient Air Care Cologne, Germany (http://www.aerome.de), for their technical support.

#P147 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Concentration as an Inappropriate Index of Chemosensory Potency

William S. Cain[1], Michael H. Abraham[2], J. Enrique Cometto-Muñiz[1], Roland Schmidt[1]
[1]UC San Diego La Jolla, CA, USA, [2]University College London London, United Kingdom

The objective is to clarify the basis of a QSAR for odor potency. Abraham et al. demonstrated applicability of a linear free energy relationship (LFER) to chemesthetic potency. The equation, with descriptors for excess molar refraction, dipolarity/polarizability, hydrogen bond acidity and basicity, resp., and lipophilicity could account for potency to humans and various rodents (rsq about 0.95). The generality seemed consistent with the simplicity of chemesthesis phenomenologically and mechanistically. When solved for odor, the variables accounted for less variance (rsq about 0.75 to 0.80), a possible reflection of greater complexity phenomenologically and mechanistically. This outcome occurred previously, and did so here with data of precision and more than 200 materials ("Nagata set"). The outcome tempted the conclusion that chemesthetic potency depends just upon selective factors related to transport, whereas odor potency depends also upon specific factors such as molecular complementarity (e.g., shape). Research on input-output functions for individual materials suggests another possibility, viz., the two LFERs account for the same amount of variance. Chemesthetic output vs. an input of concentration invariably exhibits expansion whereas olfactory output vs. input of the same variable of concentration invariably exhibits compression. When normalized for this, the Abraham equation can do as well for odor as for chemesthetic detection. If residual variance of 5% is acceptable, then we have no reason to improve the Abraham model for olfaction, except some calculational refinements. These should include a measure of potency other than simple concentration, which introduces systematic error in the chemesthetic-olfactory comparison, much as does dB(SPL) vs. dB(SWL) in sound measurement.
Acknowledgements: Supported by NIH grants R01 DC002741, R01 DC005003, and R01 DC05602.

#P148 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Relationship between Gray and White Matter Brain Volume and Body Mass Index in Healthy Adults

Sanne Boesveldt[1,2], Jessica Albrecht[1,3], Johannes Gerber[4], Thomas Hummel[4], Johan N. Lundstrom[1,5,6]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]Wageningen Universiteit, Division of Human Nutrition Wageningen, Netherlands, [3]University Hospital Aachen, Department of Diagnostic and Interventional Neuroradiology Aachen, Germany, [4]Interdisciplinary Center for Smell & Taste Research, University of Dresden Medical School Dresden, Germany, [5]University of Pennsylvania, Department of Psychology Philadelphia, PA, USA, [6]Karolinska Institute, Department of Clinical Neuroscience Stockholm, Sweden

Obesity is a growing health problem that can lead to a plethora of adverse health effects, including possibly poorer performance on tests measuring cognitive abilities (e.g. *Kerwin 2010, Gunstad 2010*). It has been demonstrated that volumetric measures of the cortex correlate with behavioral performance on both perceptual and cognitive behavioral tasks. The aim of our study therefore was to investigate the association between gray and white matter brain volume and body mass index (BMI) in healthy subjects. We hypothesized that grey matter volume in the medial frontal cortex and hypothalamus would correlate with BMI. To this end, we collected a total of 97 anatomical T1 weighted images on a 1.5 T MRI scanner from young and older adults (mean age 36.7 yrs; mean BMI 23.4 kg/m^2, range 16.8-36.2 kg/m^2; 52 women), and used voxel-based morphometry (VBM5 toolbox in SPM5) to determine gray and white matter brain volumes. Age of the subjects was parceled out in all analyses. Our preliminary results indicate that higher BMI is associated with decreased gray matter volume in the right precentral gyrus, right medial frontal gyrus, bilateral thalamus, right superior frontal gyrus, and left occipital gyri. No relationship was found for the hypothalamus. BMI was unexpectedly positively correlated with white matter volume in bilateral gyrus rectus. More detailed analyses will be presented. In conclusion, these preliminary data suggest that overweight is associated with detectable alterations in both gray and white matter brain volume in (otherwise) healthy subjects; possibly leading to future cognitive decline. Whether these neural changes precede obesity, as a clinical marker, or are the consequence of weight gain, needs to be determined in longitudinal studies.
Acknowledgements: Supported by startup funds from the Monell Chemical Senses Center awarded to JNL and a DAAD postdoctoral fellowship D/08/40252 awarded to JA.

#P149 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Sweet expectations: Inverse relationship between BMI and negative prediction error for a sweet taste

Danielle M. Douglas[1], Maria G. Veldhuizen[1,2], Darren R. Gitelman[3,6], Dana M. Small[1,2,3,4]
[1]The John B. Pierce Laboratory New Haven, CT, USA, [2]Department of Psychiatry, Yale University School of Medicine New Haven, CT, USA, [3]Department of Psychology, Yale University New Haven, CT, USA, [4]Interdepartmental Neuroscience, Yale University School of Medicine New Haven, CT, USA, [5]Department of Neurology, Northwestern University Chicago, IL, USA, [6]Department of Radiology, Northwestern University Chicago, IL, USA

Dopamine (DA) plays a primary role in learning about rewards by generating error signals during unexpected outcomes. When a reward is greater than expected a "positive error" signal is generated and when the reward is less than expected a "negative error" signal is generated. Human fMRI studies implicate the ventral striatum (VS) in coding prediction errors. Prior work has also shown that DA system differs as a function of body mass index (BMI). We used fMRI to test the influence of BMI on brain response to expected or unexpected oral stimuli. During expected trials (70%) subjects heard either "sweet" or "tasteless" and received the liquid indicated by the verbal cue. During unexpected trials (30%) subjects heard "sweet" but received tasteless (negative error signal) or they heard "tasteless" but received sweet (positive

error signal). A main effect of expectation was observed in the VS, with greater response observed during unexpected vs. expected oral stimulation, regardless of the direction of the error signal generated. BMI was then regressed against brain response to expected and unexpected oral stimuli. A positive relationship between BMI and response to expected sweet – tasteless was observed in medial orbitofrontal cortex (MOFC; $z = 4.11$; $p_{FWE} = .03$). A significant negative relationship was observed between BMI and response in the VS during the generation of negative error signals (expect sweet but receive tasteless vs. expect sweet and receive sweet; $z = 4.13$ $p_{FWE} = .03$). We conclude that body weight is positively related to MOFC response to an expected sweet taste and inversely related to generation of negative error signals in the VS when sweet is expected but not received. Results are consistent with reduced striatal DA signaling in obesity. Acknowledgements: This work was supported by NIDCD R01 DC006706-04

#P150 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Relationship Between Diet Soda Consumption and fMRI Activation to Non-Nutritive Sweetener in Young Adults

Erin Green[1], Aaron Jacobson[2], Lori Haase[1], Claire Murphy[1,2,3]
[1]SDSU/UCSD Joint Doctoral Program in Clinical Psychology San Diego, CA, USA, [2]San Diego State University San Diego, CA, USA, [3]University of California, San Diego San Diego, CA, USA

Links between consumption of sodas sweetened with non-nutritive sweeteners and increased incidence of obesity have recently been debated, although the mechanism driving this relationship remains unknown. In the present study, we examined whether individuals who do or do not regularly consume drinks containing non-nutritive sweeteners (i.e., diet soda) have differential brain activation in response to a saccharin solution. During functional magnetic resonance imaging sessions, participants were given .3ml of a saccharin solution 16 times during two separate runs; once after fasting for 12 hours and once after ingesting a nutritional preload. Participants also rated the intensity and pleasantness of the stimuli pre- and post-preload and after the scan. For both hunger and satiety conditions, participants who drink diet soda had brain responses to the non-nutritive taste stimulus that differed from those who do not regularly drink diet soda. Specifically, young adults who do not drink diet soda demonstrated significant activation of the right insula and left thalamus during hunger and right inferior frontal gyrus during satiety. Individuals who regularly consume diet soda demonstrated bilateral caudate activation in the hunger condition and thalamus activation when satiated. There were also differences in psychophysical ratings of saccharin. These results suggest that regular diet soda drinkers may have altered physiological and psychophysical responses to sweet tastes that are not associated with caloric value. Further research examining the effects of regular ingestion of natural and non-nutritive sweeteners on psychophysical properties of and physiological responses to sweet taste is warranted. Acknowledgements: Supported by NIH Grant #1 R01 AG04085 to C.M.

#P151 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Gender Differences in Cortical Activation in Response to Pleasantness Evaluation During Hunger and Satiety

Lori Haase[1], Erin Green[1], Claire Murphy[1,2,3]
[1]SDSU/UCSD Joint Doctoral Program in Clinical Psychology San Diego, CA, USA, [2]San Diego State University San Diego, CA, USA, [3]University of California, San Diego San Diego, CA, USA

Event-related fMRI was utilized to investigate differences in the neural correlates of pleasantness evaluation in response to 4 taste stimuli (sucrose, citric acid, NaCl, and caffeine) between males and females during the physiological conditions of hunger and satiety. Prior to the scans, participants fasted for 12 hours and were randomly presented with either a pre-load consisting of 474ml (two bottles) of Vanilla flavored Ensure Plus (sated) or were not administered a pre-load (hungry). Stimuli were dissolved in distilled water and were delivered to the mouth via computer-controlled pumps as 0.3ml of solution in 1 s boluses. The subject's task was to rate the pleasantness of stimuli using the General Labeled Magnitude Scale. Imaging was conducted on a 3T GE scanner using a standard gradient echo EPI pulse sequence to acquire T2*-weighted functional images. To conduct group analysis, a one-sampled t-test was calculated on the fit coefficient corresponding to each contrast at each voxel. Voxels with a fit coefficient meeting a threshold of $p < 0.005$ and belonging to clusters of at least 17 voxels were considered as activated. There were no significant differences between males and females in hunger ratings or in the evaluation of intensity and pleasantness for the taste stimuli. Females showed significantly more activation than males when sated in response to caffeine, sucrose and citric acid in regions implicated in dopamine release (insula, putamen) and the default network (precuneus, posterior cingulate). These findings suggest that despite equivalent psychophysical ratings for the taste stimuli and perceived hunger, there are differences in cortical activation between males and females in reward and default network regions. Acknowledgements: NIH grants R01AG04085 to Claire Murphy, Ph.D.

#P152 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Neural Correlates of Flavor-Nutrient Conditioning in Humans

Wambura Fobbs[1,2], Maria G. Veldhuizen[1,2,3], Danielle M Douglas[1], Tammy Lin[1], Martin Yeomans[5], Linda Flammer[6], Dana M. Small[1,2,3,4]
[1]John B. Pierce Laboratory New Haven, CT, USA, [2]Yale University New Haven, CT, USA, [3]Department of Psychiatry, Yale University New Haven, CT, USA, [4]Department of Psychology, Yale University New Haven, CT, USA, [5]University of Sussex Brighton, United Kingdom, [6]PepsiCo Valhalla, NY, USA

Long-lasting preferences develop for flavors paired with caloric nutrients, an associative process designated "flavor-nutrient conditioning". Animal studies have established a crucial role for the amygdala, but not the insula, in the acquisition and expression of flavor-nutrient conditioning (Touzani and Sclafani, 2005, 2007).

Although behavioral paradigms have demonstrated flavor-nutrient conditioning in humans, the neural correlates remain unknown (Yeomans et al., 2008). The aim of the current study was to use fMRI to determine which brain regions are sensitive to the conditioning between novel flavors and maltodextrin (M) in humans. Only subjects who could not detect M in solution with flavors were included. Subjects were exposed six times, while hungry, to flavorful drinks, containing either 0, or up to 150 calories derived from M. Pleasantness ratings for the flavors (minus M) were assessed pre and post-conditioning. Brain response to the flavors (minus M) was assessed post-conditioning with fMRI. As predicted, pleasantness ratings increased preferentially for flavors associated with calories. In addition, response in the amygdala, ventral striatum and medial orbitofrontal cortex was significantly greater during ingestion of flavors that had been associated with calories. In contrast, and in keeping with the animal literature, insular response to flavors was not influenced by prior association with caloric load. We conclude that the neural network underlying flavor-nutrient conditioning in humans includes the amygdala, striatum and medial orbitofrontal, but not insular, cortex. Acknowledgements: PepsiCo.

#P153 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

A salty-congruent odor enhances saltiness: fMRI study

Han-Seok Seo[1], Emilia Iannilli[1], Cornelia Hummel[1], Yoshiro Okazaki[1], Dorothee Buschhüter[1], Johannes Gerber[2], Gerhard E. Krammer[3]
[1]University of Dresden Medical School/Department of Otorhinolaryngology Dresden, Germany, [2]University of Dresden Medical School/Department of Neuroradiology Dresden, Germany, [3]Symrise AG/Research & Innovation Holzminden, Germany

Excessive intake of dietary salt (i.e. NaCl) may increase the risk of chronic diseases. Especially, cardiovascular diseases caused by sodium induced elevated blood pressure. Accordingly, various strategies to reduce salt intake have been conducted around the world. The present study aimed to investigate whether a salty-congruent odor can enhance saltiness on the basis of psychophysical (Experiment 1) and neuroanatomical levels (Experiment 2) using functional magnetic resonance imaging (fMRI). In Experiment 1, after receiving one of six stimulus conditions: three odor conditions (odorless air, congruent, or incongruent odor) by two taste concentrations (low or high) of salty or sweet taste solution, participants were asked to rate taste intensity and pleasantness. In Experiment 2, participants received the same stimulus condition during the fMRI scan. In Experiment 1, compared with incongruent odors and/or odorless air, congruent odors enhanced taste intensity and modulated taste pleasantness in sweet or salty taste solution. In Experiment 2, a salty-congruent combination of odor and taste produced significantly higher neuronal activations in brain regions associated with odor-taste integration (e.g., insula, frontal operculum, anterior cingulate cortex, and orbitofrontal cortex) than an incongruent combination and/or odorless air with taste solution. In addition, the congruent-induced saltiness enhancement was more pronounced in the low concentrated taste solution than in the high concentrated one. In conclusion, the current study demonstrates the congruent odor-induced saltiness on the basis of psychophysical and neuroanatomical results. These findings support the alternative strategy to reduce excessive salt intake by adding salty-congruent aroma molecules or mixtures to sodium reduced food. Acknowledgements: This research was supported by Symrise AG, Holzminden, Germany.

#P154 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Parallel pathways mediate attention to taste in humans

Maria G Veldhuizen[1,2], Darren R Gitelman[3,4], Dana M Small[1,2,5]
[1]The John B Pierce Laboratory New Haven, CT, USA, [2]Department of Psychiatry, Yale School of Medicine New Haven, CT, USA, [3]Department of Neurology, Northwestern University Chicago, IL, USA, [4]Department of Radiology, Northwestern University Chicago, IL, USA, [5]Department of Psychology, Yale University New Haven, CT, USA

In a prior study we showed that trying to detect a taste in a tasteless solution results in enhanced activity in gustatory cortex in anterior insula and frontal operculum (AIFO), in parietal operculum (PO), and in the canonical attention network (anterior cingulate cortex (ACC), frontal eye fields (FEF), and posterior parietal cortex (PPC)). The aim of the current study was to test whether the attention network modulates taste cortex activity during attention to taste. FMRI was used to measure BOLD responses in 14 subjects. Subjects performed two different tasks in the scanner: trying to detect a taste in a solution, or passively perceiving the same solution. We used a psychophysiological interaction analysis (SPM2) to find regions demonstrating increased connectivity with the attention network, by looking for areas who's activity correlated with the interaction between the neural response from regions in attention network with the psychological variable (attention vs. passive). We observed greater connectivity between the neural response in source regions including the FEF, PPC, and PO and response in the ACC, and in turn between ACC and the right AIFO, the same region that shows baseline increase in neural response during attention to taste. These results suggest that selective attention to taste is mediated by a hierarchical circuit in which signals are sent from the FEF, PPC and PO to the ACC, which in turn modulates responses in the AIFO. We used the dynamic causal modeling (DCM) module in SPM5 to evaluate this possibility, and observed evidence for direct and indirect (via ACC) modulation of the attention network on AIFO. This suggests that the ACC may have a special role in gustatory processing in acting as an intermediary between attention network and gustatory cortex. Acknowledgements: NIDCD grant R01 DC006706

#P155 **POSTER SESSION III: OLFACTION AND TASTE DEVELOPMENT; OLFACTORY PSYCHOPHYSICS; FUNCTIONAL IMAGING: TASTE AND FLAVOR**

Evidence for ventral and dorsal streams in the chemical senses

Johannes Frasnelli[1], Johan N Lundstrom[2], Simona Negoias[3], Johannes Gerber[3], Thomas Hummel[3], Franco Lepore[1]
[1]CERNEC, Université de Montréal Montreal, QC, Canada, [2]Monell Chemical Senses Center Philadelphia, PA, USA, [3]Technical University of Dresden Medical School Dresden, Germany

Basic sensory processing occurs in primary and secondary sensory regions of the brain, distinctly for the different sensory systems. Higher order processing, however, seems to follow a general subdivision into ventral and dorsal streams. Object identification in visual, auditory and tactile senses is processed in temporal structures (ventral stream), whereas object localization leads to activation of parietal structures (dorsal stream). In order to examine whether the chemical senses demonstrate a similar dissociation, we investigated odor identification and odor localization in 16 healthy young subjects by means of functional MRI. Odor localization was quantified as detection of which nostril was stimulated in a monorhinal presentation; a task only possible if the odor also activates the trigeminal system. To accommodate this, subjects were stimulated monorhinally with mixed olfactory-trigeminal stimuli, i.e., chemical stimuli which activate both the olfactory and the trigeminal system. Therefore we used eucalyptol and a mixture of a rose odor and CO2 as chemosensory stimuli. The subjects' task was either to localize (left – right) or to identify (eucalyptol – rose) the presented stimuli. Confirming a dual processing stream in the chemical senses, contrasting the two tasks to identify areas responsible for odor identification rendered a significant cluster in the left superior temporal sulcus. Conversely, several clusters in the parietal lobe were found when subjects localized the odors, including the superior and inferior parietal lobule and precuneus bilaterally. These data demonstrate that higher order chemosensory processing shares the general subdivision into a ventral and a dorsal processing stream with other sensory systems and suggest that this is a global, sensory independent function.
Acknowledgements: This research was supported by a RBIQ pilote grant. JF is supported by FRSQ and CIHR.

#P156 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Peripheral Mechanisms that Expand the Olfactory Code

Anandasankar Ray[1,2,3], Sean M Boyle[2], Stephanie L Turner[3]
[1]Entomology Dept Riverside, CA, USA, [2]GGB Program Riverside, CA, USA, [3]CMDB Program, University of California Riverside, CA, USA

Little is understood about how large ensembles of odor receptor proteins detect small volatile molecules with high specificity and sensitivity. Presence of an odorant in the environment is usually thought to cause either activation or inhibition of specific odorant receptors and neuronal circuits. Here we use fruit flies and mosquitoes to report identification of novel odorants that cause long-term changes in the ability of an olfactory receptor to respond to other odorants. The long-term response-modifiers fall into two categories – inhibitors and activators. Identification of these odorants was possible using a cheminformatics screen for ligands with new properties from a library of >400,000 compounds representing most volatiles. Subsequent functional analysis supported a high success rate (~75%) for this screen and led to the identification of numerous new activators and inhibitors for many of the odorant receptors, a small fraction of which caused long-term modification of activity. Behavioral analysis in both fruit flies and mosquitoes revealed that brief exposure to these odorants caused significant modification of behavior for several minutes beyond the stimulus period. Taken together, the identified odorants represent a new class of odor receptor ligands that can modify activity and signaling beyond the duration of odor stimulus. Since these odorants are present in natural environments our findings imply that coding of odorants by the peripheral olfactory system, and subsequent behaviors, can be influenced not only by the context within which such odors are present in a blend but also the recent history of odorant exposure of the organism. Acknowledgements: Supported by NIH grant NIAID R01AI087785 to A.R and S.M.B is supported in part by an NSF-IGERT grant.

#P157 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

The Functional Evolution of Odorant Receptor Orthologs

Kaylin A. Adipietro, Joel D. Mainland, Hiroaki Matsunami
Duke University Medical Center/Molecular Genetics and Microbiology Durham, NC, USA

The olfactory system has evolved to adapt to the environment; the odorant receptor (OR) repertoire has been subjected to rapid evolution with extensive gains and losses between species. Most OR evolutionary studies have employed bioinformatics to predict OR function. Though it is generally assumed that orthologs—genes that originated from a common ancestor and did not undergo gene duplication or loss—retain the same function, this assumption remains largely untested for ORs. Here we investigated the functional properties of primate and rodent OR orthologs to determine whether gene orthology accurately predicts functional characteristics and if amino acid changes in OR coding regions can be related to changes in OR function. Using a heterologous expression system, we matched human ORs with ligands. From this set of ORs, we identified and cloned 17 OR orthologs from chimpanzee and rhesus macaque genomic DNA into a mammalian expression vector. We also cloned 28 mouse-rat orthologous pairs. These ORs were transiently transfected into heterologous cells with accessory proteins and functionally characterized using a luciferase reporter assay; each OR was tested against a panel of chemically diverse odors.
We found evidence of functional changes across the majority of OR orthologs. 94% of orthologs showed differences in ligand sensitivity (EC50), while some ORs also showed changes in ligand selectivity. Our results suggest that orthology does not accurately predict OR function and that amino acid changes during evolution alter receptor properties. While we do not know what impact this has on the behavior of an animal, we can speculate that the functional changes of orthologs occurred to meet species-

specific demands. Acknowledgements: This work was funded by an R01 from the National Institute of Deafness and Other Communication Disorders (NIH/NIDCD) and a fellowship from the Duke University Primate Genomics Initiative.

#P158 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

An in vitro odorant receptor expression system that mimics ligand selectivity and sensitivity of olfactory sensory neurons expressing the corresponding receptors

Yi Dong, Hiroaki Matsunami
Department of Molecular Genetics and Microbiology, Duke University Medical Center Durham, NC, USA

Odorant receptors (ORs) respond to a wide variety of odorous ligands. The process of matching odors with a particular mammalian OR has been made more convenient with the advent of in vitro techniques using transfected culture cells. Though in vitro data predict odor perception and are mostly consistent with data obtained using in vivo models, potential concerns remain regarding whether data obtained through in vitro systems can match the data describing responses of olfactory sensory neurons (OSNs) in vivo. A confounding factor also exists in that different in vivo and in vitro methods are used for assessing different ORs. Here we demonstrate the efficacy of utilizing a single in vitro system based on cAMP-mediated reporter assays with five mouse ORs which had their ligand specificities elucidated using transgenic mouse models: I7, OR-EG, MOR23 and M71 and SR1. Plasmid DNA encoding each OR was transfected into Hana 3A cells with plasmids encoding RTP1S and muscarinic receptor M3 as well as luciferase reporters. Luciferase assay was run after odor stimulation. Dose-response curves of tested odors were compared to the previously published data, obtained with the same odors, which were derived from calcium imaging of dissociated OSNs and patch clamp recording of intact OSNs that expressed the corresponding OR. We found that the in vitro ligand selectivity of each OR matched closely with that of the previous studies. Regarding sensitivity, the in vitro responses of each OR roughly matched with average responses by calcium imaging (OR-EG and M71). The in vitro response also fell within the ranges of single OSNs by patch recordings (I7, MOR23 and SR1), though less responsive than average. Our data suggest that use of the in vitro system represents a valid method of determining OR function. Acknowledgements: This work was funded by an R01 from the National Institute of Deafness and Other Communication Disorders (NIH/NIDCD).

#P159 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Olfactory receptors coded by segregating pseudo genes and odorants with known specific anosmia

Kaveh Baghaei, Günter Gisselmann, Hanns Hatt
Ruhr-Universität Bochum/Cell physiology Bochum, Germany

Olfactory receptors (ORs) family has evolved to detect a wide range of chemical structures and it exhibits extensive genetic diversity within human population. In several examples it was demonstrated that variation in OR genes by SNPs and CNVs is the main origin for such kind of differences. Also it is described that some OR genes display both functional (wild type), functional mutated and nonfunctional alleles, and thus, are called segregating pseudo genes (SPGs). It's suggested, that SPGs are one of the main reason for variation in human odor perception. Regarding to these properties we focused on deorphanizing ORs of this SPG group and tried to match them with odorants related to known specific anosmia. Functional expression in HEK293 cells and Fura-2 based Ca-imaging was used as main technique for deorphanization of olfactory receptors. We used odorant mixtures derived from different chemical groups (ketones, aldehydes, alcohols, amines and acids) for stimulation. In one instance, we found that "sweet" smelling members of the alcohol group induced an elevation of Ca^{2+} in cells transfected with the functional variant of OR2F1. Application of ketone, acid, aldehyde or amine mixes didn't elicit any response in OR2F1 expressing cells. The pseudogene variant of OR2F1 contains an amino acid exchange (arginin to cystein at amino acid pos. 122) in the highly conserved DRY-motive and is thought to be non-functional. It seems that sweet smelling alcohols with relation to specific anosmia are suitable ligands for the functional variant of OR2F1.

#P160 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Functional variability in the human odorant receptor repertoire

Joel D. Mainland, Ting Zhou, Hiroaki Matsunami
Duke University Durham, NC, USA

Humans have approximately 400 odorant receptors (ORs) with an intact open reading frame, but among this set there are a large number of polymorphisms between individuals. We sequenced 78 ORs in 20 ethnically diverse subjects and phased alleles using MACH. We found 357 nsSNPs representing 376 unique alleles. Each OR had a median of 4 alleles. Two sets of segregating polymorphisms have previously been linked to variability in odor perception, but it is unclear if naturally-occuring polymorphisms generally affect the function of a receptor. Using a heterologous expression system we identified ligands for 28 human odorant receptors. We found extensive functional variation between reference alleles and minor alleles. For the odorant receptors with known ligands and multiple clones, 85% had at least one minor frequency allele with a significantly different dose response from the reference allele. This description of natural genetic variation and *in vitro* functional variation provides a platform for identifying the role of a single odorant receptor in human perception. Acknowledgements: NIH-NIDCD

#P161 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Variation in the human olfactory subgenome and its impact on olfactory perception

Jonas Kuklan[1], Günter Gisselmann[1], Laura Müller[2], Selda Olgun[2], Thomas Hummel[2], Hanns Hatt[1]
[1]Dept. of Cell Physiology, Ruhr-University Bochum Bochum, Germany, [2]Smell & Taste Clinic, Dept. of ORL, TU Dresden Medical School Dresden, Germany

The olfactory receptors (ORs) are a family of G-Protein coupled receptors that provide the molecular basis for the detection of volatile odorant molecules by the central nervous system. In humans, the *OR* gene family comprises about 400 functional genes and about 600 non-functional pseudogenes. Furthermore, the *OR* gene family shows a high degree of genetic variability between individuals. Common single nucleotide polymorphisms (SNPs) and copy number variants (CNVs) lead to specific patterns of functional and non-functional *OR* genes in each individual subject, resulting in "different noses for different people" (Menashe *et al.*, Nat Genet, 2003). At the same time, it has long been known that people differ in their ability to perceive certain odorants. In the most striking cases, some people completely lack the ability to detect certain odorants, although their sense of smell functions normally in general. This phenomenon is known as specific anosmia and in some cases has been shown to have a genetic basis. We used a range of methods from molecular biology to psychophysical studies to investigate the effects of genetic variance in the human olfactory subgenome on the physiological function of the olfactory system. We obtained genomic DNA samples from subjects with a specific anosmia for Pentadecalacton, Geraniol, Benzylsalicylate, Galaxolid, Musk-Ketone or Lyral. We then used massive parallel sequencing, DNA microarray and real time quantitative PCR techniques to identify genetic polymorphisms in the olfactory subgenome that show an association to these specific anosmias. Experiments were performed on pools of DNA samples to maximize cost-efficiency. Acknowledgements: This project was funded by SPP 1392 "Olfaktorik" of the Deutsche Forschungsgemeinschaft (DFG)

#P162 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Next Generation Sequencing Reveals Variation in Vomeronasal Receptor Gene Repertoires Across 17 Mouse Strains

Darren W. Logan[1,2], Elizabeth Wynn[1], Keren J. Carss[1], Mouse Genomes Project[1,3,4]
[1]Wellcome Trust Sanger Institute Hinxton, United Kingdom, [2]MRC Centre for Obesity and Related Metabolic Diseases Cambridge, United Kingdom, [3]Wellcome Trust Centre for Human Genetics Oxford, United Kingdom, [4]University of California, Los Angeles Los Angeles, CA, USA

Innate behaviour in mice, including aggression, sex and parenting are largely instructed by signalling pheromones detected by vomeronasal receptors (VRs). As inbred strains display stereotypical differences in such behaviour, we hypothesize that variation in VR genes may underpin these phenotypes. The Mouse Genomes Project aims to generate a catalogue of inter-strain genomic variation, including the mapping of all single-nucleotide polymorphisms (SNPs), by Illumina sequencing. Here we report the SNP content of 17 inbred mouse strains, focusing on approximately 6,000 VR genes. A consensus of several base-calling algorithms was used to produce a set of more than 65 million SNPs across the whole genomes of 17 strains. These include over 24,000 coding SNPs and result in around 700 genes with potentially truncating mutations. Analysis of VR genes found that wild-derived strains such as CAST/EiJ and Spretus/EiJ, have an order of magnitude more variation than classical strains. The SNP content across VR genes varies significantly in the 13 classical strains, but conserved correlations with chromosomal loci and phylogeny are evident, suggesting VR clusters may delineate functional units. For example, genes in the V2RB subfamily show almost no variation while other clusters, such as V2RA5, have a significant increase in SNPs. We also identified potentially truncating mutations in VR genes, finding that up to 10% of VRs in C57BL/6J may be non-functional in some divergent strains. The functional VR repertoire is therefore likely to vary considerably between mice; accordingly, many C57BL/6J VR pseudogenes may be active in other strains. We are using these data, with parallel analyses of structural genomic variation and global VR expression profiling, to assist us in assigning function to vomeronasal receptors. Acknowledgements: Wellcome Trust, Medical Research Council

#P163 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Identification of Relevant Olfactory Receptors to be used as Sensing Elements of a Bioelectronic Odorant Detection Device

Julien J. Daligault[1,2], Aurélie Dewaele[1,2], Marie-Annick Persuy[1,2], Patrice Congar[1,2], Christine Baly[1,2], Roland Salesse[1,2], Guillaume Launay[3], Stéphane Téletchéa[4], Jean-François Gibrat[3], Edith Pajot-Augy[1,2]
[1]INRA U1197 Neurobiologie de l'Olfaction et Modélisation en Imagerie 78350 Jouy-en-Josas, France, [2]IFR144 Neuro-Sud Paris, France, [3]INRA UR1077 Mathématique, Informatique et Génome 78350 Jouy-en-Josas, France, [4]INSERM U957 Laboratoire de la Physiopathologie de la Résorption Osseuse 44035 Nantes Cedex, France

In the BOND (Bioelectronic Olfactory Neuron Device) project, we use olfactory receptors (ORs) carried by nanoscale liposomes as functional sensing elements of an electrochemical array device, to detect odorants of interest for a given application on the marketplace. This project relies on a European consortium with multidisciplinary expertise on converging bio, micro/nano and information technologies to develop an integrated bioelectronic nanoplatform. The principle is to use the high sensitivity of ORs to specifically detect and/or evaluate odorant molecules at low concentrations in biological samples. In our Workpackage, we look for relevant ORs by using calcium imaging on dissociated cells from rat olfactory epithelium. Neurons responding to an odorant of interest are determined, and individually collected by whole cell content aspiration through a patch pipette. Their mRNA are prepared and retro-transcribed, and the unique sequence of the OR expressed in each neuron is amplified by

nested-PCR using degenerate primers for identification (single neuron RT-PCR). This technique has led in our hands to the determination of several receptors, cloned and wholly sequenced. We compare the 3D structures of the various ORs relevant for the same odorant to rank them relative to their binding efficiency. The next step is the production and functional evaluation of ORs in an efficient heterologous system (yeast), allowing the preparation of nanoscale liposomes to be immobilized onto nanoelectrodes. We carefully examine the addressing and functional properties of ORs in various cell membranes fractions for these newly identified ORs, as compared to previously studied ORs. Odorant binding to the ORs carried by the nanosomes can then be followed by means of electrochemical impedance spectroscopy. Acknowledgements: This work was supported by the 7th Framework Programme of the European Community, Theme Nanosciences, Nanotechnologies, Materials and new Production Technologies, Grant Agreement #228685-2, BOND European Consortium http://bondproject.org/

#P164 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Odorant receptor gene *OR7D4* and perception of the musky odorant Galaxolide

Antti Knaapila, Daniel Hwang, Anna Lysenko, Amin Khoshnevisan, Charles J Wysocki, Danielle R Reed
Monell Chemical Senses Center Philadelphia, PA, USA

Galaxolide™ is a synthetic musk commonly used in cosmetics, but some individuals are unable to perceive its odor. In a previous study, heritability of about 30% has been estimated for sensitivity to Galaxolide, but no specific gene underlying the variability has been pinpointed. Similarly, some describe androstenone (a steroid in sweat and the boar taint odor in pork) as musky, while others as urinous or odorless. Heritability of sensitivity to androstenone is also about 30%, and a gene underlying the variation, *OR7D4*, has been identified. Here we explored whether the previously reported association between alleles of *OR7D4* (SNP rs61729907, R88W) and perception of the odor of androstenone also holds for Galaxolide. A total of 226 individuals (including 100 mono- and 13 dizygotic twin pairs, 21-80 years of age, 79.6% women) sniffed Galaxolide (5% wt/wt), androstenone (0.05% wt/vol) and cinnamon (powdered cinnamon; non-musky control), rated whether they smell something (yes/no), and, if yes, rated liking and intensity using 8 cm visual analogue scales. The odor of Galaxolide, androstenone, and cinnamon was not detected by 24%, 49%, and 0% of subjects. Women rated cinnamon as more intense ($p < 0.05$) and liked it more ($p < 0.001$) than did men. Individuals with the RR genotype for *OR7D4* detected androstenone more frequently than individuals with RW or WW genotype ($p < 0.001$), but this did not apply to Galaxolide or cinnamon. Likewise, individuals with the RR genotype rated the intensity of androstenone, but not Galaxolide or cinnamon, as higher than those with the other genotypes ($p < 0.05$). No association between *OR7D4* and liking of any of the odors was found. The results suggest that *OR7D4* is associated specifically to androstenone and is not related to other musks like Galaxolide.

#P165 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Identification of Odorant Receptors Discriminating Enantiomers of Odorants

Yoshiki Takai, Kazushige Touhara
The University of Tokyo Tokyo, Japan

Enantiomers have identical chemical and physical properties except for their ability to rotate plane-polarized light. However, it is known that some enantiomers elicit different smell. Recent studies show that there exist odorant receptors (ORs) that respond only to one enantiomer and that respond to both enantiomers. However, ORs that discriminate enantiomers have not been fully characterized. In order to identify ORs that recognize optically active odorants, we adopted an *in vivo* OR identification technique that allowed for isolating an OR gene that was expressed in olfactory sensory neurons based on olfactory glomerulus activity. We exposed mice to highly optical pure *l*- and *d*-menthol as a pair of enantiomers, and performed Ca^{2+} imaging of the olfactory bulb. We first analyzed the activity of each glomerulus that responded to *l*- or *d*-menthol against a panel of odorants that varied in the chemical structure, and classified them for their ligand specificity using a cluster analysis. Next, we cloned several OR genes from glomeruli that responded to menthol. Each cloned OR was expressed in HEK293 cells and we reconstituted the menthol responses. We confirmed that these ORs responded to *l*- and/or *d*-menthol with different thresholds. We will also discuss the relationship between ligand specificity and OR sequences.

#P166 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Become a virtual ligand for a day: elucidating olfactory receptor binding-pocket interactions through enhanced visualization techniques

Peter C. Lai, Chiquito Crasto
Genetics Division of Research, University of Alabama at Birmingham Birmingham, AL, USA

From an electrostatic and steric (Van der Waals) interactions context, what do odor-ligand molecules "perceive" when they encounter the extracellular solvent-accessible surface of an olfactory receptor? Studying the steric surface and electronic character of all potential ligand binding sites is crucial to understanding binding selectivity, affinity, and ability to activate. Displaying the results of a binding pocket survey using conventional 2D graphical or tabular formats may present difficulties with data interpretation. A more direct visual approach of inspecting the interior of the binding pocket may be beneficial. We demonstrate this technique on a set of two distinctly different but possible models of hOR17-210, a receptor that has been shown to be variably functional and pseudogenic in humans, and is missing a TM domain (Lai *et al*., 2008 *J. Struct. Funct. Genomics*). The University of Alabama at Birmingham Enabling Technology laboratory has developed VisMini as an open-source and easily transportable version of Visbox, Inc.'s VisBox and VisCube immersive virtual reality 3D display system. This

advanced 3D-visualization platform implements 2 coaxial projectors to display a 3D image of our receptor models with enhanced depth perception provided by user-worn polarized glasses and six degrees of freedom interaction provided by a handheld analog controller device. We adapt the system for use here in developing the latest in molecular model visualization capability as compared to current desktop-style model renderers. Acknowledgements: Supported by NIDCD DC011068

#P167 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Excitatory response in ORs is mediated by odorant-binding in preferred binding regions

Chiquito J. Crasto[1], Brandon Guida[2,3], Peter C. Lai[1,4], Jing Shi[4]
[1]Department of Genetics, University of Alabama at Birmingham Birmingham, AL, USA, [2]Department of Biology and Environmental Science, University of New Haven West Haven, CT, USA, [3]Center for Infectious Diseases and Vaccinology, Arizona State University Tempe, AZ, USA, [4]School of Engineering, University of Alabama at Birmingham Birmingham, AL, USA

Experimental functional analysis of olfactory receptors at a mechanistic level is challenging. The specificity by which thousands (or combinations) of odors interact with a fixed number of functional ORs to effect olfaction is unknown. There exists no predictive method by which specific odorants might be identified *a priori* as eliciting excitatory responses from specific ORs. The results of a combinatorial functional analysis experiment for several mouse ORs showed varied responses to odorant ligands which differed in chain length, functional group and degree of unsaturation. The responses ranged from strongly excitatory at dilute odorant concentrations to no response at relatively high odorant concentrations. (Malnic et al., Cell [1999]). We performed computational OR protein modeling and odorant ligand binding for two mouse ORs, S79 and S86, involved in the combinatorial studies. Our protein modeling and odorant binding results showed that both receptors have two preferred binding sites: one bound by transmembrane (TM) helices I, II, III and VII, and the other bound by TMs III, IV, V and VI. The excitatory odorants, heptanoic acid, nonanedioic acid and octanoic acid (S79) and nonanoic acid (S86) preferentially bind in the region bound by TMs III, IV, V and V. The non-excitatory odorants heptanol (S79) and heptanoic acid (S86) show greater likelihood of binding in the region bound by TMs I, II, III and VII. Odorant interactions within preferred binding regions of an OR are a possible means by which excitation and non-excitation or inhibition might be elucidated. Acknowledgements: 1R21DC011068-01 (NIDCD)

#P168 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Influence of Tags on ORs Functional Properties in HEK293 cells

Alex Veithen, Françoise Wilkin, Magali Philippeau, Pierre Chatelain
TecnoScent S.A. Brussels, Belgium

The difficulty of expressing olfactory receptors (ORs) in heterologous cell systems has been a major hurdle for the deorphanization of this family of receptor. The tagging of ORs in their N-terminal moiety has been achieved to allow the detection of ORs and to improve their cell surface and functional expression. For example, the addition of the first 21 amino acids of the rhodopsin receptor (a.k.a. Rho tag), has been successfully used to improve the functional response of mouse and human ORs. This strategy remains however questionable since additional stretches of amino acids in the N-terminal part of ORs might interfere with the ligand binding and lead to changes in potency and/or efficacy or to a modification of the receptor selectivity. We investigated the influence of tags on the functional expression of two deorphanized ORs that respond with a different selectivity to a wide range of ligands. The human receptor ORL420 was tagged with the Rho tag, the 5 amino acids F5 tag or the 45 amino acids SSTR3 tag. Likewise, the mouse receptor mOREG was tagged with either the Rho tag or the F5 tag. The functional response of the different tagged versions were assessed in HEK293 cells using either a gene reporter assay or a calcium imaging assay and compared to these of the native OR. Results show that the potency of the tested ligands remains unaffected by the presence of tags. Similarly the ranking of the potency of ligands in each series is not modified. In contrast, the efficacy of the response can be modified by addition of a tag, reflecting the previously reported positive effect of the tag on the targeting of the OR to the cell surface. Based on this analysis, we conclude that tagging ORs is a suitable strategy for improving the functional expression of ORs without affecting the binding properties. Acknowledgements: This work is supported by the Brussels Region

#P169 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

MOR118 Cytoplasmic Tail Suppresses Endogenous Odorant Receptor Expression in Cultured Olfactory Sensory Neurons

Huaiyang Chen, Qizhi Gong
Department of Cell Biology and Human Anatomy, School of Medicine, University of California Davis, CA, USA

In mouse olfactory system, there are about 1000 odorant receptors (OR) responsible for distinguishing immense number of different odorants. Each olfactory sensory neuron (OSN) expresses only one odorant receptor. OR proteins are believed to play a critical role in the maintenance of the selected OR expression. The regulatory mechanism of OR selection and maintenance in OSNs is still unclear. With our established dissociated OSN culture system, we observed that exogenous OR expression, introduced by lentiviral mediated gene transfer,

suppresses endogenous receptor expression. To define whether any specific domain of OR sequence is responsible for suppressing endogenous OR transcription, we expressed MOR118 cytoplasmic fragments in cultured OSNs. Truncated T-cell surface glycoprotein CD4 precursor (CD4) was fused with different MOR118 cytoplasmic fragments to target them onto OSN membrane. Chimeric OR fragments were introduced to cultured OSNs by lentiviral mediated infection and endogenous OR expression was evaluated and compared with GFP expressing culture. We observed successful membrane localization of CD4-MOR118 fragments in cultured OSNs as early as 3 days post-infection. When MOR118 cytoplasmic terminus (292-313aa) was expressed in cultured OSNs, we saw significant suppression of endogenous OR expression, evaluated by qRT-PCR. Eighty percent of endogenous P2 receptor expression was suppressed at 5 days post lentiviral infection when compared to the expression level of GFP infected OSN culture, while 75% of endogenous MOR263-4 expression was suppressed. The function of three MOR118 cytoplasmic loops (47-58aa, 119-138 aa and 221-240aa) was also investigated and the effect of these fragments on endogenous OR expression is being validated.
Acknowledgements: NIH/NIDCD DC010237

#P170 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Homo-Oligomerization of a Mammalian Olfactory Receptor: a BRET Study Demonstrates a Correlation between Activity and Conformational State

Edith Pajot-Augy[1], Fallou Wade[1], Agathe Espagne[2], Marie-Annick Persuy[1], Jasmina Vidic[3], Régine Monnerie[1], Fabienne Merola[2], Guenhael Sanz[1]
[1]INRA UR1197 Neurobiologie de l'Olfaction et Modélisation en Imagerie 78350 Jouy-en-Josas, France, [2]Laboratoire de Chimie Physique UMR 8000 U. Paris-Sud 11 and CNRS 91405 Orsay, France, [3]INRA UR892 Virologie et Immunologie Moléculaire 78350 Jouy-en-Josas, France

Olfactory receptors (ORs) are G protein coupled receptors (GPCRs). GPCR oligomerization plays many roles in receptor trafficking and functional response. Even though heterodimerization of ORs was reported, mammalian ORs homodimerization is still not elucidated, nor the relationship between OR activation and association/conformation state. The bell-shaped dose-response curve for receptor functional response in the absence of OBPs (odorant binding proteins) prompted us to propose a model for OR-OBP-odorant interaction, based on two hypotheses : ORs homodimerization, and competitive binding of odorant and OBP to the receptor. We already elucidated OBP specific role in maintaining OR activity at high odorant concentration. Investigation of ORs oligomerization was then required to fully validate our model. We therefore explored mammalian ORs homo-oligomerization and its involvement in receptor functional response using co-immunoprecipitation and bioluminescence resonance energy transfer (BRET) approaches on membrane fraction or subfractions from yeast heterologously expressing hOR1740. We clearly demonstrate OR1740 self-association into dimers starting early in the biosynthetic pathway, and therefore constitutive dimer expression at the plasma membrane. Furthermore, odorant binding modulates BRET levels, suggesting a conformational rearrangement of receptor dimers related to their activation state. This variation exhibits a bell-shaped curve with odorant concentration, which correlates with that of the functional response. We ascribe the decrease of both measurements at high concentration to ligand-mediated protomers conformational changes, resulting in dimer inactivation. This validates our model in which ligand binding site occupancy rate affects the dimer activity level.
Acknowledgements: This work was funded by INRA and Agence Nationale de la Recherche (NOSE, ANR-07-PCVI-0027-01)

#P171 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Odorant receptor expression in the olfactory epithelium of β3GnT2 mice

Tom Knott, Pasil Madany, Tim Henion, Ashley Faden, Gary Schwarting
University of Massachusetts Medical School Department of Cell Biology Worcester, MA, USA

The glycosyltransferase β3GnT2 is highly expressed in olfactory neurons where it regulates poly-lactosamine glycan synthesis on several glycoproteins involved with olfactory axon guidance and signal transduction. Null mice for β3GnT2 have severe axon guidance defects. To identify potential changes in gene expression associated with this phenotype we analyzed mRNA isolated from β3GnT2 mutant mice and wild-type littermates using Affymetrix mouse gene arrays. These chips contain over 34,000 unique target sequences including probes for more than 600 mouse odorant receptors. Preliminary studies suggest that the expression levels in the majority of OR genes are similar in wild-type and null OEs. However, about 5% of OR genes are significantly up-regulated and approximately 30% of OR genes appear to be down-regulated in the olfactory epithelium of β3GnT2 null mice. Comparison of wild-type and null mice by in situ hybridization revealed that the number of OR37+ and M72+ neurons is unchanged, whereas the number of P2 neurons is decreased in nulls by about 75%, and the number of OR-15+, OR43+, and OR137+ neurons is increased by between 40 and 60%. In addition, mapping studies reveal that in some cases axons of OR-expressing neurons that are reduced in the OE cannot be detected in glomeruli of the olfactory bulb, suggesting that null mice have a significantly reduced OR repertoire. Of the remaining ORs examined, M72, OR37 and OR15 axons innervate inappropriate targets in the OB. In spite of these significant changes in OR expression and axon targeting, adult β3GnT2 null mice were able to complete an odor discrimination task with a greater than 80% efficiency compared to wild-type littermates.
Acknowledgements: Supported by NIH grants DC00953.

#P172 POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION

Ectopically Expressed Olfactory Receptors in Human Primary Skeletal Muscle Cells

Markus Osterloh, Elena Guschina, Hanns Hatt
Ruhr University Bochum 44801 Bochum, Germany

Olfactory receptors (ORs) comprise the largest group of GPCRs and are responsible for the recognition of different odorant molecules. Despite the original "dogma" that ORs in mammals are only expressed in the olfactory epithelium it has been shown in recent years that olfactory receptors are widely expressed among different tissues throughout the body in various organisms. In contrast to their mere expression the physiological functionality of ORs has been shown for only a few candidates e.g. OR1D2 (chemotaxis in sperm) and OR51E2 (inhibition of proliferation in prostate cancer cells). Human skeletal muscle cells also express a number of ORs which so far has been demonstrated by microarray analysis and RT-PCR. We could show via large scale microarray analysis and RT-qPCR that human primary skeletal muscle cells (SkMC) express a large variety of ORs. Additionally in calcium imaging experiments, SkMCs responded to stimulation with specific odor mixtures by showing an intracellular calcium increase in a concentration dependent manner. Interestingly the responses were also dependent on the developmental state of the cells showing a significant difference between undifferentiated and differentiated cells. We could further demonstrate the expression of olfactory signaling cascade proteins in skeletal muscle cells via western blot analysis.

#P173 POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION

Olfaction in other tissues

Na-na Kang, Jae Hyung Koo
DGIST Daegu, Korea

Animals have evolved chemosensory systems that are able to discriminate environmental stimuli and transmit it into the brain. There, the signals summate, integrate, and generate cognitive and behavioral responses. Olfactory sense is mediated by specialized olfactory receptor neurons (ORNs) that contain unique molecules (OMP, Golf, and AC3) which are mainly present in the olfactory epithelium. However olfactory signaling molecules, such as Golf and AC3, have recently been reported outside of the olfactory system, suggesting that the olfactory sense may play a role in other tissues. Little is known about the systematical analysis of olfactory components throughout the whole body. This study demonstrates whether olfactory signaling is present in other tissues by systemically using microarray databases, transgenic mice, and immunohistochemical analysis. Gene expression of OMP, Golf, and AC3 was noticeable in several other tissues by the analysis of microarray databases. Among them, tissues related to food digestion and the heart showed high levels of olfactory signal molecules compared to other tissues. A summary of the results and future directions will be investigated and discussed.
Acknowledgements: This work was supported by MEST & DGIST (10-BD-04, DGIST Convergence Science Center)

#P174 POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION

Electrophysiological Evidence for Multiple Amino Acid Olfactory Receptor Sites in Elasmobranchs

Tricia L. Meredith, Stephen M. Kajiura
Florida Atlantic University, Biological Sciences Boca Raton, FL, USA

Marine environments contain a vast number and variety of dissolved chemicals that may be detected by the olfactory systems of the resident organisms. Odorants are detected when they bind to receptors on olfactory receptor neurons (ORNs). Each olfactory receptor can detect multiple odorants and each odorant can be detected by multiple olfactory receptor types; therefore, the olfactory discrimination of odorants results from the stimulation by different *combinations* of olfactory receptors. Cross-adaptation experiments with teleosts have confirmed the existence of multiple olfactory receptor types that detect particular groups of odorants. These experiments aim to determine whether two agonists interact with the independent or overlapping receptor populations. Between four and six receptor types have been identified for amino acids based on the side-chain structure. While the presence of multiple receptor types has been shown in teleosts, it is unknown whether elasmobranchs possess similar receptor types. To test this hypothesis, we performed cross-adaptation experiments with two distantly related elasmobranch species, the lemon shark (*N. brevirostris*) and Atlantic stingray (*D. sabina*) ($n \geq 8$ each). We tested the electro-olfactogram (EOG) responses of individuals to ten test stimuli delivered separately over five different background or adapting stimuli that each varied in side-chain structure. Under all five adapting regimes, the test EOG responses were reduced from those during the unadapted state, and some differences among them resulted depending on the molecular structure of the adapting amino acid. Our preliminary results indicate potentially separate receptor types for neutral, basic, and aromatic amino acids. Further testing may reveal additional amino acid receptor types.

#P175 POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION

Using Imaging to Measure Lobster Peripheral Olfactory Coding

Yuriy V. Bobkov[1], Kirill Y. Ukhanov[1], Elizabeth A. Corey[1], Barry W. Ache[1,2]
[1]Whitney Laboratory, Center for Smell and Taste, and McKnight Brain Institute, University of Florida Gainesville, FL, USA, [2]Depts. of Biology and Neuroscience, University of Florida Gainesville, FL, USA

Recent findings [e.g., Stopfer et al., 2009] suggest that ORNs are capable of generating odorant-specific temporal patterns of responses that potentially provide a basis for spatiotemporal odor coding. As a first step towards understanding whether lobster ORNs are capable of producing stimulus-dependent temporal activity patterns and the cellular mechanisms that might underlie such heterogeneity, we developed a calcium imaging approach to

estimate population activity in lobster ORNs. The somatic calcium signal (Ca_i) faithfully reflects specific ORN activity, i.e., Ca_i increases in a concentration-dependent manner, Ca_i is ligand-specific, and Cai is sensitive to pharmacological probes known to potently suppress and/or modulate the electrophysiological responses of the ORNs. Simultaneous extracellular recording from imaged ORNs shows that the dynamics of spontaneous and odorant-evoked Ca_i matches that of the electrophysiological activity of the ORNs, and allows distinguishing several basic functional subpopulations of ORNs based on their spontaneous activity and response pattern evoked by brief complex odor stimulus, including tonic (both excitatory or inhibitory responses), phaso-tonic, and bursting ORNs. In addition, we show that different stimuli can elicit responses of different polarity within the same ORN. These findings support the hypothesis of stimulus-dependent response patterning in ORN output.
Acknowledgements: Supported by grants from the National Institute on Deafness and Other Communication Disorders.

#P176 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Anatomical and Histological Analyses of the Olfactory Epithelium of the Domestic Cat (*Felis catus)*
Karen K. Yee[1], Fritz W. Lischka[1], Mark E. Haskins[2], Charles J. Wysocki[1], Nancy E. Rawson[3], Blaire Van Valkenburgh[4]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]Department of Pathobiology, School of Veterinary Medicine, University of Pennsylvania Philadelphia, PA, USA, [3]AFB International St Charles, MO, USA, [4]Department of Ecology and Evolutionary Biology, University of California, Los Angeles Los Angeles, CA, USA

The molecular and cellular characteristics of the olfactory epithelium demonstrate remarkable conservation across many species. However, different species acquire chemical information about different aspects of their environment and they use this information in different ways. Species adaptations have evolved to suit these differing needs. Few studies have described the anatomy and biology of the feline olfactory system. Among cats, chemosensory information is used in social interactions, territory identification, and tracking of prey. Because cats are obligate carnivores, documentation of olfactory neurobiology in the feline can inform our understanding of the evolution of this behaviorally important sensory pathway. Here we present a study of the nasal anatomy of the adult, domestic, shorthaired cat at macro- and microscopic levels. Non-sensory and sensory epithelia were defined by thickness, presence or absence of goblet cells, distinctive epithelial morphology and immunohistochemical staining with neuronal markers. Rostral extent and distribution of olfactory epithelium on the ethmoturbinates and septum were determined. We determined that the extensive and intricately scrolled turbinate system is populated by sensory neuroepithelium, which is present on regions of the ethmoturbinates, septum and nasoturbinates. In an unusual arrangement, that may reflect their flattened facial structure as compared with rodents or most canines, the olfactory bulbs, although encased within the skull, extend partly into the nasal cavity, with open turbinate structures both below and above the anterior extension of the bulbs. Further study of the impact of environmental, social and/or dietary demands on chemosensory anatomy should reveal principles of evolution that may be more widely applicable. Acknowledgements: We thank the National Science Foundation (NSF0421927 to BVV) and the National Institute of Health (DK25759 and RR02512 to MEK) for financial support of this work

#P177 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Rat EOG response to nasal flow rate change predicts response distribution and retronasal response
John Scott, Lisa Sherrill
Emory University/Cell Biology Atlanta, GA, USA

Odorant solubility and polarity correlate with the growth of response as nasal flow rate increases (Flow-Slope), with response distribution, and with the size of retronasal odorant response. These facts led us to compare Flow-Slope and response distribution in the same animals. We measured orthonasal electroolfactograms (EOG) to an expanded dataset (62 odorants) with several odorants we had not tested before. Response distribution was evaluated as the normalized difference between medial and lateral EOG (Response-Difference). We recorded from the dorsomedial recess and the dorsal region of the lateral recess in 58 rats killed with overdose of pentobarbital (100mg/kg) in a protocol approved by the Emory University IACUC. The mean Flow-Slope for the medial and lateral sites was calculated for each odorant (N=3 or more rats). The medial ($R=0.82$, $p<.01$) but not the lateral ($R=0.18$, $p>0.05$) Flow-Slope correlated with the Response-Difference. This difference reflects the more complicated path through the lateral epithelium. The medial Flow-Slope correlation was stronger than with predicted solubility properties (e.g. with air-water partition coefficient $R= -0.31$, $p<.02$). It may indicate either nonlinear solubility effects on sorption or specific protein contributions (enzymatic or odorant binding) on the transport of particular odorants through the nasal cavity. Retronasal responses correlated with the medial Flow-Slope ($R= -0.54$, $p<.01$) for the 45 odorants tested, which was also stronger than the correlation with solubility properties. The correlation was even stronger for an exponential function of medial Flow-Slope ($R=0.90$). The results indicate that transit of odorants through the nasal cavity partly governs response distribution and suggests that the controlling factors may be complex. Acknowledgements: Supported by NIH grant DC 008648

#P178 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Active sensing in the olfactory system: testing the sorption hypothesis in the awake animal

Tristan Cenier[1,2], Yusuke Tsuno[1], John McGann[1], Matt Wachowiak[1,2]
[1]Department of Biology Boston, MA, USA, [2]Department of Physiology Salt Lake City, UT, USA

Odor sampling - sniffing - is strongly modulated in the behaving animal. One hypothesis, first articulated by Mozell et al., is that sniff magnitude - as defined by peak intranasal airflow - can shape olfactory receptor neuron (ORN) responses by differentially affecting the way in which odorants absorb onto the olfactory epithelium, depending on their chemical properties. Thus, animals may actively modulate sniff magnitude in order to optimally sample odorants. Here, we test this hypothesis by imaging from ORNs and monitoring sniff flowrate in the awake rat. Presynaptic calcium imaging in awake, head- fixed rats was performed as described previously (Verhagen et al. Nat Neurosci 10:631). Sniff flowrate was measured using an intranasal thermocouple. The sorption hypothesis predicts that the ORN response to strongly sorbed odorants will increase with flowrate while it will decrease or not change for weakly sorbed odorants. However, we found that peak flowrate was poorly correlated to ORN response magnitude in the behaving animal and that this correlation was weak for both high and low-sorption odors (mean r^2=0.04). We also tested whether sniff flowrates changed as rats discriminated either strongly- or weakly-sorbed odorants of progressively decreasing concentration. We found little evidence that rats actively modulate sniff magnitude for either odorant set. In contrast, in anesthetized animals, using artificial inhalation, we found that flowrate and response amplitude were well correlated (r^2=0.86), although this correlation was high for both strongly and weakly sorbed odors. Thus, overall we find that sniff flowrate has little effect on ORN response magnitude during natural odor sampling, and we find little evidence that rats modulate this parameter of sniffing while performing odor discriminations. Acknowledgements: Supported by NIH DC006441.

#P179 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

The Mouse Lateral Olfactory Tract

Peter C. Brunjes
University of Virginia Charlottesville, VA, USA

The olfactory peduncle connects the olfactory bulb (OB) with the forebrain and contains the anterior olfactory nucleus/cortex (AON) as well as the anterior portion of the lateral olfactory tract (LOT). Axons of OB projection neurons (mitral and tufted cells) course through the LOT, innervating the AON and continuing caudally to the olfactory tubercle and piriform cortex. Axons from the dorsal OB traverse the deep layers of the accessory OB before collecting and entering the LOT from the dorsal side. Remaining fibers enter the tract from the ventral side. Electron microscopy was used to reconstruct a coronal section of the LOT in the anterior peduncle at 800 X. Then, 11 equally-spaced, 50 µm-wide strips of the tract extending from the pial surface to deep regions where axon density dropped were sampled, yielding a data set of over 13,300 profiles of myelinated axons. The population was sampled to visualize the distribution of the largest and smallest caliber axons and axons with elongated shapes (and thus traveling obliquely to the plane of section). Each feature was found to be differentially distributed along the dorsal to ventral axis of the tract. The 11 sample areas were also divided into deep (A), middle (B and C) and superficial (D) bands to examine structure within this dimension in the LOT. Once again, results indicate that the tract is not uniform. For example, large area profiles are more prevalent in the superficial (C and D) regions of the LOT, both small and long profiles are less abundant in the pial-most quarter (D) of the tract, and axon density was highest in region B. Taken together, the results indicate that the LOT has a differential internal organization that may be important to understand in order to grasp mechanisms of olfactory information processing. Acknowledgements: Supported by Grant DC000338 from NIH (NIDCD)

#P180 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

GABAergic populations in the mouse anterior olfactory nucleus/cortex

Rachel B. Kay, Peter C. Brunjes
University of Virginia Dept of Psychology Charlottesville, VA, USA

The anterior olfactory nucleus (AON) is a central olfactory cortical structure residing between, and reciprocally connected to, the olfactory bulb and piriform cortex. Relatively few studies have described the area's organization, especially given the AON's key position in the olfactory circuit. For example, while the heterogeneous nature of GABAergic neurons has been elucidated in both the bulb and piriform cortex, AON interneurons have received scant attention. The present study mapped the distribution and number of subtypes of GABA-releasing interneurons in the AON to test the possibility of differential expression within internal subdivisions of the structure and to compare the AON to other olfactory regions. Tissue from adult GAD67-GFP transgenic mice underwent fluorescent immunolabeling to quantify coexpression of several molecular markers including calbindin, calretinin, parvalbumin, cholecystokinin, neuropeptide Y, somatostatin, and vasoactive intestinal peptide. Initial results indicate that all the molecular markers are present within the population of GAD67 cells, but each is expressed in different proportion of the cells. Approximately 42% (±6) of GAD67 cells coexpress calbindin, 23%(±5) coexpress parvalbumin, and 22%(±4) coexpress calretinin, with the other markers together accounting for less than 13% of the GAD67 population. Additionally, some markers showed deep-to-superficial and regional differences in expression. Double labeling indicated that combinations of two molecular markers in single GAD67 cells also varied widely. While further works needs to be done, the results suggest a heterogeneous population of GABAergic interneurons in the AON similar to that reported for the piriform (e.g., Suzuki & Bekkers, J. Comp. Neurol. 2010) and other cortical areas.

#P181 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Quantitative analysis of interneuron diversity within the mouse main olfactory bulb external plexiform layer

Bao-Feng Ma, Yu-Feng Wang, Kathryn A. Hamilton
LSU Health Sciences Center, Cellular Biology & Anatomy Shreveport, LA, USA

The external plexiform layer (EPL) of the main olfactory bulb (OB) contains Van Gehuchten and other interneurons (INs) that are morphologically diverse. To identify GABAergic IN subtypes, we used whole-cell patch clamp recording methods to label single INs in OB slices from GAD65-GFP transgenic mice. We then collected two-photon confocal z-stack images of the INs. The images were imported into Neurolucida software for 3-D reconstruction and quantification of morphological parameters, which described process complexity, soma size, and soma depth within the EPL. Next, we performed principal component analysis and hierarchical cluster analysis of the principal components. The results revealed three morphologically distinct groups of INs. Group 1 INs had smaller somata, fewer branches, and fewer varicosities than Group 2 or 3 INs. Group 3 INs branched more extensively and had a higher density of varicosities than Group 1 or 2 INs. Group 2 INs exhibited intermediate characteristics. We used three methods to validate the three IN groups: 1) Silhouette analysis showed that each interneuron was correctly assigned to its cluster on the basis of a dissimilarity matrix; 2) Plots of the first two principal components or canonical variables demonstrated clear separation between the groups; 3) Nonhierarchical cluster analysis using the K-means method produced 86.7% agreement (13/15 cells) with the hierarchical cluster analysis. These results indicate that GABAergic EPL INs of GAD65-GFP mice can be classified as three morphological subtypes. The results also support the hypothesis that different EPL IN subtypes perform different inhibitory functions. Acknowledgements: Supported by NIH DC007876.

#P182 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Activation of Metabotropic Glutamate Receptors (mGluRs) Enhances Persistent Sodium Currents in External Tufted Cells (ETCs)

Hongwei Dong, Qiang Nai, James C Davis, Matthew Ennis
Department of Anatomy and Neurobiology, University of Tennessee, HSC Memphis, TN, USA

Previous studies demonstrate that activation of Group I mGluRs with DHPG evoked an inward current and enhanced rhythmic bursting in ETCs (Dong et al 2009). These effects were due in part to activation of a non-selective cation current (I_{CAN}). However, I_{CAN} blockade attenuated, but did not eliminate, DHPG-evoked inward currents or the enhancement of ETC rhythmic bursting. In the present study, we used patch clamp electrophysiology in rat olfactory bulb slices to investigate if mGluRs also modulate the persistent sodium current (I_{NaP}) that plays a key role in generating rhythmic bursting in ETCs. I_{NaP} was isolated by applying a slow voltage ramp (-80 mV to -40 mV, 30 mV/s) in the presence of CNQX, APV, gabazine, TEA (10 mM), cadmium (200 mM) and nickel (1 mM). This elicited a TTX-sensitive voltage-dependent inward current with an activation threshold of -60 mV, as previously reported. DHPG application significantly increased the amplitude of I_{NaP} by -15.6 ± 5.7 pA (HP = -55 mV); DHPG simultaneously evoked a voltage independent I_{CAN} in ETCs. However, the enhancement of I_{NaP} by DHPG was not related to its effect on I_{CAN} as the increase in I_{NaP} persisted using a BAPTA-based internal solution that eliminated the I_{CAN}. Riluzole blocked the effect of DHPG on I_{NaP} but had no effect on the I_{CAN}. The Group II mGluR agonist L-CCG-I modestly increased I_{NaP} by -7.4 ± 4.9 pA, but had no effect on the I_{CAN}. The Group III mGluR agonist L-AP4 was without effect. In current clamp recordings using a BAPTA-based internal solution, DHPG significantly enhanced ETC rhythmic bursting. The results demonstrate that activation of mGluRs enhances ETC excitability and rhythmic bursting via modulation of multiple currents. Acknowledgements: PHS Grants: DC009049 and DC003195

#P183 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Modulation of mitral cell spikes by dendrodendritic synapse location

Thomas McTavish[1], Michele Migliore[2], Michael Hines[3], Gordon Shepherd[1]
[1]Dept. of Neurobiology, Yale University New Haven, CT, USA, [2]Institute of Biophysics, National Research Council Palermo, Italy, [3]Dept. of Computer Science, Yale University New Haven, CT, USA

On their lengthy lateral dendrites, mitral cells of the olfactory bulb form dendrodendritic synapses with granule cell interneurons. The dendrodendritic synapse operates such that backpropagating spikes in the lateral dendrites activate granule cells, and the granule cells inhibit the mitral cells. In this study, we continue our efforts to analyze signal modulation in the olfactory bulb using computational models of the mitral-granule circuit focusing on the spatial arrangements of the dendrodendritic synapses. Utilizing networks of 5 mitral cells and 100 granule cells, we demonstrate that mitral cells can synchronize at arbitrary distances as long as each mitral cell has a cluster of granule cells centered about its soma and that each mitral cell being activated also makes synapses with this cluster. Another constraint for synchrony is that the input magnitude must be balanced. When adjusting the input magnitude driving a particular mitral cell relative to another, the mitral-granule circuit induces a phase shift or delay in the weaker-driven cells. This phase shift using asymmetric inputs is absent when the granule cells are removed from the model or when the synaptic locations are randomized. Collectively, our results indicate that the spatial locations of dendrodendritic synapses of the mitral-granule operate to modulate the timing of mitral cell spikes. Acknowledgements: NIH/NIDCD R01 DC 009977

#P184 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Nitric oxide modulates juxtaglomerular neuron spiking in the mouse main olfactory bulb through cGMP mediated signaling

Ambarish S. Ghatpande, Graeme Lowe
Monell Chem Senses Ctr Philadelphia, PA, USA

Neuronal nitric oxide synthase is prominently expressed in the mammalian main olfactory bulb (MOB), yet the physiological role of nitric oxide (NO) in bulbar signaling is virtually unknown. In order to understand the roles played by NO in bulbar processing of olfactory signals and the underlying cellular mechanisms, we performed cell-attached recordings from juxtaglomerular (JG) neurons in olfactory bulb slices from wild-type (WT) and transgenic *gucy1a3* mice that co-express EGFP along with soluble guanylate cyclase (sGC), the major physiological NO receptor. Upon bath application of an NO donor (NOC7), 33% of randomly selected JG cells in MOB slices from WT mice showed increased spike frequency, which reversed on washout of donor. In slices from *gucy1a3* mice, 78 % of JG neurons targeted for recording by EGFP fluorescence showed increased spiking in response to NO. This selective effect of NO on sGC-positive neurons was reversible, repeatable and attenuated by co-application of cPTIO, an NO scavenger, as well as by ODQ, an sGC inhibitor. Increased spiking of green fluorescent JG cells was also observed following direct application of 8-Br-cGMP, a membrane-permeant cGMP analog. Taken together, these results suggest a cGMP-dependent postsynaptic excitatory effect of NO in MOB glomerular circuits. We discuss the cellular mechanisms underlying this effect, and its potential role as a facilitator of synaptic plasticity in bulb circuits for odor learning and memory. Acknowledgements: Supported by: 3R01DC004208-07S1 (GL)

#P185 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Patterned Stimulation of Olfactory Bulb Glomeruli with Light using a Dual-axis Microscope

David C. Willhite[1,2], Tomokazu F. Sato[1], Vikrant Kapoor[1], Venkatesh N. Murthy[1]
[1]Harvard University Cambridge, MA, USA, [2]Yale University New Haven, CT, USA

Transgenic mice expressing the photo-activatable protein channelrhodopsin-2 have greatly expanded the options for stimulus control in physiological studies. Using this method, the neuronal population activated can be selected by promoter-specific expression, and the spatio-temporal aspects of the stimulus can be tightly controlled. Here, we describe a dual-axis microscope design for light activation of olfactory bulb (OB) acute slices. The OBs of transgenic mice expressing channelrhodopsin-2 under the olfactory-marker protein promoter in olfactory sensory neurons (Dhawale, et al., 2010, Nat Neurosci 13:1404) were sectioned coronally for acute slice recordings. Regions of interest (resolution as low as 10 microns) were selected singly and in combinations using custom software. Synaptic currents (both inhibitory and excitatory) were recorded in mitral cells using whole-cell patch clamp electrodes with cesium-gluconate internal solution. Glomeruli were stimulated by a VGA projector from beneath the slice in an optical axis independent of the upright microscope axis used for electrophysiology and fluorescence imaging. Single stimulations and combinations of multiple stimulations were performed, and results will be presented along with the description of the dual-axis design. Acknowledgements: NIH R01 DC011291

#P186 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Benzodiazepines Selectively Increase Brief-Access Licking for Gustatory Stimuli Independent of Influences on Motivational State

David W. Pittman[1], Phillip H. Neill[1], Michael H. Schecter[1], Isaac D. Rankin[1], John-Paul Baird[2]
[1]Wofford College, Department of Psychology Spartanburg, SC, USA, [2]Amherst College, Dept. Psychology & Neuroscience Program Amherst, MA, USA

In addition to their well-known anxiolytic functions, benzodiazepines produce a hyperphagia that is thought to be mediated through enhanced affective reactivity to palatable taste stimuli, although most studies have limited analyses to normally-accepted (e.g., sweet) tastants. Previously, we reported that the benzodiazepine, chlordiazepoxide (CDP), increased consumption of both appetitive and aversive taste stimuli during long-term (1hr) tests, primarily through changes in licking patterns associated with hedonic taste evaluation with no effect on feeding behaviors associated with postingestive feedback. Here, in a complimentary study, we examined CDP effects on responses to a range of taste stimuli during brief access (15s) trials. A counterbalanced within-subject design compared CDP (10mg/kg/ip) and saline effects on licking for water and 4 concentrations of NaCl, Q-HCl, citric acid, MSG, saccharin, ethanol, and capsaicin under water-restricted (23hr) conditions and sucrose, saccharin, and MSG in water-replete conditions (n=16). CDP uniformly increased licking to taste stimuli that were normally-avoided under saline conditions with the important exception of the trigeminal stimulus, capsaicin, which was not affected at any concentration, suggesting a taste-specific effect of CDP on orosensory processing. Under water-replete conditions there was no effect of CDP on water consumption. CDP increased licking to concentrations of normally-accepted stimuli that did not already elicit maximal licking in the saline condition. This research indicates that benzodiazepines do not increase the perceived intensity of taste stimuli. Rather, they selectively enhance the hedonic acceptance of gustatory orosensory stimuli, independent of motivational states such as thirst, appetite, or general anxiolytic effects. Acknowledgements: Supported by Wofford College.

#P187 POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION

Parabrachial benzodiazepine receptor antagonist effects on licking for sucrose

John-Paul Baird[1], Ayla Mansur[1], David Pittman[2]
[1]Amherst College, Dept Psychology & Program in Neuroscience Amherst, MA, USA, [2]Wofford College, Dept. Psychology & Program in Neuroscience Spartanburg, SC, USA

Benzodiazepines such as midazolam induce hyperphagia by enhancing the hedonic acceptance of taste stimuli. Previous studies have implicated the hindbrain parabrachial nucleus (PBN) as a site for benzodiazepine stimulation of hyperphagia (Higgs & Cooper, 1996; Soderpalm & Berridge, 2000). Recent studies also show that benzodiazepine infusions to the PBN reverse potent anorexia induced by ablation of AGRP/NPY/GABA neuron input to the PBN (Wu et al., 2009). To further clarify the role of the PBN in benzodiazepine-mediated modulations of feeding and taste evaluation, we tested the effectiveness of PBN injections of the benzodiazepine receptor antagonist, flumazenil (FLZ). Our first experiment examined the effect of intraPBN FLZ injections (3000, 300, 30, 0 ng/0.4 ul DMSO per side) on licking for 0.2M sucrose during 90-min access tests. FLZ had no effect on intake or several measures of licking microstructure including first minute lick rate, mean lick-burst size, mean lick-burst duration, number of bursts in the meal, or meal duration (n=13; most F values <1) . An ongoing follow-up study indicates that midazolam injections to the PBN (2 ug or 4 ug/0.4 ul saline per side) significantly increase 0.2M sucrose consumption by approximately 50% after a 4 ug dose ($p < 0.012$), which is consistent with prior studies (Soderpalm & Berridge, 2000). The results suggest that acute blockade of benzodiazepine receptors in the PBN is insufficient to suppress behavioral responses to a moderately palatable taste stimulus. We are currently evaluating whether PBN FLZ injections block hyperphagia after GABAergic tone is increased by exogenous benzodiazepine injections. This potential outcome would suggest that the PBN is a necessary relay for benzodiazepine-mediated hyperphagia. Supported by Amherst College and HHMI. Acknowledgements: Amherst College and Howard Hughes Medical Institute Undergraduate Research Fellowship.

#P188 POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION

Effects of Astringency Sensations on Oral Fat Perception

Catherine Peyrot des Gachons[1], Paul A.S. Breslin[1,2]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]Department of Nutritional Sciences, Rutgers University School of Environmental and Biological Sciences New Brunswick, NJ, USA

Fatty foods increase the perceived basal lubricity of the mouth, naturally provided by salivary proteins such as mucins, whereas astringent drinks and foods decrease it. We hypothesized that fatty sensations and astringency are at opposite ends of an oral tribological spectrum. Therefore, both sensations should not be strongly felt simultaneously and they should tend to counter one another. Several studies have shown that viscous solutions with lubricating properties such as sucrose or oil decrease astringency sensation but very little work has been done on the effects of astringency sensations on oral fat perception. Here, through a series of psychophysical experiments, we show that astringent rinses (grape seed extract) can diminish the sensation of oral fattiness and slipperiness elicited by a lubricant, significantly more efficiently than non-astringent rinses (Urea). We also show that fat perception can be modulated both by a single strongly astringent sip and by multiple mildly astringent sips, the last procedure mimicking more closely what occurs during the course of a meal. Such phenomena might have real world meaning and explain why so many societies have developed the gastronomic customs of drinking astringent beverages during meals, such as tea and wine. The perceived high-lubricity conferred by fatty foods would be countered by oral exposure to astringent stimuli, most likely providing a sensation of "cleanness" in the mouth of individuals. Acknowledgements: Suntory and NIH DC02995

#P189 POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION

Effects of systemic injection of the glucagon-like-peptide-1 (GLP-1) receptor agonist, Exendin-4, and antagonist, Exendin-3(9-39), on intake and concentration-dependent licking of sucrose by rats

Clare M Mathes, Alan C Spector
Florida State University Department of Psychology and Program in Neuroscience Tallahassee, FL, USA

GLP-1 is expressed in taste bud cells and its receptors are found on intragemmal fibers. GLP-1-receptor knockout mice lick less to low sucrose concentrations than wild-type mice. Here we assessed the effects of exogenous GLP-1 modulation on intake and taste responses to water and sucrose in various 30-min tests. Rats were first tested with water presented continuously and then in 10-s trials while 23-h water-deprived. Then they were tested with a sucrose series (0, 0.1-1 M) presented in randomized blocks of 10-s trials while 23-h fasted or nondeprived in a lickometer. Rats (n=8/group) were injected (ip) 15 min before testing with either Exendin-4 (Ex4, 1 g/kg) or Exendin 3(9-39) (Ex9, 30 g/kg), and also with vehicle (phosphate buffered saline, 1 ml/kg). Finally, we assessed the effect of the peptides and vehicle on 0.3 M sucrose intake every 30 min in a 1-h 1-bottle test while the rats were fasted. Ex4: a) decreased total licks to water when presented continuously, b) decreased trials taken to water, c) decreased trials taken to sucrose while the rats were nondeprived, and d) decreased 1-bottle 0.3 M sucrose intake, but, importantly, e) had no effect on lick rate for sucrose during brief access trials regardless of deprivation state. Ex9 had no effect on water lick rate during 10-s trials or 1-bottle 0.3 M sucrose intake, but did significantly decrease water trials taken and lick rate to sucrose during 10-s trials while nondeprived, but the magnitude of these effects were minor. Ex9 did partially block the effect of Ex4 on 1-bottle sucrose intake, demonstrating its potency. Collectively, the findings suggest that, unlike genetic deletion of GLP-1 receptors, exogenous modulation of GLP-1 signaling does not robustly alter concentration-dependent responding to sucrose in a brief access test. Acknowledgements: This work was supported in part by NIDCD NRSA 1F32DC010517 to CMM.

#P190 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Descending modulation of taste/ethanol-responsive neurons in the nucleus of the solitary tract from the nucleus accumbens

Cheng-Shu Li
Anatomy Carbondale, IL, USA

The nucleus of the solitary tract (NST) and the parabrachial nuclei (PbN are the first and second taste relays in the rodent taste pathway, respectively. From the PbN, the ventral forebrain pathway and the thalamocortical pathway carry taste information further to the forebrain taste areas and cortex. The ventral forebrain pathway involves various forebrain gustatory nuclei including the nucleus accumbens (NAcc). It is known that NAcc plays an important role in the rewarding and subsequent addictive properties of drugs of abuse and alcohol in particular. Here, we studied the effect of electrical stimulation of the NAcc on the responses of taste/alcohol-responsive neurons in the NST. Single-unit activity was recorded from the hamster NST. After confirming the responsiveness of a NST neuron to taste/ethanol stimulation, the NAcc was stimulated bilaterally. We recorded a total of 61 taste/ethanol-responsive cells in the NST; 19 being NaCl-best, 11 being sucrose-best, 14-being citric acid-best, and 17 being quinine hydrochloride (QHCl)-best neurons. Except for one cell, all sucrose-best neurons also responded to ethanol stimulation whereas ethanol stimulation did not elicit responses of other best-stimulus cell categories. Thirty-seven cells (61%) including all sucrose-best/ethanol-responding cells were orthodromically activated following the NAcc stimulation; 34 cells after the ipsilateral NAcc stimulation and 4 cells following the contralateral NAcc stimulation. All NAcc-elicited responses of the NST cells were exclusively excitatory. These results suggest that sucrose-best cells in the NST preferentially co-respond to alcohol, and that the NAcc exerts intensive centrifugal influence to taste-responsive cells in the NST, and sucrose/ethanol-responding cells in particular. Acknowledgements: NIDCD006623 to C.-S. Li

#P191 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Activation of mu opiate receptors presynaptically suppresses afferent input to rNST neurons

Alison J Boxwell[1], Yuchio Yanagawa[2], Susan P Travers[1], Joseph B Travers[1]
[1]Ohio State University, Neuroscience Graduate Studies Program and College of Dentistry Columbus, OH, USA, [2]Department of Genetic and Behavioral Neuroscience, Gunma University, Graduate School of Medicine Maebashi, Japan

The rostral nucleus of the solitary tract (rNST) contains a rich substrate for modulation of taste processing including opioid peptides and receptors. Met-enkephalin suppresses gustatory responses (Li et al., '03) and in vitro data demonstrate that solitary tract (ST)-evoked responses in parabrachial projection neurons are attenuated by met-enkephalin via post-synaptic delta receptors (Zhu et al., '09). Behavioral pharmacological studies from our lab also suggest an influence of mu opiate receptors in rNST (Kinzeler and Travers, '08), but the underlying mechanism is unknown. To determine the site of action, we tested effects of the mu opiate agonist DAMGO in an in vitro preparation. Whole cell patch clamp recordings were made from 300um coronal brain slices from knock-in mice (P14-19) expressing EGFP under the control of the promoter for GAD67, a synthetic enzyme for GABA (Tamamaki et al., '03). Paired pulse ST stimulation was delivered through a bipolar electrode (10-120 uA). DAMGO (0.03 uM) significantly suppressed excitatory currents in 2/12 EGFP-positive (17%) and 4/6 EGFP-negative neurons (67%). DAMGO did not significantly alter membrane resistance in either cell type but did increase the paired pulse ratio by 53-195% in suppressed cells, indicative of a presynaptic effect. The magnitude of suppression was profound and similar in the 2 populations (49+/-8%; 49+/-25%), but the proportion of significantly suppressed cells was greater for EGFP-negative neurons ($p=0.046$). The membrane capacitance of EGFP-negative neurons was higher ($p=0.015$), implying a larger cell size. These findings suggest that DAMGO attenuates rNST activity via a presynaptic mechanism that preferentially affects non-GABAergic cells, perhaps output neurons contributing to ascending or local circuit pathways. Acknowledgements: NIH DC00416 & DC00417

#P192 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Insulin reduces sweet responses in the chorda tympani nerve

Arian F Baquero, Sue C Kinnamon
Department of Otolaryngology and Rocky Mountain Taste and Smell Center, University of Colorado Denver Aurora, CO, USA

Recently, Baquero and Gilbertson (AJP Cell Physiol 2010 Nov 24) reported that insulin activates the insulin receptor and increases ENaC function in taste receptors cells. We have used Chorda Tympani (CT) nerve recordings to investigate whether insulin modulates other taste qualities. Since insulin is released from the pancreas in response to carbohydrate ingestion, we asked whether insulin might affect sweet taste in addition to salt taste. Insulin (500 nM) was bath-applied to the tongue for 5 min prior to application of insulin plus tastant. In all cases (n=8), CT nerve responses to both 500 mM and 1M sucrose were significantly and reversibly decreased in the presence of insulin. As a control, we also tested the effects of insulin on NaCl responses. As expected from results on isolated taste cells, responses to 100 mM NaCl were significantly increased in the presence of insulin. In contrast, responses to 30 mM quinine, an aversive stimulus, were unaffected by the presence of insulin. Our results suggest that the effects of insulin are quality-specific and are mediated by the presence of insulin receptors on sweet-sensitive taste cells. These data suggest that insulin may act in concert with other satiety hormones such as leptin to inhibit the intake of sweet compounds. Acknowledgements: Supported by R01DC006021 and P30DC004657.

#P193 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Modulation of taste sensitivity by PYY signaling

Michael S. La Sala[1], Daniela Hurtado[1], Sergei Zolotukhin[1], C. Shawn Dotson[2,3]
[1]University of Florida, College of Medicine, Department of Pediatrics Gainesville, FL, USA, [2]University of Florida, College of Medicine, Departments of Neuroscience & Psychiatry Gainesville, FL, USA, [3]University of Florida, [3]Center for Smell and Taste Gainesville, FL, USA

Recent findings suggest that the gustatory system shares a number of common features with the gastrointestinal system. Indeed, many metabolic polypeptides have been shown to be expressed in taste cells or to be present in saliva. In addition, the cognate receptors for these peptide hormones are expressed in taste cells, found on afferent nerves fibers, or in the oral mucosa. The proximity of these agents and their cognate receptors suggested that these hormones likely have roles in taste functioning. Peptide YY (PYY) is a satiation hormone released postprandially into the bloodstream from enteroendocrine L-cells in the gut epithelia. We recently reported that PYY_{3-36} is also present in the oral cavity, in particular in murine as well as in human saliva. In addition, we found that 1) PYY is synthesized in the taste receptor cells in taste buds of the tongue, and 2) PYY_{3-36}-preferred Y2 receptor is abundantly expressed in basal layer of the tongue epithelia as well as in the salivary ducts of the Von Ebner's gland. The objective of the current report was to investigate whether PYY_{3-36}, similar to other neuropeptides, modulates taste perception. In the preliminary experiments in PYY KO mice model we showed that PYY modulated taste sensitivity and lipid sensing. Experiments are under way to extend these findings in a wild-type mouse model and to map PYY-positive taste cells in the circumvallate papilla using GFP as a surrogate marker for PYY. In summary, these data suggest a role for PYY signaling in the oral cavity in the modulation of taste sensitivity.
Acknowledgements: NIDCD (P30 DC010763) and the Children's Miracle Network Foundation.

#P194 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Leptin modulation of sweet taste behavior is context-dependent

Amanda E.T. Elson[1], Christa M. Patterson[2], Martin G. Myers, Jr.[2], Steven D. Munger[1,3]
[1]Department of Anatomy and Neurobiology, University of Maryland School of Medicine Baltimore, MD, USA, [2]Division of Metabolism, Endocrinology, and Diabetes, Department of Internal Medicine, University of Michigan Ann Arbor, MI, USA, [3]3Department of Medicine, University of Maryland School of Medicine Baltimore, MD, USA

Many hormones that facilitate glucose and energy homeostasis also regulate taste sensitivity in taste buds. Previously, we reported that glucagon, a key regulator of glucose homeostasis, is produced in taste cells and modulates sweet taste responsiveness, likely through local actions on its cognate receptor. Disruption of glucagon signaling reduces behavioral taste responses to sucrose, suggesting that this hormone normally acts to enhance or maintain sweet taste responsiveness. In contrast, the adipocyte hormone leptin has been reported to reduce sweet taste responses in behavioral assays (Shigemura, 2004). To understand how these hormones may work together to modulate taste function, we assayed taste responses in mice to two concentration ranges of sucrose using a brief access test. Signaling by glucagon, leptin, or both hormones was altered by pharmacological (addition of the GlucR antagonist L-168,049 (1 M) to the taste solution; i.p. injection of leptin (100 ng/g)) or genetic (*Scg5*$^{-/-}$ mice, which lack glucagon) manipulations. Consistent with published studies, a suppressive action of leptin was observed when testing a sucrose range of 30 mM - 1 M (high range; $P<0.05$). However, leptin had a small ($P=0.05$) enhancing effect when mice were tested at a lower sucrose range (25 mM - 400 mM). Consistent with our published results, disruption of glucagon signaling by either L-168,049 treatment or *Scg5* deletion reduced sweet taste responsiveness. Combining leptin and glucagon manipulations did not result in additional taste suppression in either range. However, leptin treatment "rescued" the suppressive effects of disrupted glucagon signaling in the low sucrose range. Together, these data suggest that glucagon and leptin modulate taste responsiveness in a context-dependent manner.
Acknowledgements: NIDCD (DC010110, DC010113), Ajinomoto Amino Acid Research Program

#P195 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Tastants and low-caloric sweeteners induce differential effects on release of satiety hormones

Maartje CP Geraedts[1], Freddy J Troost[2]
[1]Maastricht University, Department of Human Biology Maastricht, Netherlands, [2]Maastricht University, Department of Internal Medicine, div of Gastroenterology and Hepatology Maastricht, Netherlands

Recent data have demonstrated that ingestion of intact pea protein results in elevated levels of satiety hormones. As dietary components, proteins are usually not ingested as single ingredients, but are combined with other products such as tastants. However, the effect of tastants, and in particular sweet tastants, and the combination with macronutrients on satiety hormone release is still unknown. In this study, we incubated STC-1 cells, a murine enteroendocrine cell line, with different concentrations of bitter, sour, sweet, salty, and umami tastants, with several commercial sweeteners (Splenda, Natrena, Candarell, Stevia, and Tagatesse), or with a combination of pea protein and sweetener. CCK and GLP-1 concentrations were measured using RIA. All tastants increased CCK levels in both a dose- and time-dependent manner. Tastant-induced GLP-1 release was similarly dose-dependent, with the exception of acetic acid. GLP-1 release was also time-dependent with all tastants; however, bitter tastants only stimulated GLP-1 release during the first 15 minutes of exposure. All commercial sweeteners elevated both CCK and GLP-1 levels, with Tagatesse exerting the strongest effects. While all sweeteners and pea protein elevated GLP-1, only aspartame, sucralose, sucrose, pea protein, or pea protein with sucralose elicited elevated levels of CCK. Simultaneous addition of pea

protein and sucralose induced higher levels of CCK and GLP-1 that either compound alone. Together, our results indicate that different tastants, but especially sweet-tasting ones, can promote the release of satiety hormones from STC-1 cells and suggest that combinations of dietary compounds may provide a novel tool to control appetite and food intake. Acknowledgements: This research was supported by the Transnational University of Limburg.

#P196 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Melanocortin receptor mediation of glucagon-like peptide-1 conditioned taste aversion processing

Annie Handler, Christina Wright, Laura Turner, Lindsay Grigg, Julia Lord, John-Paul Baird
Amherst College, Dept Psychology & Program in Neuroscience Amherst, MA, USA

Glucagon-like peptide-1 (GLP1) is implicated in satiety, visceral illness and conditioned taste aversion (CTA). Paired with a novel tastant, GLP1 agonist injections into the lateral ventricle (LV) produce a CTA (Kinzig et al., 2002; Baird et al., AChemS 2009). Melanocortin receptor (MCR) agonists also reduce food intake and produce a CTA at high doses. We therefore evaluated whether LV injection of the MC4R antagonist SHU9119 influenced a CTA caused by LV EX4. Rats with LV cannulas were water deprived for 4 days except when daily offered two bottles of the same fluid in succession (8 min per bottle). On the first 2 days rats were offered dH2O. On day 3, rats were LV injected with SHU9119 (0.1nM/2ul) followed 30 min later by LV injection of EX4 (1µg/2µl). After 15 min, rats were offered 0.12M NaCl from two bottles. On day 4, rats were offered 0.12M NaCl as on the prior day, with no injection. Relative to dH2O, SHU9119/EX4 markedly reduced licking for NaCl ($t(10)=4.34$, $p<0.001$). Compared with prior data (Baird et al., AChemS 2009), the reduction was not different from that observed after EX4 alone ($t(17)=1.86$, NS). Thus, MC4R blockade did not affect unconditioned malaise or short-term CTA produced by EX4. However, day 4 NaCl intake after SHU9119/EX4 was significantly greater than for rats receiving EX4 alone ($t(17)=3.14$, $p=0.006$). Therefore, MC4R blockade appeared to disrupt either consolidation or recall of the EX4 CTA. We chose a SHU9119 dose (0.1nM) that is subthreshold for hyperphagia, but the increased day 4 NaCl licking in the SHU9119/EX4 group could be due to a long-lasting effect of SHU9119. Thus, we are testing CTA responses 4 days after SHU9119-EX4 injection. This caveat notwithstanding, the preliminary data suggest an interaction between MC4R and GLP1 systems in CTA processing. Acknowledgements: Amherst College, HHMI, and DC07389

#P197 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Reducing Sodium Levels in Frankfurters by Using Soy Sauce and Natural Flavor Enhancer

Megan M. McGough[1], Jeffrey J. Sindelar[1], Takuya Sato[2], Scott A. Rankin[1], Larry L. Borchert[1]
[1]University of Wisconsin-Madison Madison, WI, USA,
[2]Kikkoman USA R&D Laboratory, INC. Madison, WI, USA

Sodium chloride serves several important functions in processed meats, making significant contributions to quality and food safety characteristics; however, renewed interest exists in reducing sodium content in human diets. Because of sodium chloride's multiple functions in processed meats, it is not realistic to remove it from formulations. Current sodium chloride usage levels are near minimum sensory thresholds, therefore, new methods must be investigated to reduce sodium in processed meats. The study investigated sensory effects of partial replacement of NaCl with ingredients reported to enhance the salty taste sensation, naturally brewed soy sauce (SS) and commercial soy fermentation product (NFE). Varying levels of SS or NFE were used with NaCl and/or KCl to constitute treatments (TRT). All studies included a 100% NaCl control TRT (C; 2.5% of meat weight). Phase I included 25%, 50%, 75%, and 100% NaCl replacement by either SS or NFE (SS/NFE). Phase II used a 50% SS/NFE-50% NaCl base formulation (TRT1) as well as 10% (TRT2), 20% (TRT3), 30% (TRT4) NaCl reductions. Phase III incorporated SS/NFE, NaCl, and KCl to investigate whether the salty taste enhancing effects of SS/NFE attenuate the bitterness associated with KCl TRTs. Effects were declared significant at the $a<0.05$ level. Phase I consumer sensory responses indicated that TRTs with SS/NFE replacement were perceived as saltier than C with the panelists tending to prefer SS/NFE TRTs over C. Phase II sensory responses showed no significant differences for overall liking between SS TRTs; however, only TRTs 2 and 3 were liked more than C. NFE TRTs 2 and 3 scored equally highest for overall liking and were both higher than C. SS/NFE TRT 1 was saltier than SS/NFE TRTs 2, 3, 4, and C, showing that SS/NFE enhanced the perceived saltiness. Acknowledgements: Kikkoman USA R&D Laboratory, INC.

#P198 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

The impact of retronasal odor and taste on hedonic responses to vegetables

Arthi Padmanabhan, Juyun Lim
Oregon State University/Department of Food Science and Technology Corvallis, OR, USA

While previous research has suggested that bitterness is the key determinant of vegetable disliking, it is unknown what role retronasal odor plays in vegetable hedonics. We therefore investigated the impact of retronasal odors as well as taste on liking and disliking of four vegetables (steamed asparagus and Brussels sprouts; fresh celery and snap peas). To minimize the visual and textural cues, the vegetables were finely chopped into small pieces. Subjects tasted a small portion of the samples with

the nose open or closed, and rated the degree of liking/disliking as well as perceived intensities of sweetness, bitterness, saltiness, and vegetable flavor on the LHS and gLMS, respectively. Subjects were classified as 'likers' or 'dislikers' of each vegetable based on the valence of their hedonic ratings under the nose open condition (i.e., when retronasal odors could be perceived). The degree to which 'likers' liked and 'dislikers' disliked the vegetables was significantly *less* ($p<0.05$) in the nose closed condition, indicating that retronasal odor was a significant driver of vegetable hedonics. Importantly, perceived vegetable odor intensity did not differ significantly between 'likers' and 'dislikers' for any of the vegetables, which implies that the quality of specific vegetable odors, but not their intensity, drove hedonic ratings. In contrast, taste intensity ratings measured in the nose closed condition sometimes differed significantly ($p<0.05$) between 'likers' and 'dislikers', but not in a consistent manner across vegetables. These results suggest that retronasal odor is a more consistent driver of vegetable liking and disliking than is taste. A follow-up experiment is underway to further investigate the role that specific retronasal odors and tastes play on liking and disliking of vegetables. Acknowledgements: This work was supported by Oregon State University start-up funds.

#P199 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Color facilitation in speeded gustatory detection

Adam Y. Shavit[1,2], Timothy G. Shepard[1], Maria G. Veldhuizen[1,4], Lawrence E. Marks[1,2,3]
[1]John B. Pierce Laboratory New Haven, CT, USA, [2]Division of Environmental Health Sciences, Yale School of Public Health New Haven, CT, USA, [3]Department of Psychology, Yale University New Haven, CT, USA, [4]Department of Psychiatry, Yale University School of Medicine New Haven, CT, USA

Physical appearance is central to the identification of food and beverages, to the perception of flavor, and, ultimately, to ingestion. The present experiment tests whether the presence of a color can improve the detection of the tastant sucrose, and whether the improvement in detection reflects a change in the gustatory sensation (a lower-level, sensory enhancement), or a change in the tendency to report the presence of a gustatory sensation (a higher-level, cognitive change in decision criterion). The subjects' task is to discriminate sucrose from water when presented in clear or in red solutions. Sucrose is presented at concentrations sufficiently above threshold to be clearly perceptible, but not aversive. The stimuli are presented in both a blocked design (alternating blocks of trials containing red solutions and clear solutions) and an intermixed design (trials containing red solutions and clear solutions interspersed randomly within a given block). Thus far, we do not find significant and consistent effects of color on sensitivity or on the speed-accuracy trade-off; however, we do see consistent effects of color on decision criterion. For example, increases in hit rate are accompanied by increases in false alarm rate. The decision criterion is higher in the intermixed than the blocked design for red solutions, but lower in the intermixed design for clear solutions. These preliminary data suggest that the sensory effect of color, if any, is likely to be much smaller than the decisional effect. Thus, these results are consistent with the hypothesis that color primarily affects flavor detection through higher-level, decision processes. Acknowledgements: NIH grant DC009021

#P200 **POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION**

Identification of Gustatory-Olfactory Flavors: Effects of Stimulus Context

Timothy G Shepard[1], Adam Y Shavit[1,2], Maria G Veldhuizen[1,4], Lawrence E Marks[1,2,3]
[1]John B. Pierce Laboratory New Haven, CT, USA, [2]Yale School of Public Health/Division of Environmental Health Sciences New Haven, CT, USA, [3]Yale University/Department of Psychology New Haven, CT, USA, [4]Yale University School of Medicine/Department of Psychiatry New Haven, CT, USA

We investigated how stimulus context affects the identification of gustatory (sucrose) - olfactory (citral) flavor mixtures. In a baseline session, we determined, for each subject, concentrations of sucrose and citral that matched in flavor intensity, deriving matches from ratings of perceived intensity of several concentrations of both flavorants. The matching stimuli were then used to create seven flavor mixtures that proportionately contained either more Citral than Sugar or more Sugar than Citral (0.10S/0.90C, 0.25S/0.75C, 0.40S/0.60C, 0.50S/0.50C, 0.60S/0.40C, 0.75S/0.25C, and, 0.90S/0.10C). The main experiment then compared responses in two conditions, tested in separate sessions: In the uniform condition, each of the seven stimuli was randomly presented 12 times throughout the session. In the skewed condition, the frequency of presentations was skewed so that half of the subjects received more trials with mostly-sucrose mixtures, half more trials with mostly-citral mixtures. On each trial, subjects sipped 5ml of the mixture, expectorated, and then responded whether the mixture contained more 'sugar' or more 'citral'. Identification of the 0.50S/0.50C stimulus fell around 50% in the uniform condition (called 'sugar' and 'citral' equally often). Identification in the skewed condition showed contrast: Subjects were more likely to call 0.50S/0.50C 'citral' in the 'mostly-sucrose' condition, more likely to call it 'sucrose' in the 'mostly-citral' condition. Indeed, both psychometric functions in the skewed conditions shifted away from the function of the uniform condition. Thus, stimulus context affects the perceived identity of gustatory-olfactory flavors, perhaps by modifying the relative perceived intensities of the components. Acknowledgements: Supported by NIH grant DC009021

#P201 POSTER SESSION IV: OLFACTION: RECEPTORS AND PERIPHERY; OLFACTORY CNS; TASTE MODULATION; MULTIMODAL PERCEPTION

Sex-dependent responses to irritating chemosensory stimuli, but not pure odors, are modulated by state anxiety

Andrea Ponting[1], Sanne Boesveldt[1,2], Eva C. Alden[1], Johan N. Lundstrom[1,3,4]
[1]Monell Chemical Senses Center, Cognitive Neuroimaging Laboratory Philadelphia, PA, USA, [2]Wageningen University, Division of Human Nutrition Wageningen, Netherlands, [3]University of Pennsylvania, Department of Psychology Philadelphia, PA, USA, [4]Karolinska Institute, Department of Clinical Neuroscience Stockholm, Sweden

It has been shown that men and women differ in their cerebral responses to irritating chemosensory stimuli, but show little to no difference for "pure odors". We hypothesized that these sex-dependent differences for irritating chemosensory stimuli may be linked to a heightened autonomic nervous system (ANS) response (arousal) resulting from the painful component of the odor stimulus. To test this hypothesis, we investigated the ANS response in 40 healthy subjects (20 women), by means of evoked galvanic skin response (GSR), to a bimodal odor (menthol, with both an olfactory and trigeminal percept) and a pure olfactory odor (phenyl ethyl alcohol [PEA], rose odor). Participants were asked to rate both the intensity and irritation of the stimuli, following the presentation of each odor by an olfactometer. Sensitivity to olfactory (PEA) and trigeminal (menthol) stimuli, as well as state and trait anxiety (State-Trait Anxiety Inventory [STAI]), were assessed. No significant main differences were found between men and women for any of the individual measures, including sensitivity to each of the odors. However, there was a significant correlation between evoked GSR to menthol and state anxiety scores (STAI-S) for women (r = .458, p = .032), but not men (r = .153, p = .521). No correlation was observed between evoked GSR response to PEA and STAI-S scores for either men or women, or between evoked GSR and STAI-T scores for any condition. Results from this study indicate that greater state anxiety levels are linked to a greater arousal response to irritating stimuli in women, but not men. This suggests that sex-dependent differences to bimodal odors are, to some extent, mediated by a subset of women expressing a high level of anxiety towards the irritating portion of the odor percept.
Acknowledgements: Supported by start-up funds from the Monell Chemical Senses Center awarded to JNL.

#P202 POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING

TrpM5-expressing superficial epithelial cells in the main olfactory epithelium of mice

Anne Hansen, Marco Tizzano
University of Colorado and Rocky Mountain Taste and Smell Center Denver, CO, USA

The nasal cavity hosts an array of chemoresponsive cells including not only the elements of the extended olfactory system but also an abundance of other cells involved in detection and responses to irritants and other gaseous substances. The transient receptor potential channel TrpM5 is present in the olfactory and the taste system and in scattered cells in the respiratory epithelium (RE). The channel is an essential element in the taste transduction cascade and many of the TrpM5-positive cells in the RE share other elements of taste transduction. We and other investigators have described a diversity of TrpM5-positive cells in the RE, which make contact with nerve fibers leading to their designation as solitary chemosensory cells (SCCs). Previously, we described a population of small, apically-situated microvillous (MV) cells in the MOE (Hansen and Finger, 2008). Given the scarce information known about these MV cells, their function is still enigmatic. We re-examined these superficial TrpM5-positive MV cells with more sensitive techniques to clarify whether they express only the TrpM5 protein, or also contain other elements of the taste or SCC transduction cascade. The TrpM5 cells of the MOE, like TrpM5-positive taste cells and SCCs, exhibit PLC 2, gustducin, and in many cases a T1R3-driven GFP transgene. These findings suggest the entire taste transduction cascade is present. To test whether these cells are sensory, we used antisera against CGRP, substance P, and ChAT to test whether they are innervated. The vast majority of the superficial TrpM5-positive cells lacked innervation, but many were themselves ChAT-positive, suggesting a cholinergic phenotype. We suggest that these cells are chemoresponsive but not sensory, and may serve to modulate activity of the surrounding olfactory epithelium.
Acknowledgements: Supported by NIH NIDCD grants RO3 DC-007732 (A.H.), P30 DC-04657 and RO1 DC-06070 (D. Restrepo and T. Finger).

#P203 POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING

Potential cholinergic influences on olfactory sensory neurons and supporting cells by TRPM5/ChAT-expressing microvillous cells in mouse main olfactory epithelium

Tatsuya Ogura, Steven Szebenyi, Aaron Sathyanesan, Kurt Krosnowski, Weihong Lin
Dept. Biological Sciences, University of Maryland Baltimore County Baltimore, MD, USA

The mammalian main olfactory epithelium (MOE) constitutes multiple types of cells, among which are microvillous cells with unknown function. Previously, we reported that a large population of microvillous cells expresses transient receptor potential channel M5 (TRPM5; Lin et al. 2008). These cells are evenly distributed throughout the MOE with their cell bodies

located in the superficial layer. Because the TRPM5-expressing microvillous cells are neither neuronal nor innervated by trigeminal nerve fibers, we examined whether they could communicate with and influence neighboring supporting cells and olfactory sensory neurons (OSNs) via neuroactive substances, such as acetylcholine (ACh), which is known to be present in the airway mucosa. We found that the TRPM5-expressing microvillous cells immunoreacted to an antibody against choline acetyltransferase (ChAT), a key enzyme for ACh synthesis. In MOE of transgenic ChAT^{BAC}-eGFP mice, strong GFP expression was found in microvillous cells identical to those expressing TRPM5. These results strongly suggest that TRPM5-expressing microvillous cells are capable of synthesizing ACh. We thus determined whether ACh influences activities of supporting cells and OSNs. In Ca^{2+} imaging experiments, ACh induced increases in intracellular Ca^{2+} levels in supporting cells, which was blocked by the muscarinic ACh receptor antagonist atropine. ACh also altered the levels of Ca^{2+} increases induced by adenylyl cyclase activator forskolin in a subset of OSNs. Further, in immunolabeling using subtype-specific antibodies, we found that supporting cells and OSNs express different muscarinic receptor subtypes. Our data suggest that ACh can be released from the microvillous cells and modulates activities of neighboring cells by paracrine mechanisms. Acknowledgements: Supported by NIH/NIDCD DC009269 and ARRA administrative supplement to WL.

#P204 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

NPY release in mouse olfactory epithelium is partially dependent on the presence of functional IP3R3 receptor subtype

Sebastien Hayoz, Marcus Weera, Cuihong Jia, Colleen C Hegg
Michigan State University / Department of Pharmacology and Toxicology East Lansing, MI, USA

We previously showed that ATP induces the upregulation of NPY and increases neuroproliferation mediated by the NPY Y1 receptor subtype in adult mouse olfactory epithelium (OE). Also, ATP increases NPY release in mouse neonatal OE slices. Thus, ATP likely induces neuroproliferation by increasing both the production and the release of NPY. Interestingly, ATP upregulates NPY production specifically in the microvillous cell subtype that expresses the IP3R3 receptor subtype. IP3 receptors release calcium from the endoplasmic reticulum to regulate a wide variety of physiological processes including secretion. Thus, ATP-induced NPY release and neuroproliferation could depend at least partially on the IP3R3 receptor. To test this hypothesis, we cultured neonatal mouse OE slices for 24 hours in the presence or absence of the IP3R receptor inhibitor 2-APB (100 µM) and in the presence or absence of ATP (100 µM) and measured released NPY in the culture media by ELISA. ATP significantly increased the release of NPY compared to control slices (41.81 ± 1.84 pg/slice vs. 29.2 ± 1.14 pg/slice; $p<0.001$). Treatment with 2-APB alone did not significantly change the amount of released NPY compared to control (29.33 ± 2.1 pg/slice). Slices treated with both 2-APB and ATP released significantly less NPY compared to ATP-treated slices (35.18 ± 2.54 pg/slice; $p<0.05$). To determine the role of the IP3 receptor in proliferation, we quantified the amount of cell proliferation marker PCNA in adult mice that were intranasally administered ATP or ATP + 2-APB via Western blot. We conclude that ATP-induced NPY release in neonatal OE at least partially depends on the IP3R3 receptors. Acknowledgements: NIDCD 006897 and Swiss Fellowship For Advanced Researchers PA 00P3_131493

#P205 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Effects of cocaine on the olfactory receptor cell function

Kengo Tamari[1,3], Hiroko Takeuchi[2], Masayoshi Kobayashi[3], Takashi Kurahashi[2], Tetsuro Yamamoto[1]
[1]Department of Neurophysiology, Mie University Graduate School of Medicine Tsu, Japan, [2]Department of Physiology, Osaka University Graduate School of Frontier Biosciences Toyonaka, Japan, [3]Department of Otorhinolaryngology-Head and Neck Surgery, Mie University Graduate School of Medicine Tsu, Japan

Cocaine (actually 1-5% concentration) is often used as topical anesthetics in endoscopic sinus surgery. However, it remains to be proved if the cocaine is innocuous for olfactory function. the present study was designed to reveal if the cocaine affected olfactory system by using newt olfactory receptor cells (ORCs). Newt ORCs were isolated from the olfactory mucosa, placed on a dish and observed under an inverted microscope. Under the whole cell voltage clamp mode, membrane potential of the single ORC was depolarized from a holding potential of -100 mV in a stepwise between -90 and +40 mV and its voltage-gated currents were recorded. The puff application of 5% cocaine solution significantly reduced both inward and outward voltage-gated currents, which recovered completely after washout. The dose-suppression curves of cocaine for voltage-gated sodium and potassium currents fit the Hill's equation. Half-blocking concentration and Hill's coefficient of the sodium current were 43 µM and 1.1. Those of the potassium current were 54 µM and 0.9. We also investigated the effect of cocaine on odorant responses with cineol. the peak amplitude of cineol-induced responses was reduced after cocaine application and recovered completely after washout. These results indicate that cocaine can suppress olfactory function at the level of the ORCs but never impair them permanently.

#P206 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Neuropeptidergic Presynaptic Modulation Mediates Starvation Dependent Odor-Driven Food Search Behavior in *Drosophila*

Kang I. Ko, Cory M. Root, Jing W. Wang
University of California, San Diego/Division of Biological Sciences La Jolla, CA, USA

Early sensory processing plays a critical role in detecting environmental cues. We have investigated the effect of starvation on food finding behavior and odor-evoked activity in the first olfactory synapse in *Drosophila*. Using two-photon calcium imaging, we demonstrate that this behavioral change is accompanied by dramatic shifts in the odor map. The neuropeptide sNPF, a homolog of the mammalian NPY, is highly

implicated in hunger signaling and is expressed in *Drosophila* olfactory sensory neurons (OSNs). Using RNAi to knockdown sNPF, we found that sNPF signaling in DM1 glomerulus is necessary for the starvation-dependent modulation of both olfactory representation and food finding behavior. Furthermore, the sNPF receptor is expressed in OSNs and mediates a feedback enhancement of sensory transmission in DM1. Thus, starvation causes presynaptic facilitation of sensory transmission, which leads to increased food finding behavior. We then investigated starvation modulation in DM5—a glomerulus that mediates innate aversion behavior. Starvation suppresses DM5's sensitivity to odor stimulation, and that this suppression is mediated by tachykinin signaling. Furthermore, starvation modulation of DM5 also influences food finding behavior. Thus, early olfactory processing and appetitive behavior are profoundly controlled by metabolic states in *Drosophila*. Starvation does not simply scale up or down global activity in the antennal lobe. Rather, it upregulates activity in certain sensory channels and downregulate it in others in what appears to be an optimization strategy to fine tune local circuits towards a concerted modulation of appetitive behaviors. Acknowledgements: Funded by NIDCD (RO1DC009597, R21DC010468, 1F31DC009511)

#P207 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Cholecystokinin: a peptide modulator of olfactory bulb output

Jie Ma, Luba Dankulich, Graeme Lowe
Monell Chemical Senses Center Philadelphia, PA, USA

The olfactory bulb contains a rich assemblage of neuromodulators woven into complex networks of overlapping pathways, many of which are still poorly understood. Deciphering these signals will require elucidation of cellular targets and physiological actions. We focused on cholecystokinin (CCK), a peptide synthesized by superficial tufted cells whose projections link isofunctional glomerular columns of medial and lateral hemibulbs. Applying immunohistochemistry to mouse olfactory bulbs, we detected CCK2R, the major brain receptor for the peptide, in juxtaglomerular and mitral cells, but not in granule cells. Double immunostaining revealed colocalization in mitral cell bodies of CCK2R and transcription factor Tbx21, a mitral cell marker. In slice patch-clamp recordings, perfusion of 1 µM CCK-8S transiently elevated spiking and induced a slow inward current (10–100 pA, duration 521 ± 98 s) in mitral cells with or without intact primary dendrites. A negative reversal potential (–60 to –100 mV) suggests that this might be mediated by suppression of a K^+ conductance. In some mitral cells, there was transient inhibition, and excitatory responses were reduced by glutamatergic blockers. These may correspond to presynaptic effects due to interneuron inhibition or lateral excitation. In CCK2R knockout mice, receptor immuno-labeling was absent, and mitral cells did not respond to CCK. From these data, we formulate a new model of CCK signaling in the olfactory bulb. Strong odorant stimuli may trigger tufted cells to release peptide that diffuses from the inner plexiform layer to the mitral cell body layer to facilitate mitral cell spiking. We suggest that this amplifies selective mitral cell outputs, by exciting coactive medial/ lateral bulb circuits encoding activation of shared olfactory receptors. Acknowledgements: Supported by: R01DC004208-06 and 3R01DC004208-06A2S1 (GL)

#P208 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Increases $[Ca^{2+}]_i$ Through Multiple Pathways in Neonatal Mouse OB

Mavis A Irwin, Mary T Lucero
University of Utah, Physiology Department Salt Lake City, UT, USA

Neonatal olfactory bulb (OB) is enriched with both Pituitary Adenylate Cyclase-Activating Polypeptide (PACAP) and its G-protein coupled receptor (PAC1 R). Without PACAP, neonates often die before weaning, suggesting that PACAP is required for normal development. We used confocal imaging of slices of neonatal mouse OB loaded with membrane permeant Ca^{2+} indicator dyes, to track changes in intracellular Ca^{2+} concentration ($[Ca^{2+}]_i$) in response to various agonists and antagonists. Superfusion of PACAP onto OB slices led to a robust oscillating increase in $[Ca^{2+}]_i$ in many OB neurons. Both the levels of $[Ca^{2+}]_i$ and numbers of responding cells increased with increasing PACAP concentrations from 100 fM to 100 nM. Blockage of PAC1 receptors with the specific antagonists M65 and PACAP6-38 (100 nM each) prevented PACAP-induced increases in $[Ca^{2+}]_i$ at moderate PACAP concentrations (≤40 nM). The blockers did not block high concentrations of PACAP possibly due to downstream release of other neurotransmitters. Previous studies showed that PAC1Rs can activate PLC, adenylate cyclase, and L-type calcium channels[1]. Here, the PLC antagonist (2-APB) or zero Ca^{2+} extracellular solution applied individually slightly reduced the number of cells responding to 100 nM PACAP. When the treatments were combined, the PACAP response was significantly reduced, suggesting a requirement of both Ca^{2+} influx and store release. Overall, the neonatal OB neurons respond in a dose-dependent manner to PACAP with increasing $[Ca^{2+}]_i$ through the activation of PAC1 receptors. These studies show that PACAP and PAC1 receptors are functional in neonatal mouse OB and modulate intracellular Ca^{2+}, which may have important downstream signaling effects on development, maturation and survival of OB neurons. 1 Curr. Protein Pept. Sci. 2002; 3: 423-439. Acknowledgements: NIH NIDCD ARRA Supplement to R01 DC02994-8 12/1/10-11/30/11

#P209 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Activation of βNoradrenergic Receptors Enhances Rhythmic Activity in the Main Olfactory Bulb

Qiang Nai[1], Hongwei Dong[1], Christiane Linster[2], Matthew Ennis[1]
[1]University of Tennessee, Health Science Center/Dept. of Anatomy and Neurobiology Memphis, TN, USA, [2]Cornell University/Dept. of Neurobiology and Behavior Ithaca, NY, USA

Previous studies reported that activation of βnoradrenergic receptors increased long-lasting depolarizations (LLDs) in mitral cells. Since LLDs appear to be generated by external tufted (ET) cells, in the present study we investigated the effects of norepinephrine (NE) and βreceptor agonist isoproterenol (Iso) on the activity of ET cells using patch clamp electrophysiology in rat and mouse olfactory bulb slices. Bath applied NE or Isop

increased the frequency and regularity of ET cell bursting in normal media or in the presence of APV-CNQX-gabazine. Focal puffs of Iso into the glomerular layer, but not deeper layers, increased the frequency, strength and regularity of mitral and tufted cell discharge. The increase in discharge was accompanied by an increase in the frequency and amplitude of LLDs. The increase in LLDs elicited by Iso also occurred in the presence of APV-gabazine, indicating that the enhancement was not due to changes in GABAergic inhibition in the network. Additional current and voltage clamp recordings in the presence of APV-CNQX-gabazine showed that bath or glomerular puff application of Iso elicited slow rhythmic membrane potential or current oscillations in ET and mitral cells. The oscillations persisted when voltage-gated calcium channels were blocked, but were eliminated by TTX. These results indicate that activation of βreceptors in the glomeruli directly increase ET cell excitability and rhythmic bursting. We speculate that these effects in turn generate rhythmic oscillatory activity in mitral and tufted cells via chemical and electrical synapses in the glomerular network.
Acknowledgements: DC008702, DC003195.

#P210 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Role of bulbar NE and ACh in detection of low concentration odors

Olga Escanilla[1], Samuel Alperin[1], Monica Youssef[1], Matthew Ennis[2]
[1]Cornell University, CPL & Neurobiology and Behavior Ithaca, NY, USA, [2]Univ. Tenn, Hlth Ctr, Dept. Anatomy and Neurobiology Memphis, TN, USA

The mammalian main olfactory bulb (MOB) receives a significant noradrenergic input from the locus coeruleus as well as cholinergic input from the horizontal limb of the diagonal band of Brocca (HDB). Previous studies have demonstrated a role for both bulbar NE and ACh in the discrimination of chemically similar odorants (Mandairon et al., 2006; 2008; Doucette et al 2007). We here investigate the role of noradrenergic and cholinergic modulation in the MOB for very low concentration odor detection. Previous results have shown that infusionof NE into the MOB modulates spontaneous odor detection thresholds in a dose-dependent manner in adult rats (Escanilla et al. 2010). Recently we found that increasing bulbar ACh does not have a similar effect on low odor detection. We here test if rats engage their noradrenergic or cholinergic systems when asked to detect odors at low concentrations. Rats were trained to detect a low concentration odorant versus a blank to retrieve a sweet cereal reward. On each test day, they were first trained to detect a 10-2 Pa odorant during five trials and then tested on 10-2, 10-4 and 10-5 Pa odorants in randomized order during 15 trials. Rats MOB were infused with vehicle, noradrenergic or cholinergic anagonists. Results show that rats in which bulbar NE receptors were blocked were significantly impaired at the lowest odor concentration whereas blockade of ACh receptors had no effect on odor detection. These results together with our previous findings suggest that bulbar NE but not ACh is important for the detection of very low concentration odorants. Current studies are testing the differential roles for NE and ACh in discrimination between very low concentration odorants. Acknowledgements: R01DC009948 R01DC008702

#P211 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Developmental regulation of noradrenergic inhibition in the accessory olfactory bulb

Richard S. Smith, Tyler Treadway, Ricardo C. Araneda
University of Maryland, Department of Biology College Park, MD, USA

Modulation of dendrodendritic synapses by the noradrenergic system in the accessory olfactory bulb (AOB) plays a key role in the formation of memory in olfactory mediated behaviors. We have recently shown that noradrenaline (NA) elicited a long lasting depolarization of granule cells in the AOB, mediated by α_1-adrenergic receptor (AR) activation, thereby increasing GABA inhibitory input onto mitral cells (MCs). Studies in the main olfactory bulb (MOB) have confirmed this effect and also demonstrated a dose dependent biphasic noradrenergic regulation of MCs, an effect partially dependent on activation of α_2-ARs. Here, we show that NA (10 µM) increases the frequency of inhibitory postsynaptic currents (IPSCs) in MCs in an age dependent manner. The AR-mediated increase in GABA inhibitory activity in MCs is robust at early postnatal ages and gradually declines with age. The increase in IPSCs is mediated by activation of α_1- and β-ARs, as the increase in IPSC frequency is mimicked by phenylephrine (10 µM) and completely abolished in the presence of prazosin (300 nM). Similarly, the β-AR agonist isoproterenol (10 µM) also increased the IPSC frequency in MCs. Intriguingly, application of the α_2-AR agonist clonidine (10 µM) failed to decrease this inhibitory activity, as it has been previously described in the MOB. Taken together, our results suggest that compared to the adult, at early postnatal ages NA exerts a more powerful control of inhibition in the AOB by a mechanism involving activation of α_1- and β-ARs. Acknowledgements: This work was funded by a NIDCD RO1-DC-009817 to R.C.A.

#P212 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Cholinergic Modulation of Newborn Granule Cells in the Olfactory Bulb

Alexia Nunez-Parra, Ricardo C. Araneda
University of Maryland, NACS Program & Biology Department College Park, MD, USA

Cholinergic modulation of granule cells (GCs) in the olfactory bulb (OB) is thought to play an important role in olfactory learning. Throughout life, adult neurogenesis provides the OB with a new pool of granule cells, which functionally integrate within the existing circuitry. In addition, the OB receives an extensive cholinergic innervation from the basal forebrain and it has been suggested that cholinergic neuromodulation is important for the survival and integration of GCs born in the adult. GCs originate from neural progenitors in the subventricular zone and arrive into the OB via the rostral migratory stream. Once in the OB, immature GCs migrate radially to the GC layer and complete their maturation. Fully mature GCs are found in the OB within 2 to 3 weeks after birth, and their maturation is characterized morphologically and functionally by five distinct

stages (I-V). Here, using an in vitro slice preparation we characterized cholinergic responses in GFP-labeled GCs born postnatally. We used the non-selective cholinergic agonist carbachol (30 µM) to determine the responsiveness of GCs at different stages of their maturation. We found that carbachol produced a marked increase in excitability in GCs that extended processes that branched into the external plexiform layer (IV-V). In contrast, the response to carbachol was not present in migrating and more immature neurons (I-II). These results suggest that functional modulation of GCs by the cholinergic system could be activated during the later stages of GC integration. Acknowledgements: Work supported by a NIDCD R01-DC-009817 grant to R.C.A. and a fellowship from the Chilean government to A.N.

#P213 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Activity Dependent changes in cholinergic innervation of the main olfactory bulb

Ernesto Salcedo, Tuan Tran, Xuan Ly, Robert Lopez, Cortney Barbica, Diego Restrepo, Sukumar Vijayaraghavan
University of Colorado Denver Aurora, CO, USA

The olfactory bulb receives massive cholinergic innervation, primarily from the horizontal limb in the diagonal band of Broca; however, the interplay between olfactory activity and cholinergic modulation remains to be fully understood. This report examines the pattern of cholinergic innervation throughout the mouse main olfactory bulb across different developmental stages and in naris-occluded animals. To visualize the pattern of cholinergic innervation we used a transgenic line of mice which express a fusion of the microtubule-associated protein, tau, with green fluorescence protein (GFP) under the control of the choline acetyltransferase (ChAT) promoter. This tau-GFP fusion product allows for a remarkably vivid and clear visualization of cholinergic innervation in the main olfactory bulb (MOB). In line with previous reports, we find that cholinergic innervation of the bulb increases up to approximately post-natal day 12 and then decreases by adulthood. Similarly, we consistently find a population of atypical glomeruli that receive heavy cholinergic innervation. Interestingly, we find an uneven distribution of cholinergic innervation in the adult glomerular layer, where anterior, medial, and lateral glomerular regions of the bulb receive relatively heavier cholinergic innervation that other regions. Moreover, in contrast to previous reports, we find a marked change in the pattern of cholinergic innervation to the glomerular layer following unilateral naris occlusion between the ipsilataral and contralateral bulbs in adult animals. Together, these results suggests that olfactory activity can affect the nature of cholinergic modulation of the olfactory system. Acknowledgements: NIDCD DC008855

#P214 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Cholinergic Modulation of Neuronal Circuits in the Mammalian Olfactory Bulb

Markus Rothermel[1], Ryan Carey[2], Matt Wachowiak[1]
[1]University of Utah School of Medicine/Department of Physiology Salt Lake City, UT, USA, [2]Boston University/Department of Biomedical Engineering Boston, MA, USA

The olfactory bulb receives centrifugal input from diverse brain centers, including cholinergic and GABAergic projections from the horizontal limb of the diagonal band of Broca (HDB). Here, we examine the influences of inputs originating from HDB on sensory input and mitral/tufted cell (M/T) output from the olfactory bulb using *in vivo* imaging, electrophysiological and optogenetic approaches. First, we tested whether HDB stimulation affected odorant-evoked sensory inputs using presynaptic imaging from receptor neuron terminals. HDB stimulation had no effect on either presynaptic Ca^{2+} transients or transmitter release. Next, we tested whether HDB stimulation affected postsynaptic activity by imaging from transgenic mice expressing GCaMP2 in MT and juxtaglomerular cells (Fletcher et al. J Neurophysiol 102:817). HDB stimulation caused a transient increase in the odorant- and inhalation-evoked GCaMP2 signal. Extracellular recordings from presumptive MT cells showed that HDB stimulation enhanced inhalation-evoked as well as spontaneous spiking; HDB stimulation appeared to decrease presumptive external tufted cell spiking. To specifically examine the influences of cholinergic (as opposed to GABA-ergic) inputs to the bulb, we selectively expressed channelrhodopsin (ChR2) in cholinergic HDB neurons using and injecting a viral vector carrying a cre-dependent ChR2 construct into HDB of cre-ChAT mice. Subsequent activation of these neurons with blue laser pulses led to enhanced MT cell spiking in a manner qualitatively similar to that seen with electrical stimulation. These experiments are consistent with the hypothesis that cholinergic inputs to the olfactory bulb may enhance sensory-evoked MT responses and thus may be important in the attentional modulation of early olfactory processing. Acknowledgements: Supported by DFG (MR) and NIDCD (MW, RC).

#P215 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Glomerular nicotinic receptors mediate excitation-dependent global inhibition in the olfactory bulb

Rinaldo D D'Souza[1], Sukumar Vijayaraghavan[1,2]
[1]Department of Physiology and Biophysics, University of Colorado Denver Aurora, CO, USA, [2]Neuroscience Program, University of Colorado Denver Aurora, CO, USA

Olfactory bulb glomeruli, the initial sites of synaptic processing of sensory odor information, show high levels of nicotinic acetylcholine receptor (nAChR) expression. We examined the role of nAChRs in glomerular processing using whole-cell electrophysiology. Activation of glomerular nAChRs depolarized external tufted (ET) and mitral cells, and resulted in an increase in glutamate and GABA release at dendrodendritic synapses within

the glomerulus. GABA release was, for the most part, dependent on glutamate receptor activation. In addition to excitation of ET and mitral cells, nAChR activation strongly suppressed input driven responses on mitral cells. Our results suggest that the activation of glomerular nAChRs leads to excitation-dependent "global" inhibition within the olfactory bulb that suppresses glomerular network-driven excitation of mitral cells. However, the nAChR-mediated depolarization of mitral cells leads to robust transmission of strong sensory input. We thus provide a mechanistic model for nAChR-mediated filtering and modulation of sensory input to the olfactory bulb, via a balance of excitation and inhibition, resulting in enhanced odor detection and discrimination. Acknowledgements: NIH RO1 DC 008855

#P216 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Electrical activity in the horizontal limb of the diagonal band of Broca in awake rats

Sasha Devore[1,2], Laura C. Manella[1], David M. Smith[2], Christiane Linster[1]
[1]Cornell University Department of Neurobiology and Behavior Ithaca, NY, USA, [2]Cornell University Department of Psychology Ithaca, NY, USA

Cholinergic modulation serves a critical role in regulating olfactory perception. Lesions of the cholinergic basal forebrain neurons projecting to the olfactory system, as well as direct manipulation of cholinergic signaling within the olfactory bulb (OB) and piriform cortex, have been shown to influence a variety of olfactory behaviors. While computational modeling studies from our laboratory suggest that the cholinergic inputs to the olfactory system constitute a feedback loop that can be directly regulated by sensory processing within the OB, little is actually known about how and when cholinergic inputs to the olfactory system are activated during olfactory behavior. To investigate the relationship between olfactory processing and cholinergic modulation, we are obtaining extracellular recordings from the nucleus of the horizontal limb of the diagonal band of Broca (HDB), a component of the basal forebrain cholinergic system projecting almost exclusively to the olfactory system, in awake, freely-moving rats engaged in an olfactory cross-habituation task that is known to be influenced by cholinergic modulation in the OB (Mandairon et al., 2006). Preliminary recordings, obtained from single neurons as well as multi-neuron clusters, suggest that neurons in the HDB are tonically active during the waking state, with spontaneous rates ranging from 0.79 to 43.2 Hz (average of 6.15±3.76 Hz, n=12). In a subset of recordings (2/12, 16.7%), firing rate is significantly modulated by active investigation of olfactory stimuli, suggesting that early olfactory processing may directly activate the cholinergic inputs to the olfactory system. Acknowledgements: NIH R21 DC010420-02 to CL (DMS, co-PI)

#P217 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Regulation of Kv1.3 Channel Current Density by the Ubiquitin Ligase Nedd4-2

Patricio Velez[1], Debra A Fadool[1,2]
[1]Dept Biological Sciences,Florida State University Tallahassee, FL, USA, [2]Programs in Neuroscience & Molecular Biophysics, Florida State University Tallahassee, FL, USA

The Kv1.3 potassium channel is a main contributor to olfactory bulb (OB) electrical signaling. Gene-targeted deletion of Kv1.3 causes a *super-smeller* phenotype and resistance to obesity. Kv1.3 current density, therefore, not only modulates OB excitability, but also basal metabolism. Controlling current density could thus be potentially therapeutic for balancing energy expenditure or enhancing olfactory ability. Since Kv1.3 contains PY motifs, we explored whether Kv1.3 expression and current density is controlled by the E3 ubiquitin ligase, Nedd4-2, and the channel-interacting adaptor protein Grb10. We transiently expressed Kv1.3 in HEK293 cells and measured current density by cell-attached patch-clamp before and after co-expression with Nedd4-2 and Grb10. In response to a depolarization from -90 to +40 mV, Kv1.3 peak current amplitude was 530±95 pA (mean ±SEM, n=29). Co-expression of Kv1.3 with Nedd4-2 reduced the peak current to 220±30 pA (n=36). This reduction was due to the specific action of Nedd4-2 because co-expression of Kv1.3 with an inactive mutant Nedd4-2 produced no significant change in Kv1.3 current density (519±65 pA, n=28). Incubation with the proteasomal inhibitor MG-132 blocked Nedd4-2-induced reduction of Kv1.3 current density. These results indicate that Kv1.3 is a target for ubiquitination by Nedd4-2 and subsequent degradation by the proteasome. When Kv1.3 was simultaneously co-expressed with Nedd4-2 and Grb10, the degree of channel degradation was significantly lower (366±45 pA, n=33) than with Nedd4-2 alone, suggesting that Grb10 modulates the interactions between Kv1.3 and Nedd4-2. Taken together, our results indicate that Kv1.3 current density is regulated by a biochemical complex involving the channel, Nedd4-2 and Grb10. Acknowledgements: This work was supported by: NIH NIDCD DC003387

#P218 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Wnt5 and Drl Regulate Dendritic Targeting in *Drosophila* Olfactory Map Formation

Huey Hing[1], Yuping Wu[2]
[1]SUNY Brockport, Department of Biology Brockport, NY, USA, [2]University of Washington, Department of Pharmacology Seattle, WA, USA

In the development of the *Drosophila* antennal lobes, targeting of the dendrites of the projection neurons (PNs) is alone sufficient to create the forerunner of the olfactory map. But the mechanisms regulating PN dendrites targeting are unclear. We show that the secreted protein, Wnt5, is expressed in the nascent antennal lobe, and its receptor, Drl is expressed by the dendrites of the PNs. Loss of function of *Wnt5* led to PN dendrites targeting towards the dorsolateral region of the antennal lobe, while loss of function

of *Drl* led to PN dendrites targeting towards the ventromedial region of the antennal lobe. The loss-of-function *Drl* defects are suppressed by the removal of one copy of the *Wnt5* gene, indicating that *Wnt5* and *Drl* function antagonistically. Based on our findings, we propose that Wnt5 acts as a guidance cue that directed PN dendrites ventrally, and that Drl inhibits Wnt5, allowing PN dendrites to move dorsally. We also present evidence of an unknown ventrally localized dendritic repulsive force. Our findings begin to define the molecular and cellular mechanisms that guide PN dendrites movement and pattern the early olfactory map. Acknowledgements: NIH NIDCD

#P219 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Glia-neuron interactions in the formation of glial networks investing the olfactory nerve

Mounir Koussa, Leslie P. Tolbert, Lynne A. Oland
Universoty of Arizona/Dept. of Neuroscience Tucson, AZ, USA

The olfactory nerves of mammals and moths share a similar organization: groups of small-diameter olfactory receptor axons are ensheathed in bundles by glial cells. In the developing olfactory (antennal) nerve (AN) of the moth *Manduca sexta*, the axons of olfactory receptor neurons (ORNs) grow from the olfactory sensory epithelium toward the olfactory lobe, and glial cells from the olfactory epithelium migrate in long chains along the axons at stages when the AN glial cells are highly dye coupled. Phospho-histone 3 labeling indicates that AN glial cells proliferate during the period of axon ingrowth and glial migration, peaking at the earlier stages; we do not know if actively migrating glia also are dividing. EM at different stages shows the extension of glial processes that gradually form a network of processes ensheathing axon bundles. Whole-cell recordings from AN glia *in vivo* revealed two types of potassium currents and at least two types of calcium currents. The L-type Ca^{++} channel blocker, verapamil, blocks the majority of the Ca^{++} current. When co-cultured with ORNs, glia dissociated from the AN form networks. Verapamil exposure had no effect on the organization of these *in vitro* networks nor did it have apparent effects on the network's development *in vivo*. Experiments to block the non-verapamil-sensitive current are ongoing. Finally, during axon ingrowth, spontaneous Na-dependent activity can be recorded in the AN. Acute bath exposure to TTX *in vivo* showed no effect on the glial Ca^{++} current. However, recordings from glial cells chronically exposed to TTX (over two stages of development), at a dose previously shown to silence neuronal activity in the brain, showed a reduction in the Ca^{++} current, implicating interaction between ORNs and glial cells in the development of glial Ca^{++} currents. Acknowledgements: NIH NIDCD DC008597

#P220 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Patterns of Glomerular Innervation Recover Following Repeated Detergent Treatment of the Olfactory Epithelium in Zebrafish

Taylor R Paskin, Christine A Byrd-Jacobs
Western Michigan University Kalamazoo, MI, USA

We have shown previously that the olfactory system of adult zebrafish is capable of recovery following repeated degeneration with the detergent Triton X-100, but the specific innervation patterns in the bulb have not been examined. The objective of our current study is to examine glomerular staining patterns using specific antibody markers in control, detergent-treated, and recovered animals. Cryostat sections of zebrafish noses and brains were stained with anti-calretinin, anti-keyhole limpet hemocyanin (KLH), anti-Gαo, anti-Gαolf, and anti-Gαs/olf. All antibodies stained olfactory sensory neuron axons, and each antibody labeled glomeruli differentially. In control olfactory bulbs, anti-KLH appeared to label all glomeruli, anti-calretinin labeled many, but not all, glomeruli and each of the three G-proteins showed unique regional staining patterns. Anti-Gαolf stained the anterior region of the olfactory bulb, anti-Gαo stained clusters of glomeruli in the dorsal region as well as part of the medial region, and anti-Gαs/olf had extensive labeling with individual glomeruli stained in the dorsal, anterior, and medial regions of the olfactory bulb. Each G-protein is present in only a subset of the total neuron population in the olfactory epithelium since the staining in the bulb varies with each protein. In detergent-treated fish stained with anti-KLH or anti-calretinin, the glomeruli were diffusely labeled, while fish allowed to recover had well-defined glomeruli that were more intensely labeled. These results suggest that glomerular patterns of innervation can recover following repeated degeneration of the epithelium further illustrating the tremendous plasticity of this system. Acknowledgements: WMU Graduate Student Research Fund and WMU FRACA Award #09-019

#P221 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Role of Kirrel-3 in Wiring the Accessory Olfactory System

Janet E.A. Prince[1,2], Tyler Cutforth[3], Jean-François Cloutier[1,2]
[1]Montreal Neurological Institute Montreal, QC, Canada, [2]Department of Neurology and Neurosurgery, McGill University Montreal, QC, Canada, [3]Department of Molecular, Cell & Developmental Biology, University of California Santa Cruz, CA, USA

The olfactory systems play a critical role in the survival and mating behavior of most terrestrial vertebrates. Two different classes of odorants, general odorants and pheromones, are processed by the olfactory systems and convey several cues in vertebrates such as the presence of danger or of food, as well as social and sexual cues. In both the main and accessory olfactory systems, axons of chemosensory neurons must converge in the olfactory bulbs and form stereotypic connections with second order neurons in structures termed glomeruli. The proper convergence of olfactory axons into glomeruli and the formation

of these stereotyped connections are essential for olfactory function. In the main olfactory system, the Kirrel family members have been implicated as homophilic adhesive molecules that play an important role in axon-axon interactions[4]. These interactions have been suggested to regulate olfactory nerve fasciculation and olfactory convergence; both crucial features of accurate glomerular map formation. In contrast to the main olfactory system, the molecular mechanisms that regulate glomerular axonal convergence in the accessory olfactory system are poorly understood. To begin to investigate the role of the Kirrel family members in the development of the accessory olfactory system, we have performed a detailed analysis of the spatio-temporal patterns of expression of these molecules by in situ hybridization and immunohistochemistry in the vomeronasal organ and the accessory olfactory bulb. [4]Serizawa, S., Miyamichi, K., Takeuchi, H., Yamagishi, Y., Suzuki, M., Sakano, H. (2006). A neuronal identity code for the odorant receptor-specific and activity-dependent axon sorting. Cell 127, 1057-1069. Acknowledgements: CIHR and FQRNT

#P222 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Preferential Expression of Presynaptic Fragile X Proteins in Newly Mature Olfactory Sensory Neuron Axons

Michael R. Akins, Emily E. Stackpole, Eunice Chyung, Justin R. Fallon
Brown University Dept of Neuroscience Providence, RI, USA

Transient experiences are converted into long-lasting structural changes at synapses through the production of new proteins. A critical subset of this synthesis occurs within the synaptic compartment itself. While this process has been extensively characterized at postsynaptic sites, recent evidence highlights the potential importance of translation within the presynaptic compartment. The potential contribution of local presynaptic translation to olfactory circuit formation is largely unexplored. Studies from our laboratory have demonstrated that the RNA-binding protein FMRP (Fragile X mental retardation protein) is expressed in olfactory sensory neuron (OSN) axons and terminals, suggesting that FMRP-regulated local translation may modulate presynaptic formation and function in these axons. FMRP and its homologues FXR1P and FXR2P are transiently expressed in OSN axons in discrete granules (Fragile X Granules; FXGs) that are also observed in neuropil of select additional brain regions including the cerebral cortex, hippocampus, and cerebellar cortex. To better understand FXG function, we have systematically analyzed FXG expression in the mouse olfactory bulb. We find that the protein composition of olfactory FXGs mirrors that of FXGs in hippocampal mossy fibers but is distinct from that seen in other brain regions. Furthermore, we show that FXGs are expressed preferentially by newly mature axons (that express olfactory marker protein) but are sparse in growing or degenerating axons. Together with companion work showing altered synapse formation in FMRP knockout mice, these data point to a contribution of FMRP-regulated local protein synthesis to the appropriate integration of newly generated olfactory sensory neurons into existing olfactory circuits.
Acknowledgements: F32DA021501, K99MH090237 (M.R.A.) and HD052083 (J.R.F.).

#P223 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

A Role for Presynaptic FMRP in Olfactory Sensory Neuron Synapse Formation

Hanna E Berk-Rauch[1], Emily E Stackpole[2], Michael R Akins[3], Justin R Fallon[3]
[1]Graduate Program in Molecular Biology, Cellular Biology and Biochemistry, Brown University Providence, RI, USA, [2]Neuroscience Graduate Program, Brown University Providence, RI, USA, [3]Department of Neuroscience, Brown University Providence, RI, USA

Elucidating the molecular underpinnings of synapse formation and plasticity in olfactory sensory neurons (OSNs) is essential for understanding how information is processed in the olfactory system. One compelling candidate molecule that may influence this process is Fragile X mental retardation protein (FMRP), an RNA binding protein that regulates synaptic plasticity by modulating local protein synthesis and mRNA stability. In previous work, we demonstrated that FMRP is expressed in OSN axons and presynaptic terminals. In companion work we show that FMRP is expressed in granules (Fragile X granules) as newly born OSNs are integrating into existing circuitry, suggesting that FMRP may function in OSN synapse formation. Here we have used both in vivo and tissue culture models to test the role of presynaptic FMRP in synapse formation and maintenance. Presentation of the synaptogenic molecules neuroligin-1 or FGF22 induced robust presynaptic differentiation in cultured wild type OSNs. In contrast, neither molecule induced a response above baseline in FMRP null OSNs. Thus FMRP in the presynaptic neuron is required for at least two distinct modes of presynaptic differentiation. To address possible molecular mechanisms underlying these defects, we asked whether mRNA stability of a candidate set of presynaptic proteins is altered in FMRP null olfactory epithelium. At least one presynaptic message, that encoding neurexin 3α, is greatly reduced in the absence of FMRP. Finally, preliminary in vivo evidence indicates that FMRP is required for appropriate synapse formation and/or maintenance in the intact olfactory bulb. These data indicate that presynaptic FMRP may be a key component in regulating the formation and maintenance of OSN synapses.
Acknowledgements: HD052083 (J.R.F.) and F32DA021501, K99MH090237 (M.A.)

#P224 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

OSN overproduction caused by immature OSN-specific expression of the constitutively active G protein-coupled receptor, GPR12

Paula M Heron[1], Jeremy McIntyre[2], Timothy S McClintock[1]
[1]Department of Physiology, University of Kentucky Lexington, KY, USA, [2]Department of Pharmacology University of Michigan Ann Arbor, MI, USA

In addition to transducing odor signals, odorant receptors (ORs) also help determine where OSN axons terminate in the olfactory bulb. Anterior-posterior locations appear to be determined by

OR-generated cAMP (via Gαs) regulating the reciprocal expression of type I axon guidance proteins, Neuropilin 1 and Plexin-A1, in immature olfactory sensory neurons (OSNs). To further investigate the role for cAMP in the axon targeting of immature OSNs we made GPR12-tauGFP transgenic mice. This transgene, which produced a 3-10 fold increase in cAMP when expressed in HEK293 cells, is under the control of a promoter specific to immature OSNs and produces a bicistronic mRNA encoding GPR12, followed by tauGFP. As expected, GFP fluorescence in these mice is strongest in the immature OSN layer and fills their axons. Transgenic mice show increased phospho-CREB immunoreactivity in OSN nuclei and increased tyrosine hydroxylase immunoreactivity in the glomerular layer of the olfactory bulb, consistent with the expectation of elevated cAMP levels compared to wildtype mice. Intriguingly, preliminary studies find evidence of increased numbers of OSNs in GPR12-tauGFP transgenic mice, arguing that increased cAMP production in immature OSNs improves their ability to survive and differentiate into mature OSNs. Not surprisingly, this increase in OSN number appears to be accompanied by increased apoptosis of mature OSNs. Preliminary tests with markers of basal cell proliferation have thus far given conflicting results, so whether increased cAMP production in immature OSNs also indirectly increases basal cell production of nascent OSNs remains to be determined. Other experiments to explore effects on glomerular convergence and location in the olfactory bulb are in progress. Acknowledgements: F32 DC011427 to PMH and R01 DC002736 to TMc

#P225 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Role of GPRC5 receptors in the murine olfactory epithelium

Eva M Neuhaus[1], Stefan Kurtenbach[2], Thomas Pelz[1]
[1]Charite Berlin, Germany, [2]Ruhr-Universität Bochum Bochum, Germany

Neurogenesis in the olfactory epithelium (OE) takes place continuously throughout the whole lifespan, resulting in new neurons that grow axons to the brain where they form new connections to glomeruli in the olfactory bulb (OB). It is known that retinoids, derivates of Vitamin A, are involved in differentiation and survival of neurons. Retinoic acid (RA) is a derivate of Vitamin A and binds to retinoic acid receptors (RAR, RXR) which were shown to promote neuronal survival. In mass spectroscopy analysis of the membrane proteome from the OE we found high expression levels of RA regulated receptors with so far unknown function. These receptors comprise a class of four known receptor sequences, called GPRC5A-D, which are present only in chordates. We investigated the expression levels of these receptors in mice fed a special diet containing no vitamin A, carotenoids or other precursors of RA for different time-spans. Our aim is to get a deeper understanding of the molecular effects of vitamin A deficiency in mice. We used immunohistochemistry to get insight into the morphology of the neurons and distribution of the proteins involved in the odor induced signal transduction cascade. Additionally we made quantitative PCR analysis to observe protein expression levels and performed electro olfactogram (EOG) measurements to investigate the capability of the OE to respond to odorous stimuli.

#P226 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

An epigenetic signature for monoallelic olfactory receptor expression

Stavros Lomvardas
UCSF/Anatomy San Francisco, CA, USA

Constitutive heterochromatin is traditionally viewed as the static form of heterochromatin that silences pericentromeric and telomeric repeats in a cell cycle and differentiation independent manner. Here, we show that in the mouse olfactory epithelium, olfactory receptor (OR) genes are marked, in a highly dynamic fashion, with the molecular landmarks of constitutive heterochromatin. The cell-type and differentiation dependent deposition of H3K9me3 and H4K20me3 along the OR clusters is, most likely, reversed during the process of OR choice. In contrast to the current view of OR choice, our data suggest that OR silencing takes place before OR expression, indicating that it is not the product of an OR-elicited feedback signal. This implies that chromatin mediated silencing provides the molecular prerequisite upon which a mechanism of singular and stochastic OR selection can be applied. Acknowledgements: McKnight Endowment Fund for Neuroscience R03 DC010273 (NIH/NIDCD) 1DP2 OD006667-01 NIH Director's New Innovator Award NIH/Roadmap for Epigenomics NIH/EUREKA

#P227 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

MeCP2 Regulates Activity-dependent Gene Expression in Olfactory Sensory Neurons

Wooje Lee, Qizhi Gong
University of California/Cell Biology and Human Anatomy Davis, CA, USA

To sculpt precise neuronal circuitry, many pathways undergo different degrees of remodeling during early postnatal stages. Neuronal activity plays a critical role in this process. In the olfactory system, depriving odorant exposure during early postnatal stages results in a compromised olfactory connectivity due to the presence of supernumerary glomeruli for each odorant receptor expressing population. Several cell adhesion molecules were shown to alter their expression levels under manipulation of odorant induced activity. In this study, we investigated the function of methyl CpG binding protein 2 (MeCP2) in the regulation of cell adhesion molecule expression in olfactory sensory neurons (OSNs) and in the formation of precise olfactory connections. MeCP2 is a transcription regulator that is shown to play critical roles in postnatal neuronal plasticity. Mutations in MeCP2 are responsible for Rett Syndrome in human. In MeCP2 knockout mice, supernumerary glomeruli were observed for specific odorant receptor populations. Several cell adhesion molecules alter their expression levels in OSNs in MeCP2 knockout mice when compared to wildtype littermates by immunocytochemistry and qRT-PCR. MeCP2 binds to predicted promoter regions of Kirrel2 and Kirrel3 evaluated by chromatin immunoprecipitation. After odorant exposure, higher levels of

promoter binding were observed suggesting that regulations of MeCP2 to Kirrel2 and Kirrel3 expression can be altered by neuronal stimulation. Changes in promoter binding were accompanied by an increase of phosphorylation at serine 80 of MeCP2 while the total level of MeCP2 remains the same before and after odorant stimulation. These findings demonstrated that MeCP2 plays critical roles in modulating activity-dependent cell adhesion molecule expression in OSNs. Acknowledgements: NIH DC011346

#P228 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Endocannabinoid System Promotes Proliferation in the Mouse Olfactory Epithelium

Chelsea R. Hutch[1,3], Colleen C. Hegg[1,2,3]
[1]Neuroscience Program East Lansing, MI, USA, [2]Pharmacology & Toxicology East Lansing, MI, USA, [3]Center for Integrative Toxicology East Lansing, MI, USA

The peripheral olfactory epithelium exhibits natural regeneration which is accelerated after injury. The endocannabinoid system has been shown to stimulate adult neurogenesis in the central nervous system. We tested the hypothesis that cannabinoid receptors are located in the olfactory epithelium and assist in proliferation. First, the presence of two cannabinoid receptors in mouse olfactory epithelium was confirmed using RT-PCR. Second, using a neonatal mouse olfactory epithelium slice model and calcium imaging, we found that cannabinoid receptors are physiologically functional, responding to the pan cannabinoid receptor agonist WIN55,212-2. Transient increases in calcium evoked by either 100 nM or 1 M WIN55,212-2 were observed in 70% (30 out of 43) of ATP-responsive sustentacular cells, and in 23% (10 of 43) of odorant-responsive cells. These data suggest that cannabinoid receptors are present on both olfactory sensory neurons and sustentacular cells. We then tested if cannabinoid receptors regulate proliferation in the olfactory epithelium. Adult mice were intranasally administered either saline vehicle or 10 M WIN55,212-2 to activate cannabinoid receptors and bromodeoxyuridine (BrdU) was administered by intraperitoneal injection 45 hours later to measure proliferation. There was a significant increase in BrdU+ cells in WIN55,212-2 treated animals over vehicle control animals (26.9 ± 2.2 vs. 20.1 ± 1.0 BrdU+ cells/mm, $p<0.05$). Administration of the specific cannabinoid antagonist, AM-251 inhibited the WIN55,212-2 induced increase to control levels (15.7 ± 0.8 vs. 20.1 ± 1.0 BrdU+ cells/mm, $p<0.05$). Our data identifies expression of functional cannabinoid receptors in the mouse olfactory epithelium and suggests endocannabinoids may play a role in proliferation in the olfactory system. Acknowledgements: Support from NIDCD 006893.

#P229 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

A role for apical translation in the control of olfactory mucosa survival?

Christine Baly[1], Marie-Annick Persuy[1], Sylvie Souquere[2], Didier Durieux[1], Caroline Dubacq[3], Gérard Pierron[2], Jean-Jacques Rémy[4], Monique Caillol[1]
[1]INRA UR1197 78350 Jouy-en-Josas, France, [2]CNRS FRE-3238 94800 Villejuif, France, [3]UMR7102 UPMC 75005 Paris, France, [4]UMR 6184 USC INRA-Phase 13015 Marseille, France

In mammals, olfactory sensory neurons (OSN) are located at the interface between environment and the brain for proper odorant coding by the olfactory mucosa (OM). These neurons have developed specialized structures, the olfactory dendrites that contain elements of the olfactory transduction pathways. Moreover, several data show that odorant stimulation triggers other biological effects on OM, such as modulation of cell survival, stress response and synaptic plasticity. Upon the screening of a cDNA library enriched in neuronal dendrites ends, we have identified several mRNAs linked to survival functions for which we hypothesise a local translation for rapid responses upon exogenous stimuli. We examined whether the apical part of the rat OM, and particularly the OSN dendrites, is enriched in structures connected with RNA processing or to translation. We used combined immunohistochemistry and biochemical approaches to localize proteins involved either in mRNA translation or in RNA processing/storage and we purified polysomal fractions to check their translatability. Antibodies directed against known markers of RNA granules revealed the presence of dots scattered in the apical part of the OM, both in OSN and non-neuronal cells. Apical immunoreactivity for both the ribosomal P protein and the phosphorylated form of the translation factor Eif2, known to be involved in the translation arrest in polarized cells, strongly support a translation control in this zone. RT-PCR analyses of polysomes collected by cell sub-fractionation confirmed that some RNAs are engaged in translation. The fine ultrastructure of the apical region of the OM supports these results. Altogether, we provide evidence for a local translation in apical part of rat OM cells.

#P230 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Proliferation in the olfactory epithelium as it varies with aging and injury

Jessica H Brann[1], Stuart Firestein[1,2]
[1]Columbia University, Department of Biological Sciences New York, NY, USA, [2]Columbia University, Program in Neurobiology and Behavior New York, NY, USA

The olfactory epithelium (OE) is unique in that it exhibits plasticity throughout life, beyond a normal short developmental period. The olfactory system provides an opportunity to examine the role of environmental versus biological causes of aging because it is composed of both the main and accessory olfactory systems. In previous work, we examined the regenerative capacity

of the vomeronasal epithelium (VNE). There, we investigated the rates of proliferation and apoptosis in the VNE over the lifespan of the mouse. We showed that the aged VNE responds robustly to a lesion challenge. These data suggest the progenitor cell in the VNE retains its proliferative capacity throughout life. Here, we investigate whether the OE exhibits a similar or different aging strategy, as it must cope with constant exposure to odorants, airborne viruses, and toxins, in contrast to the more protected VNE. In initial studies, we established the basal rates of proliferation and apoptosis in the basal OE over the lifespan of the mouse. At 2 months of age, BrdU incorporation was significantly higher ($p < 0.001$) than that seen in either 6 or 24 month old mice. We next asked whether the ability to respond to acute injury, namely olfactory bulbectomy (OBX), also decreases with age. OBX results in rapid death of mature sensory neurons within five days, followed by the massive proliferation of basal cells and partial reconstitution of the epithelium within 30 days. Unilateral OBX was performed on mice 2, 6, and 24 month of age; animals recovered for 5 days and were evaluated for BrdU incorporation. BrdU labeling was significantly increased in the OBX OE versus non-surgery control in all age groups, suggesting that while proliferation rate is normally low in OE of old animals, this rate increases when challenged with an injury. Acknowledgements: J.H.B. supported by F32 DC008455.

#P231 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

ATP Mediates Neuroprotective and Neuroproliferative Effects in Mouse Olfactory Epithelium Following Exposure to Satratoxin G In Vitro and In Vivo

Cuihong Jia[1], Beth Belock[1], Sutheera Sangsiri[1], James J. Pestka[2], Colleen C. Hegg[1]
[1]Department of Pharmacology & Toxicology, Michigan State University East Lansing, MI, USA, [2]Food Science and Human Nutrition, Michigan State University East Lansing, MI, USA

Intranasal aspiration of satratoxin G (SG), a mycotoxin produced by the black mold *Stachybotrys chartarum,* selectively induces apoptosis in olfactory sensory neurons (OSNs) in mouse olfactory epithelium (OE) through unknown mechanisms. Here, we confirm SG-induced OSN apoptosis occurs in OE primary cell culture, the OP6 olfactory placodal cell line, and *in vivo*. Incubation of SG with OE primary cell culture or OP6 cells for 24 hrs significantly increased apoptosis as measured by increased propidium iodide or activated caspase-3 labeling. Intranasal aspiration of SG significantly increased TUNEL staining in the middle neuronal layer of the OE at 3 days, indicating that SG selectively induces OSN apoptosis. Next, we investigated whether ATP protects against SG-induced injury. ATP alone did not decrease apoptosis, but inhibition of purinergic receptors (PPADS and suramin) significantly increased apoptosis in OE primary cell culture and in vivo, suggesting that purinergic receptors are involved in neuroprotection. Further, intranasal aspiration of ATP significantly reduced SG-induced OSN apoptosis, indicating that ATP has a neuroprotective function in SG-induced OE injury. We investigated whether ATP has a neuroproliferative function in SG-induced injury by measuring BrdU incorporation. We found that the number of BrdU+ cells was significantly increased at 3 and 6 days post-SG aspiration. Treatment with purinergic receptor antagonists significantly reduced SG-induced cell proliferation, while post-treatment with ATP significantly potentiated SG-induced cell proliferation. These data indicate that ATP is released and promotes cell proliferation via activation of purinergic receptors in SG-induced OE injury. The purinergic system is a therapeutic target to alleviate or restore the loss of OSNs. Acknowledgements: NIDCD 006897 (CCH), Michigan State University Respiratory Research Initiative (JJP), Michigan State University Foundation Strategic Partnership Grant (JJP), NIEHS ES03358 (JJP)

#P232 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

MMP-9 is associated with the early inflammatory response following olfactory injury

Stephen R. Bakos, Richard M. Costanzo
Virginia Commonwealth University/Department of Physiology and Biophysics Richmond, VA, USA

The olfactory system has a remarkable capacity for regeneration and recovery following injury, however the molecular mechanisms underlying recovery remain poorly understood. We previously reported that matrix metalloproteinase-9 (MMP-9) levels in the olfactory bulb increased immediately after nerve transection and remained elevated for approximately two weeks. The importance of this early expression of MMP-9 in the bulb remains unknown, though in other regions of the CNS, MMP-9 has been linked to the inflammatory response. In this study, we used immunohistochemistry to examine the relationship between MMP-9 in the bulb and the inflammatory response to olfactory injury. Two markers for inflammatory leukocytes were used, myeloperoxidsae (MPO) to identify neutrophils and CD68 to indentify macrophages. Olfactory bulbs were examined at days 1, 7, and 10, time periods when MMP-9 levels are elevated. We found that MMP-9 was localized to MPO positive cells at all 3 time periods, demonstrating that MMP-9 was contained within neutrophils. In contrast, MMP-9 did not localize to CD68 positive cells. The association of MMP-9 with neutrophils, and not macrophages, suggests that MMP-9 plays an important role in the early inflammatory response to olfactory injury. This finding contributes to our understanding of the molecular mechanisms underlying the early response to injury. Future studies aimed at modulation of MMP-9 during the early injury response will determine its potential as a novel therapeutic target for improved olfactory regeneration and recovery. Acknowledgements: Support by NIH grant DC00165

#P233 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Neuregulin1 Promotes Olfactory Epithelial Sphere Formation In Culture

Woochan Jang, Melissa A. Donovan, James E. Schwob
Tufts University School of Medicine/Anatomy and Cellular Biology Boston, MA, USA

Three-dimensional (3D) air-media interface cultures of the adult olfactory epithelium (OE) reliably sustain progenitor cell potency

and closely mimic epithelial regeneration *in vivo*, as long as a feeder layer of 3T3 cells or of a lamina propria-derived line (LP_{Imm}) is there to stimulate sphere formation robustly. We tested whether selected growth factors added to the media can substitute for the stimulatory effects of the feeder cells on sphere formation, extent of sphere growth, and cellular composition of the spheres. Among the growth factors we tested, neuregulin 1 (NRG1), which we have shown to be a component of LP_{Imm} cell-conditioned media, was the most effective at stimulating sphere formation. As compared to control, serum-containing media, both the size and the number of spheres were significantly increased. The cellular composition of the spheres was assayed as a function of different growth factors including NRG1 both by staining with different OE markers and using transgenic mice (e.g., *OMP-GFP* and *Sox2-GFP* mice) to confirm whether different growth factors positively/negatively regulate different olfactory epithelial lineages. That the sphere formation and expansion achieved in response to either a feeder cell layer or media conditioned to the point of feeder cell growth exhaustion exceeded the spherogenesis achieved with the use of single factors (including NRG1) suggests that multiple signals are required to maximize sphere growth and for the fullest recapitulation of epitheliopoiesis *in vivo*. In summary, our 3D insert assay provides a new avenue to dissect the signaling influences exerted by the lamina propria on the olfactory epithelium during its maintenance and regeneration. Acknowledgements: NIH R21 DC010920

#P234 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Activation of Horizontal Basal Cells in the Olfactory Epithelium is Required for Multipotency

Nikolai Schnittke[1,2], Adam I Packard[1,2], James E Schwob[1]
[1]Tufts University School of Medicine/Anatomy and Cellular Biology Boston, MA, USA, [2]Tufts University Sackler School of Graduate Biomedical Sciences/Program in Cell, Molecular, and Developmental Biology Boston, MA, USA

The olfactory epithelium (OE) has a remarkable capacity to regenerate its neuronal and non-neuronal populations throughout adult life. At least two distinct progenitor cells types are able to subserve epitheliopoiesis. Globose basal cells (GBCs) are multipotent and can generate all epithelial cell types following transplantation, independent of the status of the donor epithelium (i.e., lesioned or not). A second type of multipotent cell, the horizontal basal cell (HBC), adheres directly to the basal lamina. HBCs contribute to epithelial regeneration only after severe injury (e.g., following methyl bromide [MeBr] exposure) that ablates all the differentiated cells as well as a substantial portion of the GBC population. Thus, in contrast to GBCs (from which they emerge during embryonic development), HBCs appear to function as a reserve progenitor population that requires activation by injury. We tested that hypothesis directly by transplanting FACS-purified HBCs from MeBr-lesioned donor epithelium (activated HBCs) vs. normal epithelium (dormant HBCs) into a transplant-competent (MeBr-lesioned) animal. We find that HBCs can engraft and generate the different cell types of the OE only after isolation from MeBr-lesioned epithelium, but not from normal OE. As expression of the transcription factor p63 is required for the embryonic emergence of HBCs, we hypothesize that downregulation of p63 is an important feature of HBC activation. Indeed, IHC analysis reveals a transient reduction in p63 expression in HBCs 1 day after lesion. Conversely, over-expression of p63 in a post-lesion environment by retroviral transduction causes proliferating progenitor cells to differentiate into HBCs. Thus, manipulation of p63 levels looks to be critical to activation of HBCs and their potential therapeutic use. Acknowledgements: F30 DC011241 (NS), R01 DC002167 (JES)

#P235 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Diet-induced obesity affects structure and function of the olfactory system

Suk-Hee Cho[1,3], Christopher Kovach[1], Kristal Tucker[1,3], James M. Overton[2,3], Michael Meredith[1,3], Debra A. Fadool[1,3,4]
[1]The Florida State University/Department of Biological Science Tallahassee, FL, USA, [2]The Florida State University/Department of Biomedical Sciences Tallahassee, FL, USA, [3]The Florida State University/Program in Neuroscience Tallahassee, FL, USA, [4]The Florida State University/Institute of Molecular Biophysics Tallahassee, FL, USA

Mice with gene-targeted deletion of the voltage-dependent potassium channel, Kv1.3, are resistant to diet-induced obesity (DIO) when maintained on a moderately high-fat diet (MHF, 32% fat) for 26 weeks, due to an increase in basal metabolic rate (BMR). Bilateral olfactory bulbectomy causes a loss in resistance to DIO, whereby mice exhibit a 30% increase in body weight, a decrease in BMR, and an increase in adiposity. To determine the impact of DIO on olfactory bulb function, the excitability of Kv1.3-expressing mitral cells was measured via slice electrophysiology in MHF-fed mice. Following a 30 day MHF diet, mitral cells failed to fire full amplitude action potentials, exhibited spike adaptation, and developed a resistance to insulin-evoked enhanced spike frequency. Following a 60 day MHF diet, pause durations between spike clusters were shortened, cluster length was reduced, and modulation by insulin remained subdued. Considering the energetic demands of neuronal activity, we found that mitochondrial size within the mitral cell layer increased following DIO while total abundance decreased. Mitochondrial size was smaller in Kv1.3-null animals, which concomitantly were not affected by the fat diet. Presynaptically, the MHF diet evoked a 45-52% loss in M72-expressing OSNs, independent of the expression of the Kv1.3 channel and thus body weight gain. OB-targeted delivery of the Kv1.3 pore blocker, ShK-186, via an osmotic mini-pump designed for infusion over five days, evoked an increase in BMR in wildtype mice fed either a control or MHF diet. These data indicate that obesity or a change in energy balance causes loss of OSNs, change in OB mitochondria, and perturbation of metabolic sensing that is normally performed in mitral cells at the level of Kv1.3 channel. Acknowledgements: This work was supported by: NIH NIDCD DC003387 & DC00044 and support from the Tallahassee Memorial Regional Medical Center (TMRMC) Robinson Foundation.

#P236 POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING

Local and Regional Network Function in Behaviorally Relevant Cortical Circuits of Adult Mice Following Postnatal Alcohol Exposure

Benjamin Sadrian[1,2], Donald Wilson[1,2], Jesse Peterson[1], Balapal Basaravaj[3,4], Mariko Saito[5,6]
[1]Emotional Brain Institute, Nathan S. Kline Institute for Psychiatric Research Orangeburg, NY, USA, [2]Department of Child and Adolescent Psychiatry, New York University Langone Medical Center New York, NY, USA, [3]Division of Analytical Psychopharmacology, Nathan S. Kline Institute for Psychiatric Research Orangeburg, NY, USA, [4]New York State Psychiatric Institute, Department of Psychiatry, College of Physicians & Surgeons, Columbia University New York, NY, USA, [5]Division of Neurochemistry, Nathan S. Kline Institute for Psychiatric Research Orangeburg, NY, USA, [6]Department of Psychiatry, New York University Langone Medical Center New York, NY, USA

Ethanol consumption during pregnancy often leads to Fetal Alcohol Spectrum Disorder (FASD), which consists of a complete spectrum of developmental deficits. Ethanol exposure in neonatal rodents induces widespread apoptotic neurodegeneration and long-lasting behavioral abnormalities similar to FASD. Here we used multi-site local field potential (LFP) recording and behavioral analyses to examine how binge exposure to ethanol affects information flow through the late developing olfactory system. Male and female C57BL/6By mice were injected with ethanol (20%, 2.5g/kg) or an equal volume of saline (controls) on postnatal day 7 (P7). At this age, the olfactory system was largely spared from cell death. Mice were tested at 3 months on a piriform cortex-dependent odor habituation task, a hippocampal-dependent place memory task, and had spontaneous and odor-evoked LFP activity examined. No impairment of odor investigation or habituation was observed. However, hippocampal-dependent memory was significantly impaired. In parallel with these behavioral outcomes, spontaneous LFP's were unaffected in the olfactory bulb, but depressed in both the piriform cortex and dorsal hippocampus. Odor-evoked activity expressed relative to basal activity was normal in both the olfactory bulb and piriform cortex of P7 ethanol treated mice, however, the hippocampal formation showed hyper-excitability/hyper-synchrony to this input compared to controls. In addition, functional coherence between the piriform cortex and dorsal hippocampus was enhanced in the beta frequency range in P7 ethanol treated mice compared to controls. These results suggest that P7 ethanol induces long-lasting, system-specific changes in local circuit and regional network function that correspond to system-specific changes in neurobehavioral performance. Acknowledgements: NIH/NIAAA R01 AA015355 to M.S. NIH/NIAAA R01 AA019443 to B.S.B. NIH/NIDCD R01 DC003906 to D.A.W.

#P237 POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING

Degeneration of olfactory eloquent structures in an animal model of Niemann-Pick disease

Martin Witt[1], Robert Ladegast[2], Volker Gudziol[2], Thomas Hummel[2], Arndt Rolfs[3], Andreas Wree[1]
[1]University of Rostock, Department of Anatomy Rostock, Germany, [2]Technical University of Dresden Medical School, Smell & Taste Clinic, Department of Otorhinolaryngology Dresden, Germany, [3]Albrecht-Kossel Institute for Neuroregeneration, University of Rostock Rostock, Germany

Niemann-Pick type C disease (NPC) is a rare autosomal recessive lipid storage disease characterized by progressive neurodegeneration. We used a knock-out mouse model (NPC1-/-) to examine the effects of this disorder to morphologically distinct regions of the olfactory system. For histochemistry, we applied antibodies against a series of neuronal and glia marker proteins, proliferation antigens, apoptotic and macrophage markers. Mutant animals present myelin-like lysosomal deposits in virtually all types of cells of the peripheral and central olfactory system. Especially supporting cells of the olfactory epithelium and central glia cells are affected resulting in astrocytosis and microgliosis in the olfactory bulb and other olfactory cortices. Unmyelinated olfactory afferents of the lamina propria seem less affected than ensheathing cells. Interestingly, first data suggest that new neurons such as neuroblasts of the subventricular zone and the rostral migratory stream do not contain pathological inclusions. Electroolfactograms of the olfactory mucosa suggest that NPC1-/- animals exhibit severe olfactory deficits. Treatment with an inhibitor of glucosylceramide synthase, miglustat, may prevent the accumulation of glycosphingolipids and is likely to improve olfactory function. These changes will be investigated based on the present model including immunohistochemistry and electrophysiology.

#P238 POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING

The Smell of Disease: Human Body Odor Changes in Response to Systemic Inflammation

Mats J Olsson[1], Bianka Karshikoff[1], Amy R Gordon[1,2], Bruce A Kimball[2,4], Malin Brodin[1], Johan N Lundström[1,2,3], Anne Soop[1], Mats Lekander[1], Nishteman Hosseini[1], John Axelsson[1]
[1]Karolinska Institutet Stockholm, Sweden, [2]Monel Chemical Senses Center Philadelphia, PA, USA, [3]University of Philadelphia Philadelphia, PA, USA, [4]USDA-APHIS-WS-National Wildlife Research Center Philadelphia, PA, USA

Ability to detect diseases in conspecifics would be advantageous for the individual. In line with this, rodents avoid body odors of infected individuals. To learn whether this is possible by way of human smell, we tested whether body odor would reveal an acute inflammatory response to endotoxin. Eight individuals donated odorous body substances two times by wearing cotton T-shirts following an injection of endotoxin (0.8 ng lipopolysaccharide / kg body weight) at one time and placebo (Saline) at the other. In each case, T-shirts were collected from the donors after 4

hours. The armpit regions of the shirt were cut out, put in plastic bottles, frozen to -35C, and were thawed one hour before presentation. Forty naive panelists smelled all 16 T-shirts from the plastic bottles twice and judged their pleasantness, intensity, and healthiness. Preliminary results were that endotoxin-exposed individuals smelled less pleasant [$F(1,39) = 37.67$, $p < .000$, $\eta_p^2 = .49$], more intense [$F(1,39) = 32.62$, $p < .000$, $\eta_p^2 = .46$], and less healthy [$F(1,39) = 4.22$, $p = .047$, $\eta_p^2 = .10$]. The effect size of the perceived difference between body odors of endotoxin- and placebo-exposed donors was largest for pleasantness and smallest for health. The T-shirt samples were also submitted to GC/MS analysis. Preliminary inspection of the GC/MS data collected from the t-shirt samples suggests some subtle differences between placebo and endotoxin body-odor samples. These data support the notion that humans can smell the disease of others. The nature of this perception is discussed and is currently pursued in a follow-up study. Acknowledgements: This study was supported by grants from the Swedish Research Council (2009) (to MJO), National Institutes of Health – NIDCD (R03DC009869) (to JNL), and Osher Center for Integrative Medicine (to JA)

#P239 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Olfactory Hallucinations in Primary Headache Disorders

Elisheva R Coleman[1], Brian M Grosberg[2], Matthew S Robbins[2]
[1]Albert Einstein College of Medicine Bronx, NY, USA,
[2]The Montefiore Headache Center, Department of Neurology, Albert Einstein College of Medicine Bronx, NY, USA

Objective: To gain greater understanding of the phenomenon of olfactory hallucinations (phantosmias) in primary headache disorders by analyzing patients reported in the literature and those encountered at our center. **Background:** 30% of migraine patients experience aura symptoms, primarily visual. While olfactory hallucinations are well known in temporal lobe epilepsy and schizophrenia, they have been reported rarely in migraine. Understanding this phenomenon may provide insight into central olfactory processing and migraine aura physiology. **Methods:** Retrospective chart review of 13 patients with phantosmias seen at the Montefiore Headache Center from 2008-2010 and PubMed searches yielding 24 published cases of phantosmias in headache patients. Data on quality and timing of phantosmias were extracted and analyzed from the aggregate 37 cases. **Results:** Prevalence of phantosmias among patients seen at our center during the period surveyed was 0.72%. 81% of patients described phantosmia as an identifiable smell (e.g. wood smoke), while 19% described it as vague (e.g. foul). 76% perceived unpleasant smells, 11% pleasant, and 5% neutral. Commonly described odor classes were burning (20%), fecal/decay (16%), foodstuffs (14%), and chemical (10%). Regarding timing and duration of phantosmias, 49% closely preceded headache, 32% occurred during headache, and 11% occurred >2 hours prior to headache. In 82% of patients, phantosmias lasted 5-60 minutes in some or all attacks. In 14% phantosmias always lasted >1 hour, and in 1 patient <1 minute. **Conclusions:** Phantosmias are a rare but distinct manifestation of migraine, with timing similar to common aura symptoms. They are usually identifiable and unpleasant, with burning the most common odor described. Phantosmia should be recognized as a form of migraine aura. Acknowledgements: None

#P240 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Cocainization of Olfactory Epithelium Gives Short but Not Long Term Relief of Phantosmia

Donald A Leopold[1], David E Hornung[2]
[1]University of Nebraska Medical Center Omaha, NE, USA,
[2]St Lawrence University Canton, NY, USA

Because Zilsdorf (J Laryngol Nov 1966) reported it was an effective treatment, 6 patients with phantosmia were treated with topical cocaine. The phantosmias, present for an average of 11 years, could all be blocked by occluding the involved nostril(s). Four patients were treated uninasally, 2 binasally. The phantosmia was present at the time of the treatment with 1 ml of 4% cocaine HCL applied topically to the olfactory cleft. Prior to treatment, the olfactory abilities of the affected nostrils were evaluated using the 40 odor UPSIT. In 3 patients the UPSIT score was above 34 and in the other 3 it was less than 22. All patients lost both their olfactory ability and phantosmia immediately after the cocainization. In 5 patients both the olfactory ability and phantosmia returned in 6 to 54 hours. In 1 patient, the olfactory ability returned in 1 week but the phantom smell did not appear for 6 weeks. Based on Zilsdorf's recommendations, 3 of the patients had repeat cocainization at intervals of 2 weeks to 3 months, with the phantosmia relief lasting less than 5 days each time. Eighteen months after the last treatment all patients reported their phantosmias were still present, although two noted that the quality of the phantom smell had changed (more "chemical" or more "like chlorine"). The olfactory ability of 6 treated nostrils returned to the pre-treatment controls, in one nostril the UPSIT score decreased by 9 odorants and in one it improved by 8. Although the patients took some comfort in the temporarily relief from the phantom, cocaine does not seem to offer an effective long-term solution. The observation that the olfactory ability returns before the phantom might argue against the phantom being due to a rogue neuron and in favor of a central problem.

#P241 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Flavorful Eructation - A sentinel marker for chemosensory recovery

Sameer Sharma, Alan R. Hirsch
The Smell & Taste Treatment and Research Foundation Chicago, IL, USA

Both pathways – orthonasal and retronasal – have been hypothesized to be responsible for detection of different milieu viz., external and internal respectively. While chewing, the retronasal olfactory pathway seems to be more important than it's orthonasal counterpart in perceiving the flavor of the food. A 56 year old woman, presented with one and a half year history of gradual reduction of her ability to taste, culminating in total ageusia within a year. She also observed that for last year she couldn't detect any flavor or aroma with eructation. She underwent an extensive evaluation.The patient underwent a 1 month course of 5.6 mg of L-methylfolate, 2 mg of methylcobalamine and 600 mg of N-acetylcystein. Within

10 days of beginning the regime, she noted abrupt change in her chemosensory perception of her eructations. Now, her eructations tasted of the food that she had recently eaten. Despite the "flavorful eructations", the food itself in the mouth remained flavorless. Within a week of the "flavorful eructations", her intraoral ability to taste food returned to what she perceived to be 60% of her normal. Recrudescence of flavor in eructation may be a sentinel marker in imminent recovery of perceived gustation. Eructation induced retronasal chemosensory perception appears to be more sensitive to aroma than mastication initiated retronasal olfaction. This case further supports the paradigm that complaints of taste in general may in fact be caused by selective aberration in the retronasal olfactory pathway rather than gustation in general. In this patient, the perceived improvement in her taste may be secondary to recovery in her olfactory perception. This case illustrates the importance of documenting eructation flavor in clinical evaluation of chemosensory disorders.

#P242 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Pseudohyposmic Hyperosmia

Syed. A Hassan, Gurprit.S Bains, Sunith Vijayakumar, Alan.R Hirsch
The Smell and Taste Treatment and Research Foundation Chicago, IL, USA

OBJECTIVE While hyposmia, hyperosmia and pseudohyperosmic normosmia has been delineated, subjective hyposmia with true hyperosmia has not been reported. CASE STUDY A 23 y/o single male presented with 16 months of chemosensory complaints following an episode of depression and substance abuse (marijuana and cocaine). There was gradual olfactory loss to 10% of normal, unresponsive to systemic and nasal steroids, fexofenadine and montelukast. Concomitantly their was gradual taste loss to 10% of normal. He noted flavorful eructation but no dysosmia, phantosmia or olfactory windows. Metallic -mothball phantgeusia was also noted. Food was bland, unfulfilling and without complexity. There was loss of appetite and a 45 pound fluctuation of weight. He is depressed and feels cognitively and socially impaired from his loss. Evaluation included normal ENT exam, fiberoptic endoscopy and head CT. Psychiatric assessment noted rapid, pressured speech, circumstantial, hypomanic, anxious and depersonalization. Neurologic exam: saccidization of horizontal eye movement and generalized hyperreflexia. Olfactory testing: dirhinus Quick Smell Identification Test 3/3 and a phenyl ethyl alcohol Smell Threshold Test: left nostril -4.5: right nostril -7.0, consistent with hyperosmia. DISCUSSION The etiology of the above is unclear. His baseline may have been a high hyperosmic state and fell to a lower (but still hyperosmic) range. He may have manifested chemosensory focused somatic complaints has a primary psychiatry disorder. Drugs may have induced distortions, which were perceived as hyposmia. This case highlights the importance of chemosensory testing. Creation of additional test to delineate hyperosmia is worthy of pursuit. Acknowledgements: None

#P243 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Coping with isolated congenital anosmia

Thomas Hummel[1], Simona Negoias[1], Lenka Novakova[1], Basile N. Landis[1,2], Ilona Croy[1,3]
[1]Univ of Dresden Med School, Dept of ORL, Smell & Taste Clinic Dresden, Germany, [2]Univ of Bern, Dept of ORL Bern, Switzerland, [3]Univ of Dresden Med School, Dept of Psychosomatic Medicine Dresden, Germany

Isolated congenital anosmia (ICA) is characterized by the lack of the sense of smell since birth in otherwise healthy people. Although this phenomenon is known among clinicians, there is only little knowledge about how these people cope with this serious handicap. Questionnaires of 32 patients with ICA (aged 18-46 years) were analyzed. ICA was diagnosed using detailed medical history, psychophysical examination, electrophysical measurements, and magnetic resonance imaging. Forty healthy participants (aged 18-57 years) served as controls. The smell disorder was noted first at the age of 11 years; mean age at the medical diagnosis was 20 years. About one-third of the patients avoid talking about the disorder. If they could, many of them would like to smell food (35%), parfume (23%), their spouse (16%), or "nature" (12%). Both groups did not differ significantly in weight, height, Body Mass Index, or eating behavior. However, almost all of the controls named preferred food with only one component, while ICA-patients significantly more often named preferred food with more than one component. ICA- patients reported more household accidents than healthy controls. ICA-patients also reported more worries about social situations than controls. There was no significant difference between both groups in the partnership status or satisfaction with their partnership. However, ICA-patients reported to have had significantly less sexual partners than controls. Finally, ICA-patients exhibited higher scores in the Depression Inventory compared to controls. Overall differences between the two groups are relatively subtle. ICA seems to be a handicap patients can cope with very well. Acknowledgements: We would like to thank Monika Roesner and Selda Olgun for their contribution to the collection of the data. BNL was supported by a Grant of the Swiss National Fund for Scientific Research (SSMBS grant n° PASMA-119579/1).

#P244 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Vulnerability of the Trigeminal System to Environmental Exposures

Christopher Maute, Tamika Wilson, Justine Durmala, Cristina Jaen, Pamela Dalton
Monell Chemical Senses Center Philadelphia, PA, USA

The potential for exposure to chemical vapors or particulates to impair olfactory function has long been acknowledged. In contrast, exposure-induced changes in upper airway somatosensation, mediated through activation of free nerve endings of the trigeminal nerve in the nasal mucosa, have been less well documented. Importantly, however, the compromised function of this sensory system may represent a more significant

deficit than that of olfactory loss, as the trigeminal system is responsible for our ability to detect irritants and initiate reflexes that reduce exposures to the lower airways. Across multiple studies, we have found that individuals exposed to volatile irritants such as formaldehyde or mixed exposures to vapors and particles, show significant loss of sensitivity in upper airway irritation. Anatomy students exposed to formaldehyde vapor showed significant decrements in irritant sensitivity ($p < .01$) following as little as 4 weeks of exposure to concentrations equal to or greater than 0.05 ppm. In populations with longer-term exposures to complex mixtures (i.e. firefighters) deficits in trigeminal sensitivity appeared years earlier than did olfactory losses. Although these decrements can render the workplace environment more tolerable, they also reduce the protective nature of the irritant reflexes and render the upper and lower airways vulnerable to chronic damage. Acknowledgements: Supported by NIH-NIDCD P50 –DC 006760

#P245 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Lateralized olfactory differences- an early indicator for future global olfactory dysfunction

Volker Gudziol, Irene Paech
Smell and Taste Clinic, Dresden Medical School Dresden, Germany

About 15% of subjectively normosmic individuals demonstrate clinical significant unilateral smell deficit. That smell loss will not be noticed as long as olfactory function of the better nostril remains in the normal range. We questioned whether individuals demonstrating side differences of olfactory function are at risk to develop bilateral olfactory loss. Therefore those individuals ("difference- group", n=35) were re-tested on average 4.6 years after baseline investigations. Additionally, 58 subjects who did not demonstrate olfactory side differences ("control- group") were also re-investigated. All participants performed detailed olfactory testing using the "Sniffin' Sticks" involving tests for odor threshold, odor discrimination, and odor identification.
The "difference-group" and the "control-group" were not significantly different regarding age ($p=0.19$), follow up period ($p=0.41$) and olfactory function of the better nostril at baseline ($p=0.35$). Olfactory testing at follow- up indicated lower olfactory function ($p=0.005$) in the "difference- group" than in the "control- group". The degree of side difference at baseline correlated negatively with the results from olfactory testing at follow-up ($r=-0.29$; $p=0.01$). These results suggest that individuals with side differences of olfactory function are at risk to develop bilateral olfactory loss within 4.5 years. Thus, the degree of unilateral smell loss is an indicator for the intensity of future bilateral olfactory loss.

#P246 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Effects of Soccer Ball Heading on Scent Identification and Olfactory Functioning

Bryan Raudenbush, August Capiola
Wheeling Jesuit University Wheeling, WV, USA

Past research shows that head injury decreases both the ability to identify scents and olfactory functioning. The present study examined the effects of soccer ball heading frequency, soccer ball heading intensity, past concussions, and dizziness post-soccer ball heading had on scent identification from pre- to post-season. Men and women soccer teams were asked to identify scents from scratch and sniff booklets (Brief Smell Identification Test and Provista Memory Test) before and after their soccer seasons. Athletes were chosen for further study if they showed a decrease in scent identification performance from pre- to post-season. The correlations for degree of scent identification loss and various actions are as follows: $r = .75$ for heading frequency, $r = .47$ for heading intensity, $r = .80$ for concussion, and $r = .58$ for dizziness. The data were subjected to a multiple regression analysis and it was found that the variables of heading frequency, heading intensity, concussion, and dizziness accounted for 75% of the decreased ability to identify scent and olfactory functioning. Thus, a significant proportion of variance in decreased scent identification ability and olfactory function in these athletes is accounted for by behaviors related to soccer ball heading. Future research may help to explain which particular head area injuries and related actions would effect scent identification and olfactory functioning the most in an attempt to uncover solutions to avoiding these particular circumstances. Acknowledgements: This research was funded by a grant from the NASA WV Space Grant Consortium.

#P247 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Posttraumatic olfactory disorders - a long term follow-up

Antje Welge-Lüssen[1], Andrea Hilgenfeld[1], Thomas Meusel[2,3], Thomas Hummel[3]
[1]Dept. of Otorhinolaryngology, University Hospital Basel Basel, Switzerland, [2]Dept. of Otorhinolaryngology, University Hospital Basel Basel, Switzerland, [3]Dept. of Otorhinolaryngology, Head and Neck Surgery, University Hospital Erlangen Erlangen, Germany, [4]Smell & Taste Clinic, Dept. of ORL, Technical University of Dresden Medical School Dresden, Germany

Objectives: Olfactory disorders following trauma are common and can be present not only after severe trauma but also in cases of light head trauma. In contrast to postinfectious disorders recovery is considered to be less common. However, long-term follow-up data in larger cohorts are rare. It was the aim of our study to perform a long-term follow-up examination in these patients trying to identify prognostic factors. **Methods:** Olfactory function in sixty-seven (38 men, 29 women, mean age: 40 years, range: 17-66) was psychophysically evaluated using the Sniffin Sticks test battery based on the TDI score (composite score of threshold, discrimination and identification). Olfactory testing

was performed separately for the right and left nostril. Moreover, subjective impairment was rated on a visual analogue scale (ranging from 0 ["no impaiment at all"] until 10 ["maximum impairment"]). Additionally, olfactory function was rated on a visual analogue scale (ranging from 0 ["no olfactory function"] umtil 10 ["very good olfactory function"]). **Results:** First examination was performed on average 16 months after trauma, the second examination 74 months after trauma. According to the better side initially 37 patients were anosmic, 27 hyposmic and 3 normosmic. Mean TDI score of the better side increased significantly from 16.7 to 19.4 ($p<0.001$). At the second examination 25 patients were anosmic, 35 hyposmic and 7 normosmic. Subjective impairment decreased significantly from 6.6 to 4.7 ($p<0.001$). Olfactory function was rated higher on the second examination (1.4 vs. 2.8, $p<0.001$). Neither age, sex or side difference between both nostrils on initial testing correlated with improvement. **Conclusion:** Overall, almost 30% of all patients with posttraumatic disorders improved within the follow-up period of 74 months.

#P248 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Relationship between the olfactory abilities and the structure of the olfactory bulb and sulcus in human

Jane Plailly[1], Djaber Bellil[1,2], Aïcha Ltaief-Boudrigua[3], Frédéric Faure[2]
[1]Centre de Recherche en Neurosciences de Lyon, INSERM U1028, CNRS UMR 5292, Université Lyon 1 Lyon, France, [2]Service d'oto-rhino-laryngologie, hôpital Edouard Herriot Lyon, France, [3]Service de radiologie, hôpital Edouard Herriot Lyon, France

The olfactory bulb (OB) is the first central nervous system relay for olfactory input. Until recently, it was technically impossible to determine its structural characteristics in leaving human. Recent advances of imaging techniques have allowed such an approach. However, OB structural data are still scarce and many questions are to be answered. Among them is the link between the function and the structure of this brain region. To address this question, we compared accurate structural data obtained in 3 Tesla MRI (volume of the OB and length and depth of the olfactory sulci, OS) of three groups: healthy normosmic subjects (NORM) and patients presenting a partial loss (hyposmic, HYPO) or a complete loss (anosmic, ANOS) of olfactory abilities. In order to validate the clinical diagnostics of the participants, we tested their detection threshold for phenylethyl alcohol, and their ability to detect and identify suprathreshold odors. Preliminary results revealed a strong difference between the olfactory abilities of the three groups of participants. NORM (n = 11) had lower detection threshold and higher abilities to detect and identify odors than HYPO (n = 9; all $P<0.001$). Same pattern of behavioral differences was observed between HYPO and ANOS (n = 17, all $P<0.01$). Interestingly, and following behavioral evaluations, OB was bigger in NORM than in HYPO ($P<0.01$) and bigger in HYPO than in ANOS ($P<0.05$). Additionally, OS was longer and deeper in NORM and HYPO than in ANOS (all $P<0.05$). Our findings suggest a strong relationship between olfactory abilities and structure of OB and OS. These results highlight the interest of the measurements of such structural data in the diagnostic of olfactory dysfunction in dysosmic patients.

#P249 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

The Implication of Respiratory Patterns on Olfactory BOLD Signal

Jianli Wang[1], Xiaoyu Sun [1], Christopher W. Weitekamp[1], Prasanna Karunanayaka[1], Qing X. Yang[1,2]
[1]Penn State Hershey/Radiology Hershey, PA, USA,
[2]Penn State Hershey/Neurosurgery Hershey, PA, USA

The subject's respiratory pattern can modulate the experimental paradigm in terms of timing and odor perception (intensity and threshold), which presents a critical issue for fMRI post-processing and data interpretation. Here we report an event-related olfactory fMRI technique and related data post-processing methods that can effectively remove the effect of unwanted respiratory pattern effect on the olfactory blood oxygen level dependent (BOLD) signal. The experimental setup includes an olfcatometer that can monitor and record the respiratory information and synchronize the odorant delivery with the subject's respiration and fMRI image acquisition. We compared the olfactory BOLD signals from four healthy normal adults with different respiratory patterns. Our data showed that the subject's respiratory modulation of the olfactory stimulation paradigm significantly confounded the BOLD signal. Thus, it is critical to incorporate respiratory information into both paradigm design and data processing in order to produce reliable olfactory fMRI data. The presented event-related paradigm design and corresponding data processing method are simple and effective for generating olfactory fMRI results with minimal confounding variability. These methods can also be used in studying the implication of respiratory patterns (e.g., sniffing and breath-holding) on olfactory BOLD signal. Acknowledgements: The Leader Family Foundation Laboratory for Alzheimer's disease Research and NIH RO1 EB00454

#P250 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Delay Phenomenon in Olfactory Brain Habituation to Odor Stimuli: An fMRI Study

Yongxiang Wei[1], Wei Xiao[1], Hua Gu[2], Kunyan Li[1], Jinfeng Zhang[3], lifang Si[2]
[1]The Department of Otolaryngology-Head & Neck Surgery, Beijing Chaoyang Hospital, Capital Medical University Beijing, China, [2]The Department of Radiology, Beijing Chaoyang Hospital, Capital Medical University Beijing, China, [3]The Department of Otolaryngology-Head & Neck Surgery, Beijing Tongren Hospital, Capital Medical University Beijing, China

Objective To observe the brain activation before and after recovery of olfactory perception from adaptation using functional magnetic resonance imaging (fMRI), and discuss the mechanisms of olfactory adaptation. **Method** 10 right-handed, normosmic subjects underwent 2 the same olfactory stimulation tasks with the interval of 20 minutes. The odorant used was isovaleric acid. The fMRI data was processed by the SPM5 software. Rating odor intensity and valence using visual analogue scale (VAS), and the results of 2 tasks were statistically analyzed. **Result** There was no

significant difference between 2 tasks on both intensity and hedonicity scores. In task 1, the brain activation in bilateral cerebellum, frontal (including orbitofrontal gyrus), insula, thalamus, cingulate gyrus, Putamen, amygdala / piriform cortex, the left inferior parietal lobule, precentral gyrus, right hippocampus, pallidum, middle temporal gyrus, supramarginal gyrus. In task 2, only the right middle frontal gyrus activated, and the voxels decreased significantly. Paired t-test results show that: (task1-task2) activated regions in left precentral gyrus, frontal lobe (including the orbitofrontal gyrus), insula, right superior temporal gyrus, cerebellum; (task2-task1) activation in the left inferior parietal lobule and right lingual gyrus. **Conclusion** There is obvious 'delay phenomenon' in habituation of brain cortex compared with subjective olfactory sensation. Piriform cortex plays an important role on olfactory adaptation. Underwent repeated olfactory stimulation, second olfactory cortex plays less role on olfactory perception and advanced processing. Acknowledgements: China Natural Scientific Foundation under project No. 30973284, Beijing Natural Scientific Foundation under project No. 7102063, Capital Medical Development Foundation under project No. 2007-1034.

#P251 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Independent Component Analysis on Habituation Effects of the Olfactory System on fMRI: A Non-Linear BOLD Respond

P Karunanayaka[1], CW Weitekamp[1], J Wang[1], QX Yang[1]
[1]Penn State College of Med./Radiology Hershey, PA, USA

Introduction: The goal of this research is to characterize the habituation effect on BOLD signals in the central olfactory system. We investigated the dynamics of BOLD response pattern during a four strength olfactory stimulation paradigm using group independent component analysis (ICA). This information is essential for olfactory fMRI data acquisition, analysis and interpretations. **Methods:** *Human Subjects:* Ten healthy subjects (mean age 24.7 + 1.8 years) completed two identical runs of a four strength olfactory fMRI paradigm at 3.0T. *Odor Stimulus Paradigm:* Four concentrations (weak [0.032%], medium [0.10%], strong [0.32%], and very strong [1.0%]) of lavender odorant (Quest International Fragrance Co.) were sequentially presented to subject's nostrils (6s/stimulation) three times with a 30s fresh air period in between. Visual cues of "Rest" and "Smell?" were also presented to the subjects. Conclusion: Group ICA method revealed several neuronal circuits involving detection, sniffing and cognitive activities with each having unique temporal characteristics. Time courses exhibit a task-related behavior, even for the condition where the cue "Smell?" was presented when no odorant was delivered. Based on individual ICA time courses, possible linear responses in the BOLD signal with the odorant concentration was investigated using a regression model. Our analyses indicated that the independent components of the BOLD time courses exhibited a complicated **non**-linear behavior that is likely associated with habituation effect. These new findings demonstrated that ICA is effective analysis tool for olfactory fMRI investigations. Acknowledgements: The Leader Family Foundation Laboratory for Alzheimer's Disease Research and NIH R01 AG 027771

#P252 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Reward-related Representations of Natural Food Odors in the Human Brain

James D. Howard, Jay A. Gottfried
Department of Neurology, Northwestern University Feinberg School of Medicine Chicago, IL, USA

The smell of food serves as a powerful cue to guide feeding behavior. After a food is eaten to satiety, the rewarding properties of the smell of the consumed food are reduced, often prompting organisms to turn their attention toward non-sated items that may promise other nutritive benefits. Despite the importance of smells for initiating and terminating feeding, few studies have examined how hunger and satiety states modify food-based odor information in the human brain. This may be attributable to the relative complexity of food odors, which are often mixtures of dozens of molecular components. As such, the role of food odor components in feeding behavior, and their corresponding representations in the human brain, is unknown. We have used a gas chromatography /mass spectrometry (GC/MS) system to identify and quantify 16 distinct molecular components of peanut butter. Currently, high-purity synthetic versions of these components are being combined in mixture dilution-series to establish their relative concentrations within the natural food odor. These components, along with natural peanut butter odor and an unrelated control food odor, will then be intermittently presented to human subjects while they undergo fMRI scanning both before and after eating peanut butter to satiety. We expect to observe a selective decrease in brain activity from pre- to post-satiety in the amygdala and orbitofrontal cortex elicited by peanut butter odor but not the control odor. The critical question will be to determine how satiety-related brain activity changes in response to smelling the components of peanut butter odor. The extent to which these changes coincide with either the perceptual or physical makeup of these components could shed light on the neurobiological processes underlying feeding behavior in humans. Acknowledgements: Supported by NIH grants 1R01DC010014-01 and 3R01DC010014-01S1 (American Recovery and Reinvestment Act of 2009 ARRA), and a Seed Grant from the Brain Research Foundation.

#P253 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

An fMRI Investigation of Negative Olfactory Alliesthesia: Enhanced perception of subthreshold negative odors in anxiety

Elizabeth A Krusemark, Wen Li
University of Wisconsin Madison, WI, USA

Research on threat processing has recognized a variety of biases associated with anxiety, particularly evident among attention and other downstream cognitive processes. However, little work has addressed the influence of anxiety during early stages of sensory-perceptual processing. Odors often elicit intense emotional reactions, given the intimate neuroanatomical ties between olfaction and emotion systems. Olfactory perception can involve alliesthesia as hedonic odor evaluation varies with the internal

state of an individual such that the negative internal state of anxiety may negatively bias evaluation of one's olfactory environment. Combining functional magnetic resonance imaging (fMRI) with an odor discrimination triangular task using neutral odorants and their mixtures with highly diluted negative odorants, the current study (N=14) examined the influence of anxiety on olfactory perception of threat-related smells. We predicted anxiety would increase aversion to odors (negative olfactory alliesthesia) and facilitate discrimination of subtle negative odors from neutral odors. Moreover, we predicted response enhancement to negative odors in olfactory cortices in anxious individuals, further isolating the neural mechanisms of this threat-related bias to the sensory system. Preliminary analyses indicated anxiety ratings were associated with discrimination accuracy between pure odors and their negative mixtures (relative to discrimination between pure odors and their neutral mixtures; $p<.05$). Analyses of neuroimaging data are ongoing; however, these data suggest a role for anxiety in sharpening olfaction, transforming fleeting, unnoticeable malodors into sensory violations among anxious individuals.
Acknowledgements: Parts of this research were funded by a Training grant from the National Institute of Health T32-MH018931 to E.A.K.

#P254 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Differential effects of satiety on brain response to ortho- vs. retronasally perceived food and nonfood odors

Maartje S. Spetter[1,2,3], Genevieve Bender[2,3], Thomas Hummel[4], Simona Negoias[4], Maria G. Veldhuizen[2,3], Dana M. Small[2,3]
[1]Image Sciences Institute, University Medical Center Utrecht Utrecht, Netherlands, [2]John B. Pierce Laboratory New Haven, CT, USA, [3]Department of Psychiatry, Interdepartmental Neuroscience, Yale Univerisity School of Medicine New Haven, CT, USA, [4]University of Dresden Medical School Dresden, Germany

We used fMRI to investigate the influence of route of odorant delivery (orthonasal vs. retronasal) on the effect of internal state (hungry vs. full) on neural response to two odor categories (food = F and nonfood (NF) odors). 16 subjects were scanned four times, twice hungry and twice after consuming a food associated with a target F odor to satiety. All odors were delivered as vapors through tubes inserted under endoscopic guidance so that one tube ended shortly beyond the nasal valve (ortho-) and the other at the nasopharynx (retro-). F odors included chocolate, pineapple, peach, and tomato aromas. NF odors included rose and lilac. F odors evoked greater response in the hypothalamus and cerebellum, irrespective of route of administration and internal state. Response in the hippocampus and piriform cortex was greater to F vs. NF odors when hungry compared to full, irrespective of route. Three way interactions between route, odor category, and internal state were observed in the insula, anterior cingulate cortex and amygdala. In all three areas response was greater to F vs. NF odors when hungry compared to when full. In keeping with prior work (Small et al., 2005), in the insula and amygdala this was greater for ortho- vs. retronasal stimulation, whereas in the anterior cingulate cortex the effect was greater for retro- vs. orthonasal stimulation. In addition, consistent with prior work (Gottfried et al., 2003) we found that when full, but not when hungry, the nontarget F odor elicited significantly greater response in the amygdala than the target F odor during ortho- but not retronasal stimulation. The findings extend knowledge by showing that internal state impacts the influence of route on brain response and that route influences the sensitivity of the amygdala to reward devaluation by satiety.

#P255 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Olfactory-visual synthesis sharpens subthreshold threat perception: An fMRI investigation

Lucas R. Novak, Wen Li
University of Wisconsin - Madison Madison, WI, USA

Evolution has frequently favored the development of multiple senses, and all species equipped with multiple senses integrate them to optimize perception and action (Stein and Meredith, 1993). This crossmodal synthesis is particularly prominent when minimal sensory information is available (i.e., the principle of inverse effectiveness), thereby serving a crucial function in resolving perceptual uncertainty and ambiguity. We surmise that this synergy would be especially pronounced for subtle threat-related signals, given their ecological importance. In this functional magnetic resonance imaging (fMRI) study, fourteen participants performed a bimodal discrimination task where they were asked to simultaneously smell an odor (either a pure neutral odor or its mixture with a highly diluted negative odor) and view a picture of a human face (either a neutral expression or a mixed expression of a neutral and a very faint fearful expression). Participants then pressed buttons to report whether each stimulus was neutral or negative. As we predicted, accuracy for both face and odor judgments improved when the bimodal cues were affectively congruent (p's <.1). Additionally, sharpening of odor perception via olfactory-visual synthesis was further elevated when participants displayed heightened levels of disgust sensitivity and obsessive thoughts of contamination (p's <.05).

#P256 **POSTER SESSION V: OLFACTORY MODULATION; OLFACTORY DEVELOPMENT; CHEMOSENSATION AND DISEASE; FUNCTIONAL IMAGING**

Riech-O-Mat: A small and simple olfactometer for fMRI studies

J. Ulrich Sommer[1], Clemens Heiser[1], Martin Griebe[1], Boris A. Stuck[1], Thomas Hummel[2]
[1]University-ENT-Clinic Mannheim, Germany, [2]Department of Otorhinolaryngology - Smell and Taste Clinic - University of Dresden Medical School Dresden, Germany

To perform fMRI studies with olfactory stimulation the device has to be of a non-conductive and non-magnetic material and the unit should be highly transportable. At this point of time, only a limited number of devices fulfill these criteria and are associated with high costs. Aim of the study was to investigate whether a newly developed, relatively simple and inexpensive stimulation device is suitable for fMRI measurements. Our stimulation device

was made of standard industrial and laboratory components and consists of 3 sections. 1 : The air inlet, control and distribution section. 2: The odorant-section and 3: The delivery-section. Air is taken from a regular clean air wall outlet and a constant flow is achieved using a ball-flow-meter. Computer controlled electro-pneumatic valves direct the airflow to 4 gas-washing bottles where the liquid odorants and the control fluids are filled in. 4 silicone hoses are connected with t-fittings at the end just before entering the subject's nose. For the actual fMRI study 5 healthy normosmic subjects were stimulated with Phenylethylalcohol (PEA) at a flow rate of 5 l/min. Distilled Water was used a control fluid. Blood-oxygen level dependent (BOLD) fMRI was performed on a 1.5T scanner (Siemens Sonata) in a block design. The statistical evaluation was performed by the MATLAB based SPM8 software package. Statistical threshold was $p<0.01$ (uncorrected). fMRI data were superimposed on a T1 anatomical reference. Analysis of fMRI data showed bilateral activations within the insula, the orbitofrontal Cortex and ipsilateral activations in the mesiotemporal area. The experiment proved that our easy-to-use and cost-effective stimulation device is able to create a reproducible and adequate stimulation with fluid odorants, measureable with fMRI sequences.

#P257

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Messenger-RNA Deep Sequencing Analysis of the Human and Mouse Olfactory Transduction Subgenome

Ifat Keydar[1], Tsviya Olender[1], Miriam Khen[1], Edna Ben-Asher[1], Ester Feldmesser[1], Arisa Oshimoto[2], Diego Restrepo[2], Yoav Gilad[3], Doron Lancet [1]
[1]The Weizmann Institute of Science Rehovot, Israel, [2]Cell and Developmental Biology, University of Colorado Denver, CO, USA, [3]Department of Human Genetics, University of Chicago Chicago, IL, USA

While the human olfactory receptor (OR) subgenome and its genomic variability are well-studied, less is known on olfactory-related non-receptor genes. We focus on two gene categories, collectively termed accessory olfactory genes: genes mediating OR signal transduction and genes involved in olfactory sensory neuron (OSN) development and integrity. We perform association studies between genetic variations in such genes and across-odorant sensitivity disparities. An extreme case is congenital general anosmia (CGA), for which we are conducting whole exome DNA sequencing of 6 samples, to discover relevant mutations in olfactory accessory genes likely to underlie the non-odorant-specific phenotype. In parallel, we study the weaker 'general olfactory factor' phenotype, previously reported by Cain & Gent (1991) and confirmed by us in specific anosmia cohorts (see Wysocki et al., AChemS 2010 #11). We reckon that such average olfactory sensitivity may, at least in part, be explained by genetic polymorphisms in accessory olfactory genes. For producing a candidate accessory gene list we scrutinized the literature of olfactory transduction and OSN development, including mouse gene knockouts. In parallel, we have conducted next-generation RNA sequencing of 5 autopsy and biopsy specimens from normosmic individuals, in comparison to transcriptomes from 8 standard tissues. For further comparison, mouse olfactory epithelial samples are similarly scrutinized. We have identified ~150 human olfactory accessory gene candidates, with evidence for olfactory specific RNA splice variants, and with data on significant inter-individual genome differences. The emerging map of the accessory olfactory subgenome may assist in rationalizing the great inter-individual variation in human general olfactory sensitivity.
Acknowledgements: Crown Human Genome Center at Weizmann Institute

#P258

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

PDZ-Proteins – Functional units of the olfactory signal transduction cascade

Sabrina Baumgart[1], Ruth C. Dooley[2], Fabian Jansen[1], Benjamin Fraenzel[1], Dirk Wolters[1], Hanns Hatt[1], Eva M. Neuhaus[3]
[1]Ruhr-University/Department of Cell Physiology Bochum, Germany, [2]Beaumont Hospital/Molecular Medicine Dublin, Ireland, [3]Chartite /Neuroscience Research Center Berlin, Germany

The olfactory signal transduction cascade is a complex machinery of various proteins which enable organisms to detect and discriminate between a myriad of different odorants. Most of the essential components of this pathway are already known but the question arises how those get orchestrated. To address this question we performed peptide microarray studies. With this we could show that a PDZ-Protein, called MUPP1, can interact with a great variety of different olfactory receptors. With its 13 individual PDZ domains MUPP1 has a great ability to form complex and dynamic networks. This is already known for different G-Protein coupled pathways. In mass spectroscopy interaction assays with single in *E.coli* produced PDZ domains and lysate of olfactory epithelium of mice we could show that different components of the olfactory signal transduction cascade like G_{olf} can interact with MUPP1. To further elucidate the function of this scaffolding protein we designed an inhibitory peptide which contains the last 15 C-terminal amino acids of the mOR-EG. Herewith we want to interrupt the interaction between MUPP1 and the olfactory receptor in transgenic mOR-EG mice by introducing the synthetic peptide via a patch clamp pipette. Afterwards we will measure the odorant responses to the specific ligand vanillin of single olfactory sensory neurons in slices in comparison to untreated neurons. Acknowledgements: Studienstiftung des deutschen Volkes, the International Max-Planck Research School for Chemical Biology, the Ruhr University Research School and the Deutsche Forschungsgemeinschaft (SFB642).

#P259 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Phosphorylation of Adenylyl Cyclase III at Serine-1076 does not contribute to olfactory adaptation in mice

Katherine D. Cygnar, Sarah H. Sarah Collins, Chantal Bodkin-Clarke, Haiqing Zhao
The Johns Hopkins University/Department of Biology Baltimore, MD, USA

In the nose, odorants are detected on the cilia of olfactory sensory neurons (OSNs), where a cAMP-mediated signaling pathway transforms odor stimulation into electrical responses. OSNs continuously modulate response sensitivity based on previous odorant stimulation, a process termed adaptation. Adaptation is likely mediated by a number of Ca^{2+}-dependent feedback mechanisms that may be differentially recruited depending on the intensity and duration of the adapting stimulus. However, which targets of Ca^{2+}-feedback underlie olfactory adaptation remains poorly understood. Feedback inhibition of adenylyl cyclase III (ACIII) via phosphorylation by CaM Kinase II (CaMKII) has been hypothesized to be a mechanism of olfactory adaptation, particularly adaptation that is induced by long odor pulses. This hypothesis was derived from experiments using either CaMKII inhibitors or a heterologously-expressed mutant form of ACIII, and the phosphorylation on a single serine residue, ser-1076, was identified to account for ACIII inhibition. To directly determine the consequences of this feedback mechanism for olfaction *in vivo*, we genetically mutated the ser-1076 of ACIII to alanine in mice. Immunohistochemistry and Western blot analysis show that this mutation does not affect the cilial localization and the expression level of ACIII in OSNs. Electroolfactograms showed no differences in the responses between wildtype and mutant mice to single stimulations or in several adaptation paradigms. The mutant mice also show a similar slowing of the activation kinetics in responses following a prolonged stimulation, a phenomena that is dependent on CaMKII activity and had been suggested to be due to inhibition of ACIII. These results suggest that phosphorylation of ACIII on Ser-1076 does not contribute to olfactory adaptation. Acknowledgements: Supported by NIH DC009946 and DC007395.

#P260 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Differential Expression Patterns of G-protein βγ subunits in the Mouse Peripheral Olfactory System

Aaron Sathyanesan, Saloni Mehta, Chiamaka Nnah, Weihong Lin
Dept. of Biological Sciences, University of Maryland Baltimore County Baltimore, MD, USA

Olfactory signal transduction involves a G-protein-coupled signaling cascade in which the α subunit of the G-protein, $G\alpha_{olf}$ plays a critical role. Other Gα subunits, such as $G\alpha_s$ are known to play roles in axonal sorting and targeting. Whereas much is established about the distribution, identity and function of the different Gα subunits in the olfactory epithelium, little to nothing is known about their βγ components. Previously, we reported the presence of multiple Gβγ subunits in the mouse olfactory epithelium through RT-PCR and RNA *in situ* hybridization (Sathyanesan *et al.,* 2010, AChemS abstract). To further characterize the spatial and temporal distribution of the Gβγ subunits, we performed RNA *in situ* hybridization against different subunits at four developmental time-points – postnatal day 0, 7, 14 and adult. In adult mice, $G\beta_1$ and $G\gamma_{13}$ mRNA were found in mature neurons of the MOE. In the VNO, labeling for $G\beta_1$ mRNA was observed in both upper and lower layer of vomeronasal sensory neurons, whereas $G\gamma_{13}$ expression was restricted to neurons in the upper layer (presumably these are $G\alpha_{i2}$-expressing neurons). Further, in the MOE, $G\gamma_2$ is expressed in the globose basal cell layer in P7 and P14 mice, however in adult, it is expressed in the supporting cells. Our results suggest a differential distribution of Gβγ subunits expression during post-natal development of the MOE. Our results also indicate differences in Gβγ expression patterns among the upper and lower populations of neurons in the VNO. Acknowledgements: Supported by NIH/NIDCD 009269 and ARRA administrative supplement to WL.

#P261 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Signal Transduction Pathways of IP3R3-containing Microvillous Cells

Tania R Iqbal[1], Colleen C Hegg[2]
[1]Michigan State University, Neuroscience Program East Lansing, MI, USA, [2]Michigan State University, Department of Pharmacology and Toxicology East Lansing, MI, USA

The function of olfactory epithelium microvillous cells is unclear. Recently, we reported that a subset of microvillous cells identified by the presence of inositol triphosphate receptor 3 (IP3R3) responded to odorants and adenosine triphosphate (ATP) with increases in intracellular calcium. The objective of this study is to determine signal transduction mechanisms of ATP and odorants in IP3R3-containing microvillous cells (IP3R3-mv) using confocal calcium imaging of olfactory epithelial slices obtained from neonatal $IP3R3^{tm1(tauGFP)}$ mice. To determine if odorant-induced calcium increases are G-protein mediated, GDPβS was used to block G-protein transduction. In 75% of IP3R3-mv cells (16 of 20 cells, 3 slices), GDPβs inhibited the odorant mediated increase in calcium by 43 ± 0%. Co-application of forskolin and 3-isobutyl-1-methylxanthine, agents that increase cAMP levels, evoked calcium transients, but they were smaller than odorant evoked-calcium transients (n=22 cells, 2 slices). These data suggest odorant-induced calcium increases may be mediated by a G-protein/Adenylyl cyclase pathway. Although we previously showed the presence of ionotropic P2X3 on IP3R3-mv cells, GDPβs decreased ATP-evoked calcium transient by 48 ± 0% (n=19 cells, 3 slices), suggesting the presence of G-protein coupled P2Y receptors. Indeed, the P2Y agonist UTP induced robust calcium transients (n=5 cells, 1 slice). 2-aminoethoxydiphenyl borate, an IP3 receptor inhibitor, decreased Ca^{2+} responses to ATP by 35% (n=14 cells, 5 slices), suggesting ATP induces calcium responses in microvillous cells via a G-protein coupled receptor that activates PLC. Collectively, our data suggest that IP3R3-mv cells act as a secondary chemosensory cell that is directly activated by odorants. Acknowledgements: NIDCD 006897

#P262

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Odorant Responses in Phosphodiesterase 1C Knockout Mice

Michele Dibattista, Johannes Reisert
Monell Chemical Senses Center Philadelphia, PA, USA

Stimulation of mammalian olfactory sensory neurons (OSNs) begins with binding of an odor molecule to odorant receptors (ORs), which in turn, via a G protein-coupled cascade, activates adenylyl cyclase III (ACIII) and increases intracellular cAMP. cAMP opens the cyclic nucleotide-gated channel to initiate the odorant-induced depolarization. Two phosphodiesterases (PDEs), one located in the cilia (PDE1C) and one restricted to the dendrite and cell body (PDE4A) degrade cAMP thereafter (Cygnar & Zhao, 2009). Using the suction pipette technique, we recorded from PDE1C knockout mice (which also expressed GFP in OSNs expressing the I7 OR for identification) to investigate the role of PDE1C in the odor response. We found that the odorant response in I7-positive OSNs (stimulated with the I7 agonist heptanal at 100 µM) is significantly smaller in PDE1C knockout OSNs compared to wildtype (I_{peak}=-106±17 pA in wt and I_{peak}=-70±10 pA in ko, n=12-16, *t-test p<0.05*) and might also have a slower time course. Wildtype I7-postive OSNs also display a large response to the PDE inhibitor IBMX (Reisert, 2010) indicating a high basal ACIII activity in these neurons. We also stimulated PDE1C knockout OSNs with IBMX to investigate if PDE activity remains in PDE1C knockout OSNs. I7-positive OSNs retained very small and slow responses (I_{peak}= -8.47±1.59 pA with a time to peak of 1157±505 ms, n=16) compared to wildtype OSNs (I_{peak}= -47.23±7.17 pA with a time to peak of 62 ±10 ms, n=12), suggesting that PDE4A might play a marginal role in cAMP degradation. In conclusion, our data suggest that PDE1C plays an important role in maintaining odorant-response sensitivity in I7-positive OSNs, but might only contribute to overall response kinetics in a limited way.

#P263

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

PI3K-dependent Antagonism In Mammalian Olfactory Receptor Neurons

Kirill Ukhanov[1], Daniela Brunert[1], Elizabeth A. Corey[2], Barry W. Ache[1,2]
[1]University of Florida, McKnight Brain Institute Gainesville, FL, USA, [2]University of Florida, Whitney Lab St.Augustine, FL, USA

Phosphoinositide (PI) signaling, in particular PI3Kinase (PI3K) signaling, has been implicated in mediating inhibitory odorant input to mammalian olfactory receptor neurons (ORNs). To better understand this phenomenon we investigated PI3K-dependent inhibition between single odorant pairs. The concentration-dependent inhibition of the response of native rat ORNs to octanol by citral is PI3K-dependent; blocking PI3K activity with the β and γ isoform-specific inhibitors AS252424 and TGX221 eliminated or strongly reduced the inhibition. Interestingly, blocking PI3K also changed the apparent agonist strength of the otherwise non-competitive antagonist citral. The excitation evoked by citral after blocking PI3K, could be suppressed by the adenylate cyclase III (ACIII) blockers MDL12330A and SQ22536, indicating that citral could also activate ACIII, presumably through the canonical OR. The G protein Gβγ subunit blockers suramin, gallein and M119 suppressed citral's inhibition of the response to octanol, indicating that the activation of PI3K by citral was G protein dependent, consistent with the idea that inhibition acts through the canonical OR. Lilial similarly antagonized the response to isoamyl acetate in other ORNs, indicating the effect generalizes to at least one other odorant pair. The ability of methyl-isoeugenol, limonene, α-pinene, isovaleric acid and isosafrole to inhibit the response of other ORNs to IBMX/forskolin in a PI3K-dependent manner argues the effect is not mediated by a specific OR. Our findings collectively raise the interesting possibility that the OR serves as a molecular logic gate when mammalian ORNs are activated by natural, complex mixtures containing both excitatory and inhibitory odorants. Acknowledgements: NIDCD DC001655, DC005995

#P264

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Pheromone-dose-dependent and Zeitgebertime-dependent modulation of pheromone responses by DAG in antennal trichoid sensilla of the hawkmoth *Manduca sexta*

Monika Stengl, Petra Gawalek, Andreas Nolte, Christian Flecke
University of Kassel/FB10, Biology, Animal Physiol. Kassel, Germany

Manduca sexta males detect sex pheromones with long trichoid sensilla on their antennae. One of the two olfactory receptor neurons which innervate the pheromone-sensitive sensilla always responds to bombykal (BAL), the main pheromone component. The pheromone-dependent signal transduction cascade in insects is still under lively debate. Thus, in tip recordings of pheromone-sensitive trichoid sensilla of the hawkmoth antenna we examined the role of the diacylglycerol analogues DOG and OAG in BAL-transduction. A non-adapting stimulation protocol was employed (1 µg or 10 µg BAL, duration 50 ms, interstimulus interval 5 min) for 3 hours at Zeitgebertimes (ZT) 1-4, the end of the moth s activity phase and at 8-11, the resting phase. At ZT 1-4 OAG increased and DOG decreased the 1 µg BAL-dependent action potential (AP) response, while both analogues increased the sensillum potential amplitude (SP). At ZT 8-11 with 10 µg BAL stimulation DOG decreased both measured parameters while OAG was ineffective. All effects appeared to depend on the intracellular Ca^{2+} concentration, which possibly changed daytime- and BAL-dose-dependently. Our results are consistent with our previously posted hypothesis of pheromone transduction suggesting that BAL activates a phospholipase Cβ-dependent signal transduction cascade, with IP_3-dependent activation of Ca^{2+}-permeable ion channels. The increase of intracellular Ca^{2+} and DAG is suggested to decrease the sensitivity of the signal transduction cascade via activation of protein kinase C. In contrast, we hypothesize that DAG-dependent rises in pheromone-dependent SP and AP responses were due to DAG-dependent activation of transient receptor potential (TRP)-ion channels which are blocked in the presence of high intracellular Ca^{2+} concentrations during pheromone transduction. Acknowledgements: Supported via DFG grant STE 531/20-1 to MS

#P265 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

On the role of mitochondrial calcium in mouse olfactory sensory neurons

L M Moeller[1], D Fluegge[1], J Spehr[1], S Veitinger[1], S Cainarca[2], S Lohmer[2], S Corazza[2], E M Neuhaus[3], M Spehr[1]
[1]Department of Chemosensation, Institute for Biology II, RWTH-Aachen University Aachen, Germany, [2]Axxam SpA, San Raffaele Scientific Institute, DIBIT Milan, Italy, [3]Charité - Universitätsmedizin, NeuroCure Berlin, Germany

Ionized calcium is key to a variety of sensory signaling pathways. In olfactory sensory neurons (OSNs), odorant receptor - ligand interaction is translated into an electrical signal by opening of cyclic nucleotide-gated (CNG) channels and Ca^{2+} influx into the cilia. This primary event triggers a number of secondary cellular responses that ultimately shape an individual neuron's sensory output. Therefore, cytosolic Ca^{2+} concentrations are tightly controlled. Here, we investigate a functional role of mitochondria in shaping the odor-mediated Ca^{2+} response in OSNs. Using both genetically engineered mice that express a Ca^{2+}-sensitive photoprotein associated to the inner mitochondrial membrane and a new dedicated bioluminescence microscope, we establish a novel imaging approach to selectively record Ca^{2+} signals in OSN mitochondria at high temporal resolution. Electrophysiological recordings from identified OSNs reveal the functional consequences of mitochondrial perturbation on both the odor-mediated primary receptor current and the electrical output signal. We show that mitochondria play a vital role in olfactory Ca^{2+} signaling, controlling both primary and secondary Ca^{2+} pathways. When Ca^{2+} sequestration by mitochondria is pharmacologically inhibited, the distinct time course of the odor-mediated cytosolic Ca^{2+} signal is significantly changed. Moreover, we report activity-dependent mitochondrial translocation to dendritic compartments upon odor stimulation. Based on electrophysiological recordings, we suggest that mitochondrial Ca^{2+} mobilization exerts a regulatory function that shapes an individual neuron's dynamic response range and provides a novel mechanism of olfactory input-output gain control.
Acknowledgements: This research is funded by the Volkswagen Foundation and the Deutsche Forschungsgemeinschaft(MS).

#P266 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Effect of cytoplasmic Ca buffer on the lateral spread of olfactory information in the olfactory cilium

Hiroko Takeuchi, Takashi Kurahashi
Graduate School of Frontier Biosciences, Osaka University Osaka, Japan

It has been shown in the olfactory cilium that the olfactory signal does not spread laterally when the small area was locally stimulated. The short-term olfactory adaptation that is regulated by the Ca-feedback was not observed when two spots (2 µm distance) were succeedingly stimulated. However, such measurements were performed in the presence of the Ca-chelator that is responsible for the reduction of Ca ions. In the present study, we checked the effect of reduced Ca-chelator on the signal spread. Olfactory cells were recorded with the whole-cell version of patch clamp and the cell interior was filled with caged cAMP. When the sub-micron UV spot was applied to a local area by ROI function, an inward current was and fluorescent imaging (Fluo3 or 4) simultaneously. Even in the complete absence of cytoplasmic Ca chelator in the whole-cell pipette, the diffusion of cytoplasmic elements was still observed in the single cilium, while the effective distance was slightly farther (average 4 µm) than in the presence of the Ca-chelator. Considering that the whole-cell recording configuration actually reduces the intrinsic buffering capacity, the results indicate that the olfactory response in the native cilia is a spatially linear-summation of local responses, which is achieved by the functional compartment within the single cilium.

#P267 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Proton-sensitivity of vomeronasal sensory neurons in mice

Annika Cichy, Jennifer Spehr, Marc Spehr
RWTH Aachen, Institute for Biology II, Dept. Chemosensation Aachen, Germany

The mouse olfactory system is organized in at least four subsystems. Among those, the vomeronasal organ (VNO) plays an important role in the detection of pheromones and other social signals. However, key mechanisms underlying signal detection in the VNO remain unknown. Here, we investigate acid-sensing mechanisms in mouse vomeronasal sensory neurons. Using both current-clamp and voltage-clamp whole-cell recordings from optically identified vomeronasal neurons in acute tissue slices, we show that acidic media of different pH values dose-dependently induce inward currents in voltage-clamp recordings. The same stimuli elicit robust action potential firing in current-clamp measurements. The pharmacological profile of the underlying ionic conductances indicates the possible contribution of multiple acid-sensing ion channels. On-going molecular and biochemical studies as well as electrophysiological recordings will provide insight into the functional role of acid-sensing in the mouse vomeronasal organ.

#P268 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Molecular characterization of ionotropic glutamate receptors in lobster olfactory receptor neurons

Elizabeth A Corey[1,2], Yuriy Bobkov[1,2], Barry W Ache[1,2,3]
[1]Whitney Laboratory St Augustine, FL, USA, [2]2Center for Smell and Taste and McKnight Brain Institute Gainesville, FL, USA, [3]Depts. of Biology and Neuroscience, University of Florida Gainesville, FL, USA

The molecular basis of olfactory signal transduction in crustaceans, a major group of arthropods, is still uncertain. Compelling evidence for G protein activation of metabotropic signaling pathways in lobster olfactory transduction needs to be weighed against recent evidence for lobster olfactory-specific ionotropic glutamate receptors. As part of an effort to further

identify and characterize the olfactory receptors of the spiny lobster, *Panulirus argus*, we sampled its olfactory transcriptome using 454 sequencing technology. We have identified multiple orthologs of *Drosophila* olfactory variants of ionotropic glutamate receptors (IRs) in the olfactory transcriptome that are not present in transcriptomes from the brain or stomatogastric ganglion. We have cloned and analyzed full length sequences of multiple lobster IR orthologs as well as partial sequences from other additional potential IRs. Expression of the IRs in olfactory tissue was confirmed by RT-PCR using an ORN cDNA library and/ or *in situ* hybridization. One lobster ortholog can be localized to the transduction compartment (outer dendrites) by western blot and immunocytochemistry. These results support a role for IR-mediated signaling in lobster olfactory transduction. Using heterologous expression, we are currently attempting to determine whether the lobster IR orthologs indeed function as ionotropic olfactory receptors and the extent to which they are capable of driving metabotropic signaling in lobster ORNs. Acknowledgements: Supported by the NIDCD (DC001655, DC005995, and DC009730).

#P269

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

BBS Genes are Key to Channel Function in Cilia

Judith L. Van Houten[1], A. Rajendran[2], Megan S. Valentine[1], S.D. Weeraratne[3], J. Beisson[4,5], Junji Yano[1], Jean Cohen[4,5], France Koll[4,5]
[1]University of Vermont, Biology Burlington, VT, USA, [2]Harvard University School of Medicine, Deaconess and Children's Hospital Boston, MA, USA, [3]Harvard University School of Medicine, Brigham and Women's Hospital Boston, MA, USA, [4]Center for Molecular Genetics, National Center for Scientific Research Gif-sur-Yvette, France, [5]Université Paris-Sud, Orsay, France

Cilia are sensory organelles that mediate mechano-, chemo- and photo-sensory transduction. They are key to olfactory transduction, and have an array of channels and signaling proteins that are not represented elsewhere in the cell. The mechanisms by which signaling molecules are partitioned into the cilia are not yet clear. *Paramecium tetraurelia* with thousands of cilia per cell is attractive for the study of ciliary function and dysfunction, especially neuronal cilia. We have used feeding RNAi to down regulate the expression of orthologs of Bardet Biedl Syndrome (BBS) ciliopathy genes. *P. tetraurelia* expresses 10 genes for BBS proteins, 7 of which are known to form a BBSome in other organisms. RNAi for all *P. tetraurelia* BBS sequences except BBS2, produces behavioral changes that are consistent with the loss of K channels from the cilia, i.e. a "BBS phenotype." We expressed a tagged Small Conductance Calcium –activated K channel (SK1a) and tagged PKD2, both of which are channels found in the *Paramecium* cilia. After down regulation of BBS 7, 8 or 9, these channels were no longer found in the cilia. However, a GPI anchored folate chemoreceptor that is normally found in cilia was unperturbed by these RNAi conditions. The voltage gated calcium channel that is exclusively in the ciliary membrane appears to be unaffected as well, judging by the behavior that is attributable to this channel's activation. We are now confirming this hypothesis with a tagged ciliary voltage gated calcium channel. Immunoprecipitation of BBS9 and BBS8 from whole cell lystates followed by mass spectrometry identified all members of the BBSome described in other organisms. We are currently following the fate of other channels such as a Mg channel in BBS down-regulated cells. Acknowledgements: Sequencing was performed in the VT Cancer Center DNA Analysis Facility and was supported in part by grant P30CA22435 from the NCI. The views expressed are those of the author and do not represent the views of the NCI. Deconvolution microscopy was completed with the help of NIH grants R01GM59988 and P20 RR016435-06.

#P270

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Expression and Channel Properties of Anoctamin 2 Splice Variants

Samsudeen Ponissery Saidu[1], Aaron B. Stephan[2], Haiqing Zhao[2], Johannes Reisert[1]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]Department of Biology, Johns Hopkins University Baltimore, MD, USA

Anoctamin 2 (ANO2) was recently suggested to be the olfactory calcium-activated chloride channel that amplifies the odorant-evoked receptor potential in olfactory receptor neurons. Analysis of *Ano2* transcripts in olfactory mucosa identified a major splice variant that contained Exon 4, which encodes 33 amino acids in the predicted intracellular N-terminal region of ANO2, and a minor splice variant that lacked Exon 4 (Stephan et al, 2009). We further found that two alternative transcription initiation sites, which lead to different N-terminal ends of ANO2, generate greater diversity among *Ano2* splice variants. To understand the functional significance of these splice variants, we expressed them individually in HEK293t cells and investigated their channel properties using patch clamp electrophysiology. The heterologously-expressed Exon 4-containing ANO2 variants displayed biophysical properties that are largely similar to those of the native calcium-activated channel. However, the Exon 4-lacking variants did not generate any recordable currents in response to Ca^{2+} stimulation. Consistent with the electrophysiology, imaging of the GFP-tagged Exon 4-lacking variants showed diffuse fluorescence within the cells, suggesting that the Exon 4-lacking ANO2 protein failed to target to the plasma membrane. Among the Exon 4-containing ANO2 variants, we observed a difference in Ca^{2+} sensitivity between the variants with different N-terminal ends. We propose that the Exon 4 domain is necessary for membrane targeting of the ANO2 channel, and that the N-terminal sequence may have a role in determining the channel sensitivity to Ca^{2+}. Acknowledgements: Supported by a Morley Kare Fellowship, the Human Frontiers Science Organization and NIH DC007395.

#P271 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Q8BH53, a novel protein in olfactory sensory neuron cilia

Anna K Talaga, Aaron B Stephan, Haiqing Zhao
The Johns Hopkins University/Department of Biology Baltimore, MD, USA

The cilia of olfactory sensory neurons (OSNs) are specialized for transducing odor information. Proteins that are specifically enriched in the cilia of OSNs, but not enriched in other cell types, often play roles in olfactory signal transduction. Here we report that a novel protein, Q8BH53, is highly and specifically expressed in murine OSN cilia. We initially found Q8BH53 from a mass spectrometry-based proteomic screen of OSN cilial membrane preparations. Further bioinformatic analyses revealed that Q8BH53 is conserved among ciliated eukaryotes, has no paralogs within the mammalian genome, and contains a predicted ARM-repeat domain. RT-PCR and Western blot analyses in mice showed that *q8bh53* transcripts and Q8BH53 proteins (~90kDa) are abundant in the olfactory epithelium and several other ciliated tissues, including kidney and testes. Interestingly, immunohistochemistry showed that Q8BH53 localizes specifically to the cilial layer of the olfactory epithelium but is excluded from the cilial layer of the respiratory epithelium within the nasal mucosa. The immunostaining signal was also negative in the cilia of cells in kidney and testes. Further functional analyses using molecular genetics should reveal the role of Q8BH53 in the olfactory system and perhaps in other tissues.
Acknowledgements: NIH DC007395

#P272 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

The impact of NKCC1 knock-out on olfactory sensitivity in mice

Janine Wäring, Stefan Kurtenbach, Martha Rozynkowski, Nicole Schöbel, Hanns Hatt
Department of Cell Physiology Bochum, Germany

NKCC1 is the main chloride-accumulating transporter in olfactory receptor neurons. Thus its function enhances the amplification of incoming signals when the calcium-dependent chloride channel TMEM16b is activated as the final step of the transduction cascade. Using NKCC1 gene-deficient mice, we investigated the role of the transporter in olfactory perception with the help of a behavioral paradigm as well as electroolfactograms (EOG). First we performed a cookie finding test with wild type and NKCC1-deficient mice in which the animals had to find a cookie that was buried under litter. The conditions of the experimental setup were chosen to be more and more difficult by gradually increasing the litter weight. The behavioral experiments revealed that the knock-out mice needed significantly more time to find the cookie compared to the wild type mice. Therefore, we suppose an impaired olfactory sensitivity in the NKCC1-deficient mice. Differences between the mouse lines were most pronounced in the difficult trials. At the same time, the knock-out mice did not show an impaired mobility, indicating that there was no influence of possible coordination problems in the knock-out mice. Additionally, we performed EOG recordings with the same mice used for the cookie finding test. Field potentials were recorded at different positions of the epithelium while a mixture of 100 odors (Henkel 100) was repeatedly applied via a constant airstream. Mean EOG amplitudes were about 75% smaller in the NKCC1-deficient mice. Furthermore, we noticed a slightly faster, although not significant, desensitization upon repeated stimulation in the knock-out mice. Our results give new input to the understanding of NKCC1 as the main chloride-accumulating transporter in olfactory neurons.

#P273 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Does OMP bind apo-Calmodulin?

Frank L Margolis[1], Joyce W Margolis[1], Kristen Varney[2], Hyun J Kwon[3], David J Weber[2]
[1]University of Maryland, School of Medicine/Anatomy and Neurobiology Baltimore, MD, USA, [2]University of Maryland, School of Medicine/Biochemistry and Molecular Biology Baltimore, MD, USA, [3]Andrews University/Engineering and Computer Science Berrien Springs, MI, USA

Olfactory marker protein (OMP) has been implicated as a modulator of olfactory transduction. This derives from studies of OMP-KO mice using behavioral, electrophysiological, and Ca-imaging approaches. Furthermore, the defects can be rescued by OMP replacement with OMP-expressing adenovirus. Nevertheless, the mechanism by which OMP acts is still unclear. We have demonstrated that Bex1, OMP's interacting partner protein, can bind either OMP or Ca/CaM at the same site ($Bex1_{50-75}$) suggesting that the mechanism of OMP action is in some way linked to that of Ca/CaM at various steps in the olfactory signal transduction pathway. This was supported by our observation that OMP can interact with peptides derived from several proteins in the olfactory cascade known to bind Ca/CaM (e.g. PMCA, CaMK II, NCX1). Our prior observations have now been extended to include Ca/CaM binding site peptides on the CNG channel subunits CNGA2, CNGB1, and CNGB1b which exhibit 2-10 uM affinities for Ca/CaM and 3-10 x lower affinities for OMP. In the course of these studies we unexpectedly observed that OMP and CaM can be chemically cross-linked to form a heterodimer indicating that they are proximal in solution. This interaction of OMP and CaM is more robust in the absence of Ca^{2+} indicating a preference for apo- vs. holo-CaM. NMR analyses of the interactions of 15N-OMP with apo- or holo-CaM by HSQC are consistent with this and in addition indicate multiple sites of interaction with OMP. This observation that OMP preferentially interacts with apo-CaM suggests that OMP might act to sequester free CaM in low Ca^{2+} conditions and provides additional insight to the potential role of OMP in olfaction. Acknowledgements: Supported by NIH DC003112 (FLM), Andrews-FRG (HJK) and NIH GM58888 (DW)

#P274

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Role of Olfactory Marker Protein in regulation of Na-Ca exchange activity in olfactory cilia of WT, OMP-KO and Bex1-KO mice

Manoj Tyagi, Joyce W Margolis, Frank L Margolis
Anatomy and Neurobiology, University of Maryland Baltimore Baltimore, MD, USA

Olfactory Marker Protein (OMP) is an abundant cytosolic protein of mature olfactory receptor neurons of all vertebrates. Analyses of OMP-KO mice showed that OMP plays a key role as a regulator of several steps in the olfactory signaling cascade. We previously reported that OMP regulates calcium homeostasis by controlling the efficiency of Ca^{2+} extrusion by sodium-calcium exchanger (NCX) activity in olfactory sensory neurons of OMP-KO mice. Bex1 protein has been identified as an interacting partner of OMP. Here, we address the interactions among OMP, Bex1 and NCX and their functional consequences in regulating NCX activity. We assayed reverse-mode NCX activity by sodium-dependent ^{45}Ca uptake in olfactory cilia preparations from Wild-type (WT), OMP-KO and Bex1-KO mice. The magnitude of Na^+-dependent NCX activity, in comparison to its K^+-dependent control activity, was much higher in cilia preparations from WT mice than in those from either OMP-KO or Bex1-KO mice. This reduction in NCX activity in the KO mice is consistent with our previous report of reduced Ca^{2+} extrusion from OSNs of OMP-KO mice. These functional assays demonstrate a difference in NCX activity between cilia from WT and KO mice. By contrast, RT-PCR for NCX1-3 and NCXKX1-4 mRNAs in total olfactory epithelium of WT and OMP-KO mice does not show any difference. This discrepancy between expression of exchanger mRNAs and functional activity in cilia suggests a possible defect in localization of NCX protein in the KO mice. In addition, western blot analyses of AC III, a key component of olfactory signal transduction, revealed no difference in olfactory cilia from WT, OMP-KO and Bex1-KO mice. These data collectively suggest that both OMP and Bex1 play significant roles in the regulation of NCX activity in olfactory cilia. Acknowledgements: NIH Grant RO1 DC003112

#P275

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Effect of Indole on the structure and Interactions of the odorant binding protein OBP4 from Anopheles gambiae

David N Jones, Foteini Davrazou, Emma Murphy
Dept of Pharmacology, University of Colorado School of Medicine Aurora, CO, USA

A*nopheles gambiae* mosquitoes that transmit the most dangerous form of malaria use a series of olfactory cues to locate their human hosts for a blood meal. Recognition and discrimination of the specific chemical cues occurs through the interplay of odorant receptors and odorant binding proteins. Recent studies have implicated two different odorant binding proteins, AgOBP-1 and AgOBP-4, as potential key players in the perception of indole by the mosquito. Here we show that AgOBP-4 exists in a molten globule state in solution and that binding of indole induces a shift to a more ordered conformation of the protein. This conformational shift correlates with its ability to interact with AgOBP-1. The crystal structure of AgOBP-4 solved in the presence of the indole, reveals a classical odorant binding protein fold, with indole bound at one end of a central hydrophobic cavity. We suggest that intrinsic disorder in OBPs presents a common mechanism for regulating OBP function that has not previously been fully recognized. Acknowledgements: NIH NIDCD R01DC008834

#P276

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

β-defensin Expression in the Canine Nasal Cavity

Michelle Aono[1], Jishu Shi[2], John Dennis[1], Edward Morrison[1]
[1]Anatomy, Physiology and Pharmacology Auburn, AL, USA,
[2]Anatomy, Physiology Manhattan, KS, USA

Detector dogs are exposed to many harmful or infectious agents so it is desirable to select dogs with strong immune systems. The innate immune system incudes small cationic peptides called defensins. Since the dog's olfactory acuity is its most valuable asset, â-defensin expression in the dog nasal cavity was investigated as a possible marker for immune system robustness. We posed a three part hypothesis: â-defensins are expressed in the nasal cavity; there is an olfactory specific â-defensin; and dogs, like humans, have heterogeneous expression levels of â-defensins. Tissues were collected *post mortem* from 13 dogs at the alar fold, maxiloturbinate, ethmoid labyrinth, vomeronasal organ, and olfactory bulb. RTPCR primers were designed to detect canine â-defensins 1, 2, 3, 102, 103, olfactory marker protein (OMP) and GADPH. RTPCR conditions were determined empirically. RTPCR products were sequenced to ensure primer specificity and products were run on a 2% agarose gels and stained with ethidium bromide. â-defensin 103 RNA was strongly expressed in the rostral portion of the canine nasal cavity. â-defensin 103 was not ftlineexpressed in (OMP+) tissues. The â-defensin 103 RNA expression pattern was consistent for all 13 dogs but expression levels varied between dogs. â-defensins 1, 2, 3, 102 were not detected. Of the five defensins, â-defensin 103 is highly expressed in the rostral nasal cavity. This tip of the nose expression ensures that â-defensin 103 is localized in the area of initial pathogenic exposure. â-defensin 103 is not expressed in olfactory sensory epithelia. The variation of defensin expression between individuals may correlate with the dog's ability to ward off infection. Acknowledgements: Supported in part from Grant #CA 01-G-022 from DHS (EEM)

#P277

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

The Neural Substrate for the Transformation of Olfactory Inputs into Locomotor Output in the Sea Lamprey

Elias Atallah[2], Dominique Derjean[1,2], Warren W. Green[3], Barbara S. Zielinski[3], Réjean Dubuc[1,2]
[1]Université du Québec à Montréal, Département de kinésiologie Montréal, QC, Canada, [2]Université de Montréal, Groupe de Recherche en Sciences Neurologiques, Département de physiologie Montréal, QC, Canada, [3]University of Windsor, Department of Biological Sciences Windsor, ON, Canada

Olfactory inputs can generate motor behaviours, including locomotion, in different behavioral contexts such as food seeking, social communication, and reproduction. The stereotyped nature of some of these motor behaviours (escape, attack) suggests that there is a strong neural link between olfactory inputs and motor command centers in the CNS. Using the lamprey model, we identified a neural substrate underlying the transformation of an olfactory input into a locomotor output. We found that the olfactory inputs relevant for locomotor behaviour are relayed in the medial part of the olfactory bulb and project to the posterior tuberculum in the ventral diencephalon. From there, the signal is sent to the mesencephalic locomotor region to eventually reach reticulospinal cells that act as command neurons for locomotion. Activation along this olfactory-motor pathway generates rhythmic ventral root discharges as well as swimming behavior. We also found that this pathway is present at all life stages of lampreys. On the other hand, the behavioral responses to pheromones, for instance, are elicited only at specific life stages. Modulatory mechanisms may thus be involved in this olfactory-locomotor pathway. We also found that GABA inputs powerfully modulate transmission within the olfactory bulb. The injection of the $GABA_A$ receptor antagonist, gabazine, into the olfactory bulb amplified pre-existing reticulospinal responses to olfactory nerve stimulation. Immunohistochemistry reveals a dense GABA innervation in the olfactory bulb. This suggests that GABA inputs may filter transmission at the level of the olfactory bulb in lampreys and could thus modulate locomotor output depending on the state of the animal. Acknowledgements: Funding provided by the Great Lakes Fishery Commission, the Natural Sciences and Engineering Research Council of Canada, and the Canadian Institutes of Health Research.

#P278

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Physiological and Morphological Specificity of the Medial Olfactory Bulb Region in the Sea Lamprey

Warren W Green[1], Alfred Basilious[1], Huiming Zhang[1], Réjean Dubuc[2,3], Barbara S Zielinski[1]
[1]Department of Biological Sciences, University of Windsor Windsor, ON, Canada, [2]Groupe de Recherche sur le Système Nerveux Central, Département de Physiologie, Université de Montréal Montreal, QC, Canada, [3]Département de Kinésiologie, Université du Québec à Montréal Montreal, QC, Canada

In the olfactory bulb of the sea lamprey, medial located mitral cells are part of an oligosynaptic pathway that transits through the posterior tuberculum to reach locomotor command neurons, whereas nonmedial mitral cell axons mostly project to forebrain structures. In this study we compare properties of odour specificity and mitral cell morphology between this medial region and the remaining olfactory bulb regions. Multi-unit responses to odours in an *ex vivo* preparation revealed that the medial region was responsive to basic amino acids, lamprey sex pheromones, and migratory pheromones while the lateral region was responsive only to amino acids. These properties support previous findings of diverse odour qualities triggering olfactory-locomotor transformation through the medial region of the olfactory bulb and suggest that the remaining bulbar regions exhibit chemotopy. The dendrites and cell bodies of the medial mitral cells were confined to the glomerular layer compared to the cell bodies of non-medial mitral cells, which were located largely proximal to the glomerular layer. Although the cell morphology did not differ between the medial and non-medial mitral cells, the medial somata were larger. These findings show unique neurophysiological and neuroanatomical properties in the medial region of the olfactory bulb that is involved in olfactory-locomotor transformation. Acknowledgements: Funding provided by the GLFC and NSERC.

#P279

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Subsystem-specific odorant processing in larval *Xenopus laevis*

Ivan Manzini[1,2], Sebastian Gliem[1], Eugen Kludt[1], Detlev Schild[1,2]
[1]Neurophysiology and Cellular Biophysics Göttingen, Germany, [2]DFG Research Center for Molecular Physiology of the Brain (CMPB) Göttingen, Germany

The main olfactory epithelium (MOE) of larval *Xenopus laevis* contains at least two subsets of olfactory receptor neurons (ORNs). One subset lacks the canonical cAMP transduction pathway and responds to amino acid odorants, the second subset responds to pharmacological agents activating the cAMP cascade. The ORN axons of these subsystems project to distinct glomerular clusters in the main olfactory bulb (MOB). The 'amino acid-sensitive' cluster is situated in the lateral MOB, the 'cAMP-specific' cluster in the medial MOB. Together, this clearly shows that the main olfactory system of this species is made up of different and substantially diverse subsystems. Here we set out to allocate groups of waterborne odorants (bile acids, amines, alcohols, amino acids and nucleotides) to the abovementioned subsystems using functional Ca^{2+} imaging in acute slices of the olfactory system. To do so, we recorded odorant- and forskolin-induced responses of ORNs in the MOE and odorant-induced responses of glomeruli of the MOB. By retrograde labeling of ORNs via electroporation of biocytin into the lateral and medial part of the glomerular layer, we were able to show that the two subsets of ORNs have a spatially distinct distribution in the MOE. The outcome of the present work is a further step in an effort to understand the functional significance of the different subsystems that coexist in the main olfactory system of larval *Xenopus laevis*. Acknowledgements: Supported by DFG Schwerpunktprogramm 1392 (project MA 4113/2-1) to I.M., and DFG CMPB (Project B1/9) to I.M. and D.S.

#P280

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Isomer-specific Response to Herbivore-induced Plant Volatiles in the Antennal Lobe of *Manduca sexta*

Anna M Henning[1], Silke Allmann[2,3], Sonja Bisch-Knaden[1], Andreas Reinecke[1], Silke Sachse[1], Ian T Baldwin[2], Bill S Hansson[1]
[1]Max Planck Institute for Chemical Ecology / Evolutionary Neuroethology Jena, Germany, [2]Max Planck Institute for Chemical Ecology / Molecular Ecology Jena, Germany, [3]Swammerdam Institute for Life Sciences / Plant Physiology Amsterdam, Netherlands

Volatile signals in plant-insect communication typically change composition depending on physiological state of the plant. The ability to perceive, discriminate and classify volatile compounds is crucial for insects to generate appropriate behavioral responses. In insects, olfactory sensory neurons project to the antennal lobe (AL), the first olfactory neuropil consisting of subunits known as glomeruli. We examined the olfactory response of the tobacco hawkmoth *Manduca sexta* to a group of herbivore-induced plant volatiles, so-called green leaf volatiles (GLVs). GLVs released by the host plants *Nicotiana attenuata* and *Datura wrightii* exhibit a shift from *(Z)*-3- to *(E)*-2-isomers induced by feeding *M. sexta* larvae, thereby attracting a hemipteran predator. Ovipositing *M. sexta* females should avoid plants with increased *(E)*-2-isomer emission to protect their offspring. To test whether *M. sexta* females can discriminate between isomeric ratios of GLVs, we investigated the glomerular responses in the AL to different blends of *(Z)*-3- and *(E)*-2-isomers of hexenol, hexenyl acetate and hexenyl butyrate via functional imaging. We found two isomer-specific glomeruli that were either activated by *(Z)*-3-hexenyl acetate or its *(E)*-2-isomer. Stimulation with different ratios of both isomers resulted in distinguishable activation patterns. In field oviposition experiments *Datura* plants scented with *(Z)*-3-hexenyl acetate or a *(Z)*-3-biased GLV mix were preferred over plants scented with *(E)*-2-hexenyl acetate or a GLV mix containing equal amounts of each isomer, respectively. These findings suggest that the isomer-specific AL activation pattern may contribute to discrimination of plant physiological states by female *M. sexta* for oviposition. Wind tunnel experiments are currently being performed to test this hypothesis.
Acknowledgements: Funded by Max Planck Society and BMBF.

#P281

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Medial Amygdala Circuits Involved in Response to Chemical Communication Signals

Ariel Simonton, Lindsey Silz, Michael Meredith
Florida State University/Program in Neuroscience, Dept. Biolgical Science Tallahassee, FL, USA

The hamster and mouse vomeronasal organs (VNO) detect chemosensory signals from both conspecific and heterospecific species and have strong direct projections via accessory olfactory bulb (AOB) to anterior medial amygdala (MeA), and on to posterior medial amygdala (MeP), and hypothalamic regions implicated in social behaviors. VNO, AOB and MeA respond to natural conspecific and heterospecific social signals. Conspecific and especially salient heterospecific stimuli also increase FRAs immediate early gene (IEG) expression in MeP, suggesting selective responses to biologically relevant stimulus categories. There is evidence for GABA-ergic suppression of MeP by some stimuli, either via intrinsic medial amygdala interneurons, or by inhibition from the adjacent GABA-ergic intercalated-nucleus (ICN) cells. To date we have shown that (presumed GABAergic) intrinsic interneurons expressing the calcium binding proteins, parvalbumin (PV), and calretinin (CR) in hamster appear not to be activated in circumstances where GABA-ergic inhibition is expected. Calbindin (CB) expressing interneurons are activated differentially, but largely in a pattern suggesting local feedback function rather than as a contribution to stimulus-category-specific responses. Ongoing research focuses on a potential population of inhibitory neurons not expressing calcium binding proteins, and on the ICN. A circuit comprised of a separate ICN cell-group and the basolateral amygdala is involved in fear conditioning and is modulated by dopamine via D1 receptors. Initial experiments, using D1 agonist and antagonist injections (in mice) suggest a different response to DA in the medial amygdala-ICNc circuit.
Acknowledgements: Supported by NIDCD grant DC005813 and FSU Neuroscience Program fellowships

#P282

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Effects of Sniffing on the Temporal Structure of Mitral/Tufted Cell Output from the Olfactory Bulb

Ryan M. Carey[1], Matt Wachowiak[1,2,3]
[1]Dept. of Biomedical Engineering, Boston University Boston, MA, USA, [2]Dept. of Biology, University of Utah Salt Lake City, UT, USA, [3]Dept. of Physiology, University of Utah Salt Lake City, UT, USA

Olfactory receptor neuron (ORN) inputs to the olfactory bulb (OB) are strongly patterned by sniffing, and their temporal structure is glomerulus- and odorant-specific. To investigate how sniffing affects OB outputs, we recorded from mitral/tufted (MT) cells in anesthetized rats using a custom device to "play back" sniff patterns produced by behaving rats. We found (as have others) that odorant stimulation elicited strong temporal patterning in MT cells, with each sniff evoking a stereotyped action potential burst 50-200 ms in duration. The latency to the excitatory burst was consistent across sniffs but varied across cells over the range of previously-reported ORN input latencies (~50-200 ms), suggesting that MT activity is largely driven by ORN input. The precision of spike timing varied across MT cells, with the s.d. of the time-to-first-spike ranging from ~5 to ~40 ms. Increasing sniff frequency altered MT responses, reducing peak firing rate, total spike count, and burst duration; during sustained high-frequency sniffing, the firing of some - but not all - MT cells attenuated and became less coupled to inhalation. These results are consistent with the frequency-dependent attenuation of ORN inputs, as well as a predicted increase in glomerular inhibition at higher sniff frequencies (Wachowiak and Shipley 2006). We found that MT response latencies were relatively frequency-invariant, while response phases (relative to the sniff cycle) changed significantly with sniff frequency, consistent with a role for latency rather than phase in odor coding. Together, these results suggest that ORN input dynamics play an important role in

shaping MT response properties and that rodents may modulate MT firing patterns - and thus higher-order odor representations - by changing sniffing behavior. Acknowledgements: NIDCD NRSA F31DC010312, R01DC006441

#P283 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Respiration functionally modulates lateral inhibition in the olfactory bulb

Matthew E Phillips[1,2], Robert NS Sachdev[3], David C Willhite[2], Gordon M Shepherd[2]
[1]Yale University, Department of Physics New Haven, CT, USA, [2]Yale University School of Medicine, Department of Neurobiology New Haven, CT, USA, [3]Yale University School of Medicine, Department of Neurobiology, Kavli Institute for Neuroscience New Haven, CT, USA

Respiration and olfactory processing are intimately related. In the olfactory bulb (OB), each breath phasically activates and suppresses the spiking of different neurons. However, it is not known if respiratory-coupled spiking in different OB cells is produced by nasal airflow alone, driven by excitatory circuit activity, suppression from inhibition, or both. In addition, the functional consequences of these epochs of activity and inactivity are also not well understood. Here, we recorded OB mitral and external tufted (ET) cells *in vivo* to measure the effect of nasal airflow on spiking, membrane potential, and inhibitory post-synaptic potentials arising from lateral inhibition. Mitral and ET cells showed respiration-coupled spiking, driven by excitatory synaptic inputs, only when cyclic nasal airflow was present. However, ET cells maintained high average spiking rates (~15Hz) without airflow. Furthermore, lesioning the output tract of the OB, thus removing cortical feedback, had little effect on the generation of rhythmic activity. Functionally, nasal airflow produced and modulated the strength of lateral inhibition with respiration in both mitral and ET cells. Mitral cells with strong respiration-coupled spiking showed the greatest modulation of lateral inhibition with respiration. This modulation was independent of cortical feedback, but required nasal airflow. We conclude that synaptic inputs to OB neurons, resulting from nasal airflow, shape bulbar output by producing both a baseline level of activity (respiration-coupled spiking of mitral and ET cells), and a functional modulation of lateral inhibition. Acknowledgements: This work was supported by NIH/NIDCD grants DC000086 (GMS), DC008874 (DCW), FDC009921A, and 5T32NS007224 (MEP).

#P284 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Associative Conditioning Alters Olfactory Bulb Glomerular Odor Representations

Max L. Fletcher
University of Tenn Health Science Center Memphis, TN, USA

The anatomical organization of receptor neuron input into the olfactory bulb (OB) allows odor information to be transformed into an odorant-specific spatial map of glomerular activity. In other sensory systems, neuronal representations of stimuli can be reorganized or enhanced following learning. While the OB has been shown to undergo experience-dependent plasticity, it is unclear if similar representational reorganization occurs at the glomerular level following learning. To address this, odorant-evoked glomerular activity patterns were observed in mice expressing the calcium indicator G-CaMP2 in OB output neurons. In these mice, glomerular odor representations were imaged before and after associative conditioning with foot shock. After conditioning, the responses of the individual glomeruli to the paired odor were significantly altered. These results suggest that conditioning can enhance the representation of the learned odor. Current experiments are focused on addressing on whether the plasticity observed is specific to the trained stimuli only, or if there is generalization to other, related stimuli. While the mechanisms responsible for this plasticity are currently unknown, one interesting possibility is the effect of centrifugal neuromodulation. The OB receives input from several neuromodulatory regions known to be involved in learning and memory. Of these, cholinergic fibers from the basal forebrain project heavily to the glomerular layer and can affect neuronal excitability. Experiments are underway investigating the effect of this cholinergic neuromodulation on OB glomerular plasticity. Together, these studies will further our understanding of the role that plasticity plays in shaping neural responses to sensory stimuli.

#P285 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Decorrelation of odor representations via spike timing-dependent plasticity

Christiane Linster[1], Thomas A. Cleland[2]
[1]Dept. Neurobiology & Behavior, Cornell University Ithaca, NY, USA, [2]Dept. Psychology, Cornell University Ithaca, NY, USA

The nontopographical representation of odor quality space differentiates early olfactory representations from those in other sensory systems. Decorrelation among olfactory representations with respect to physical odorant similarities has been proposed to rely upon local feed-forward inhibitory circuits in the glomerular layer that decorrelate odor representations with respect to the intrinsically high-dimensional space of ligand-receptor potency relationships. A second stage of experience-dependent decorrelation is likely to be mediated by the circuitry of the external plexiform layer. Computations in this layer, or in the analogous interneuronal network of the insect antennal lobe, are dependent on fast network oscillations that regulate the timing of

mitral cell and projection neuron (MC/PN) action potentials; this suggests a largely spike timing-dependent metric for representing odor information, here proposed to be a precedence code. We first illustrate how the rate coding metric of the glomerular layer can be transformed into a spike precedence code in MC/PNs. We then show how this mechanism of representation, combined with spike timing-dependent plasticity at MC/PN output synapses, can progressively decorrelate high-dimensional, nontopographical odor representations in third-layer olfactory neurons. Reducing MC/PN oscillations abolishes the spike precedence code and blocks this progressive decorrelation, demonstrating selectivity for these sparsely synchronized MC/PN spikes even in the presence of temporally disorganized background activity. Finally, we apply this model to odor representations derived from calcium imaging in the honeybee antennal lobe, and show how odor learning progressively decorrelates odor representations and the abolition of PN oscillations impairs odor discrimination. Acknowledgements: This work was supported by grants R01DC009948 and R01DC008702 from the National Institute on Deafness and Other Communication Disorders (NIDCD).

#P286 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Odor value represented in mitral cell synchronized firing?

David H. Gire[1], Wilder Doucette[1], Jennifer Whitesell[3], Vanessa Carmean[3], Mary T. Lucero[2], Diego Restrepo[1]
[1]Department of Cell and Developmental Biology and Neuroscience Program, University of Colorado Denver Aurora, CO, USA, [2]Department of Physiology, University of Utah Salt Lake City, UT, USA, [3]Department of Physiology and Biophysics and Neuroscience Program, University of Colorado Denver Aurora, CO, USA

Synchronized firing of mitral cells in the olfactory bulb has been hypothesized to help bind information together in olfactory cortex. In this first survey of synchronized firing by suspected mitral cells in awake-behaving vertebrates we find the surprising result that synchronized firing conveys information on odor value (is it rewarded?) rather than odor identity (what is the odor?). We observed that as mice learned to discriminate between odors synchronous firing responses to the rewarded and unrewarded odors became divergent. Further, adrenergic blockage decreases the magnitude of odor divergence of synchronous trains suggesting that mitral cells contribute to decision-making through adrenergic-modulated synchronized firing. Thus, in the olfactory system information on stimulus reward is found in mitral cells one synapse away from the sensory neuron. This is yet one more difference between the olfactory system and other sensory systems. We speculate that integration of information relevant to decision-making into the earliest stages of neural encoding helps solve the challenging problem of odor-based decision-making with a large number of input dimensions through hundreds of olfactory receptors. David H. Gire and Wilder Doucette are co-first authors of this study. Acknowledgements: Funded by NIH grants DC00566 (DR), DC04657 (DR), DC008855 (DR), DC008066 (WD), and DC002994 (ML).

#P287 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

***In vivo* functional imaging of individual neurons of the same glomerular module in the mouse main olfactory bulb**

Shu Kikuta[1,2], Shin Nagayama[1], Wei R. Chen[1]
[1]University of Texas Medical School at Houston Houston, TX, USA, [2]University of Tokyo Tokyo, Japan

Olfactory sensory neurons, expressing same type of odorant receptor, converge on specific glomeruli in which their axonal terminals synapse onto the multiple types of neurons. Based on this, neurons under same glomerulus would process similar of odor information via intra- and inter-glomerular module interactions. Although morphological differences of these neurons have been known, it is unclear how these neurons cooperate with each other during odor information processing within an individual glomerulus. For a comprehensive understanding of individual neuronal contributions to odor information processing within a glomerular module, we compared the odorant response properties of these neurons using *in vivo* two-photon calcium imaging. Individual glomeruli were visualized by the fluorescence of the Synapto-pHluorin protein expressed under the olfactory marker protein promoter. Dextran-conjugated calcium indicators were loaded into targeted glomeruli by electroporation and multiple neurons associated with a same glomerulus were labeled for the functional imaging. Juxtaglomerular cells (50-150 µm deep from the surface) showed a wide excitatory molecular receptive range and were activated by lower odor concentration. Tufted cells (150-250 µm) showed relatively wide excitatory molecular receptive range and higher odorant sensitivity too. Mitral cells (250-300 µm) showed narrowly tuned excitatory odorant selectivity and higher threshold of excitatory responses to odor stimulation compared with those of tufted and juxtaglomerular cells. These data indicate that each glomerular module is composed of different types of neurons, which have different odorant response properties. These functional differences could be related to the neuronal arrangement from the surface to deep within glomerular modules. Acknowledgements: This work was supported by NIH NIDCD grants (DC010057 to Shin Nagayama, DC003918 and DC009666 to Wei Chen).

#P288 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Human olfactory electrocorticography and stimulation

Christina Zelano, Keng Nei Wu, Stephan Schuele, Micheal Macken, Joshua Rosenow, Jay Gottfried
Northwestern University/Neurology Chicago, IL, USA

We used electrocorticography (ECoG), in which electrodes are placed directly on the exposed surface of the brain in patients undergoing pre-surgical evaluation for medically refractory epilepsy, to record odor-evoked activity patterns simultaneously from multiple human olfactory brain regions, including olfactory bulb, piriform cortex, amygdala, and orbitofrontal cortex. In 32 trials, subjects were presented with odors contained in squeeze bottles and asked to rate the pleasantness of each smell. Event

related spectral perturbation plots from a single patient (patient V) indicate that oscillatory activity in both the olfactory bulb and piriform cortex varies with pleasantness of the delivered smell. Specifically, gamma-band power increased upon presentation of unpleasant odor while beta-band power increased upon presentation of pleasant odor. In addition to recording data from the surgically implanted electrodes, we also stimulated cortex through them, allowing us to record cortico-cortical evoked potentials (CCEPs). Bipolar stimulation of electrode pairs in piriform cortex enabled us to record CCEPs from orbitofrontal cortex, providing a way to characterize network functional connectivity within the olfactory network. Maximal CCEPs were recorded from the central-posterior area of orbitofrontal cortex of patient V upon stimulation of posterior piriform cortex. Our preliminary data indicates that odors varying in perceptual pleasantness evoke unique electrical responses in the beta (13-25 Hz) and gamma (>25 Hz) frequency bands. Furthermore, our data may help resolve the question of where the olfactory projection site in human orbitofrontal cortex resides, which on the basis of functional imaging data appears to be situated 3 cm anterior to the site in primates. Acknowledgements: NIH to cmz: 1F32DC010530-01A1 NIH to jag: 610-5210200-60023390-01

#P289

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Coding of Pleasantness and perceived Intensity in the Human Olfactory System

Per Moeller, Ida Viamose, Ondrej Lassak, Gert Christoffersen
University of Copenhagen Frederiksberg, Denmark

Objective: To investigate how the human brain codes pleasantness and perceived intensity of odours. **Background:** Despite the fundamental nature of the question of how pleasantness and perceived intensity of odours are coded by the human olfactory system no consensus seems to exist. Some previous studies suggest that temporal differences (different delays of characteristic responses) code perceived intensity while others argue that perceived intensity is rather coded by amplitudes of characteristic responses. **Methods**: In this experiment we used four odours, two unpleasant (a chicken smell at high and low perceived intensity) and two pleasant (an orange smell at same high and same low perceived intensities as the chicken odours) with clearly different pleasantness (+3 and -3.5 on a scale from -5 to +5 for the most intense odours (7 on a scale from 0 to 10) and +1 and -2 for the less intense odours (2 on the intensity scale from 0 to 10). Twenty Ss each received 36 trials of each of the four odours and a blank control in semirandom sequences divided over 3 sessions with 15 min in between. Each odour was delivered for 2.5 sec followed by a blank interval of 9.5 sec before next odour delivery by a constant flow olfactometer. EEG signals were recorded from 64 electrodes. **Results and conclusions:** For all odours we find significantly more smell induced modulation of power in the delta band (0.5-3.5 Hz) than in both the theta band (3.5-7.5 Hz) and the alpha band (7.5-12.5 Hz). The power of responses is modulated by perceived intensity, but the modulation is much larger for the unpleasant odour, suggesting a contribution of delta band activity in coding of olfactory pleasantness. Furthermore, there are indications in the data that alpha activity codes *identity* rather than pleasantness or perceived intensity.

#P290

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Pleasantness of food odor negatively associated with hunger

Lorenzo D. Stafford
University of Portsmouth Portsmouth, United Kingdom

Recent research has provided evidence that olfactory sensitivity to food and non-food odors varies as a function of hunger state, with additional differences according to BMI. The research here aimed to extend this research using a larger sample and simplified measure of olfactory threshold. Participants (n=104) attended one session where following baseline ratings of hunger, they completed pleasantness ratings and threshold tests for a food and non-food odor. Results revealed that for those participants tested with the food odor first, there was a significant negative association between hunger state and pleasantness of the food odor, with no corresponding effect for the non-food odor. This suggests that when hunger state has not been manipulated directly, against expectation, increases in hunger predict lower pleasantness ratings for an odor associated to food.

#P291

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Behavioral Learning of Complex Odor Mixtures

Robert L. Rennaker[1], Adam Lovitz[1], Donald Wilson[2]
[1]University of Texas At Dallas Richardson, TX, USA, [2]Nathan Kline Institute Orangeburgh, NY, USA

In order to more fully understand olfactory processing we trained animals to discriminate complex odor mixtures. The mixtures consist of 10 standard components with one component removed. We then add in another odor component at 4 different increasing concentrations from 0ppm up to 1000ppm. Our results demonstrate it takes longer to learn to discriminate the lower concentration mixtures compared to the higher concentration mixtures. We also found that if the animals trained on another odor set and were returned to a previous odor set, they were still able to discriminate all concentrations. We plan to use this paradigm to understand odor learning in awake behaving subjects. Acknowledgements: NIDCD R01 DC008982

#P292

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

The behavioral characteristics of odor preference decision during multi-AFC task

Shiori Nakano, Saho Ayabe-Kanamura
University of Tsukuba Tsukuba, Japan

We have investigated the contribution of smelling behavior to odor preference decision and choices in humans. For vision study, a bias in looking behavior prior conscious decision during two alternative forced choice (2-AFC) preference decisions (a gaze bias effect) was demonstrated. It is also demonstrated that, in

Multi-AFC task, from the very first dwell the task relevance of the fixated items are evaluated, and this evaluation partially determines the first dwell duration. Then we confirmed whether this phenomenon would occur in olfaction as well. In our study, participants chose the most preferred odor among six pleasant odors (flavored tea) with their way of free choice. Participants were not instructed odors name and they could smell same odor repeatedly and there was no limit of the required choice time. There were two condition of alternative similarity. In one experimental condition, alternatives were very similar each other. In second condition, there were dissimilar. We measured participants' smell duration by each item for the first smell in a trial and compared finally chosen odor with other not-chosen odor. As results, a smell bias that referred vision preference decision was not shown. However, reflective participants showed a tendency to smell odor eventually prefer longer than the other at the first smell in trial on similar condition. And there were several behavior patterns on the choice. Some participants could distinguish like odors from other alternatives briefly after once they experienced each odor. And they were also shown a smell bias like reflective participants as well. It is concerned that information process style is one of the factor for odor preference decision.

#P293 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Consistently Naming and Remembering Odors

Adriana M. Reedy, Robert A. Frank
University of Cincinnati Cincinnati, OH, USA

The current study investigated the relationship between odor naming and odor memory by examining how the ability to name an odor affects remembering the odor at a later time. Many past studies have found little or no decline in odor memory over time. However, Olsson et al. (Chemosensory Perception, 2009) showed that accurately named odors were remembered better than inaccurately named odors after a 15-minute retention interval and that there was significant forgetting of both accurately and inaccurately named odors after a seven day retention interval. More recently, Frank et al. (Chemical Senses, 2010) established that consistent naming is a better predictor of odor memory compared to accurate naming. Therefore, the current study measured both naming accuracy and consistency. The current study used a conventional two-phase recognition memory test where participants were to remember whether odors had been presented in Phase 1 during a Phase 2 test. The first phase contained ten odors and the second phase contained the same 10 odors (the old odors) along with10 new odors. Using logistic regression to analyze the results, the current study found that there was no effect of a retention interval on memory when both new and old odors were combined. However, just as Olsson et al. (2009) demonstrated, significant forgetting was observed for the old odors over time. There was no effect of the retention interval on memory when only new items were assessed. A significant decrease in the consistency of odor naming that paralleled the memory findings was also observed. Naming consistency was the best predictor of memory performance. These findings provide evidence for a decline in odor memory over time, and replicate the strong relationship between consistent odor naming and recognition memory.

#P294 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

The Relationship Between Odor Naming, Consistency, and Memory is Not Affected by the Difficulty of an Odor Naming Task

Konstantin A. Rybalsky, Adriana Reedy, Robert A. Frank
University of Cincinnati Cincinnati, OH, USA

A very strong relationship was observed between consistent odor naming and recognition memory in a recent series of studies (Frank et al., Chemical Senses, 2010). The current studies sought to expand the scope of the previous work by systematically examining the relationship between consistent naming and memory under conditions known to affect odor naming performance. Engen (American Scientist, 1987) demonstrated that odor naming performance declines when a list of possible odor labels is categorically similar as compared to when the odor label alternatives are dissimilar. The current series of studies manipulated the similarity of odor labels in the odor naming component of a dual odor naming/recognition memory task. Participants chose among four odor labels as part of the naming task- one correct, normative label and three other, incorrect labels. Participants named and remembered odors under conditions where response alternatives were categorically unrelated, moderately related, or closely related. In an additional study, the normative ("veridical") odor label was not provided at all, leaving only closely related (but incorrect) response alternatives. As expected, naming accuracy declined as the response alternatives became more similar, and a modest decline in memory performance was also observed. However, the relationship between consistent naming and recognition memory remained strong across experimental conditions. Across conditions, consistent naming was associated with 94% correct memory responses while inconsistent naming was associated with 27% correct memory responses. These findings provide additional support for a strong connection between episodic odor memory and consistent odor labeling in the dual odor naming/memory task.

#P295 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

The Relationship Between Odor Naming and Consistency is Procedure-Dependent

Trevor C. Cessna, Robert A. Frank
University of Cincinnati Cincinnati, OH, USA

We have reported a strong relationship between odor naming and recognition memory in a recent series of experiments. The basic finding of these studies is that consistently and/or correctly named odors are associated with excellent recognition memory while memory is poor or non-existent for odors named inconsistently and/or incorrectly. The findings were observed when a dual odor naming/memory task was employed. The aim of the current studies was to determine if a similar relationship between naming and memory would be observed when odor recognition memory and odor naming were tested separately. Participants smelled 10 odors and were told to remember each

during the first phase of the study, and then, following a 10 minute retention interval, they were asked which of a set of 20 odors had been experienced during the initial phase of testing. During this second phase, half the stimuli were from the first phase, and half were new. In the third phase, participants named each of the 20 odors twice in two blocks of trials. One group of participants was provided with four possible odor labels for each stimulus as they named them, while another group was required to generate their own names for the odors. We found that while overall recognition memory performance was akin to that observed in previous studies, the predictive power of consistently and/or correctly named odors was *not* predictive of recognition memory performance. We conclude that the predictive power of odor naming is dependent on the experimental procedure employed. The results make it clear that a variable such as odor knowledge is not responsible for the strong relationship between odor naming and memory observed in the dual odor naming/memory task.

#P296 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

More often people contacted the odor even unconsciously, the more they come to like it?

Midori Ogawa[1], Saho Ayabe-Kanamura[2]
[1]Graduate School of Comprehensive Human Sciences Master's Program in Psychology, University of Tsukuba Tsukuba, Japan, [2]Comprehensive Human Science, University of Tsukuba Tsukuba, Japan

The effect that human comes to form favorable attitude to stimuli when one consciously or unconsciously contacts them repeatedly is called "mere exposure effect". This effect has been investigated by using various kinds of stimuli such as characters, faces, melodies, and odors. However in many researches using odors, they were exposed to participants under conscious. In our pre-experiment, this effect was examined using odor stimuli in concentration below and upper their thresholds. As results, when subthreshold odor stimuli were presented twenty times, these odors were evaluated more pleasant than these presented five times. On the other hand, when suprathreshold odors were presented five times, these odors were evaluated more pleasant than these presented twenty times. The contact even with subthreshold odor stimuli seems to form favorable attitude to the odor. Therefore, it is necessary to examine this effect when odor stimuli were contacted unconsciously. So the aim of this study was to confirm that human comes to form favorable attitude to the odor stimuli when one unconsciously contacts it repeatedly. In this study, commercial shampoos were used as odor stimuli. In order to contact odors unconsciously, participants were performed color matching task between colored-shampoos with color samples without explaining the object of this study. In the color matching task, three shampoos were used and each shampoo was presented in the different frequency. After the task, hedonic value of three exposed shampoos and three unexposed novel shampoos were rated and compared. It is predicted that odors that are presented repeatedly are more pleasant than novel odors.

#P297 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Olfactory Training in Older Adults

Beverly J. Cowart, Kaitlyn Abrams, Ryan Crawford, Marcia L. Pelchat
Monell Chemical Senses Center Philadelphia, PA, USA

Age-related olfactory loss (presbyosmia) is the rule rather than the exception, and there is currently no effective treatment or preventative measure for this form of loss. Numerous studies on adult rodents and humans show that periodic exposure to an odorant under a variety of conditions can lead to improvements in olfactory sensitivity. Most notably, Hummel et al. (Laryngoscope, 119:496-9, 2009) reported improvements in threshold sensitivity to some odorants in hyposmic patients who sniffed 4 odorants twice daily for 12 weeks, with 30% of patients showing clinically significant improvement. However, this approach has not been specifically tested in an aging population. We report here a pilot study to determine if daily sniffing of odorants can lead to improvements in olfaction in elderly adults. We further examined whether exposure to complex, natural odor stimuli (n=15) might lead to greater or more generalized improvement than exposure to pure compounds (n=15). Thresholds to the exposed odorants and one unexposed odorant were assessed at baseline, 6 and 12 weeks. Control subjects (n=10) who sniffed odorless compounds were tested with either the complex or pure odor stimuli. Subjects ranged from 64 to 82 years of age, and were predominantly women (n=32). Not all subjects have completed the protocol, but preliminary analyses indicate that significant improvements in sensitivity to exposed odorants occurred at 12 weeks in those exposed to complex stimuli, with approximately 50% of these subjects showing substantial individual improvement. However, those exposed to the pure compounds do not appear to have improved, and control subjects also appear to show no change in olfactory sensitivity. Finally, neither of the exposed groups evidence improved sensitivity to the unexposed odorant.
Acknowledgements: P50 DC006760-5, P50 DC006760-05S2, ARO W911NF-11-1-0087

#P298 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Do More Recent Generations Have Better Olfaction?

Carla R. Schubert[1], Weihai Zhan[2], Alex Pinto[1], Mary E. Fischer[1], Guan-Hua Huang[3], Barbara E.K. Klein[1], Ronald Klein[1], Karen J. Cruickshanks[1,4]
[1]University of Wisconsin, Department of Ophthalmology&Visual Sciences Madison, WI, USA, [2]Yale University School of Medicine New Haven, CT, USA, [3]National Chiao Tung University, Institute of Statistics Hsinchu, Taiwan, [4]University of Wisconsin, Department of Population Health Sciences Madison, WI, USA

The prevalence of olfactory impairment is high in the general population and is more common in older age groups. But does the prevalence of impairment vary by generation? By comparing the prevalence of olfactory impairment among people when they are the same age, but born in different years, it can be determined if the prevalence of olfactory impairment is changing across

generations in the United States. The San Diego Odor Identification Test (SDOIT) was used to measure olfactory ability in two longitudinal cohort studies; the population-based Epidemiology of Hearing Loss Study (EHLS; 1993-present) and the Beaver Dam Offspring Study (BOSS; 2005-present). SDOIT data from three examinations in the EHLS (1998-2000, 2003-2005, 2009-2010) and one examination in the BOSS (2005-2008) were combined for participants aged 53 years and older. Alternating logistic regression models were used to determine the effect of birth year on the prevalence of olfactory impairment. Preliminary analyses found the odds of having impaired olfaction decreased by 8.2% for women and 18.7% for men with every 5 year increase in birth year in a model adjusted for age (Odds Ratio (OR)=0.92, 95% Confidence Interval (CI) = 0.84, 1.01 and OR= 0.81, 95% CI = 0.74, 0.89, respectively, for every 5 birth years). Adjusting for additional covariates associated with the prevalence of olfactory impairment, a history of stroke or epilepsy and cold or sinus problems, did not change these results. These preliminary results indicate the prevalence of olfactory impairment is decreasing among adults, particularly men, in more recent generations suggesting modifiable risk factors are associated with the development of olfactory impairment. Acknowledgements: R01AG021917 R37AG011099 U10EY06594

#P299

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Human sweat odor precursors detected in human milk, colostrum and amniotic fluid

Constanze Hartmann[1], Sébastien Doucet[4], Yvan Niclass[3], Ralf Dittrich[5], Susanne Cupisti[5], Benoist Schaal[4], Christian Starkenmann[3], Andrea Buettner[1]
[1]University of Erlangen, Food Chemistry Erlangen, Germany, [2]Fraunhofer IVV, Sensory Analytics Freising, Germany, [3]Firmenich Geneva, Switzerland, [4]Université de Bourgogne, Developmental Ethology and Cognitive Psychology Group Dijon, France, [5]University of Erlangen, University Hospital Erlangen (OB/GYN) Erlangen, Germany

Objectives. The volatile thiol (R)/(S)-3-methyl-3-sulfanylhexan-1-ol [1] and the fatty acids (E)/(Z)-3-methylhex-2-enoic and (R)/(S)-3-hydroxy-3-methylhexanoic acid [2] are the major components of human axillary odor. They are secreted as glutamine- and cysteinglycine-S-conjugates in the axilla and released by human skin bacteria. This study aimed to identify these human specific odor precursors in human milk (HM) and amniotic fluid (AF). **Methods.** The identification of the odor precursors in HM and colostrum was performed by means of UPLC-MS/MS after fat extraction and purification by column chromatography. The AF samples were filtered and injected directly without concentration. The quantification was carried out by using external calibration. **Results.** The glutamine-*N*-α-conjugates of the (E)/(Z)-3-methylhexen-2-enoic and the (R)/(S)-3-hydroxy-3-methylhexanoic acid and the cysteinglycine-S-conjugate of the (R)/(S)-3-methyl-3-sulfanylhexan-1-ol were identified in HM and colostrum samples. Concentrations are in the range of µg/kg for the glutamine conjugates and pg/kg for the cysteinglycine-*S*-conjugate respectively, with a tendency towards higher contents in colostrum compared to HM. In AF, only the precursor of the 3-hydroxy-3-methylhexanoic acid could be identified, whereas in cow milk, none of these precursors were detectable. **Conclusions.** The odor precursors formerly found only in human sweat could now be identified also in HM and AF, whereby the concentration in HM seems to vary with the lactation period. They seem also to be human specific as they were not detectable in cow milk and were never found in other animals or plants until now. **References**. [1] Natsch et al. 2004. Chemistry & Biodiversity 1(7): 1058-1072 [2] Zeng et al. 1991. Journal of Chemical Ecology 17(7):1469-1492 Acknowledgements: Financed by the German Federal Ministry of Education and Research (BMBF)and the Bavarian Research Foundation.

#P300

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Human communication of emotion via sweat: how specific is it?

Monique A. Smeets[1], Alexander Toet[2], Rosalie Duinkerken[1], Jasper H de Groot[1], Annemarie Kaldewaij[1], Marcel van den Hout[1], Gün R. Semin[1]
[1]Utrecht University Utrecht, Netherlands, [2]TNO Human Factors Soesterberg, Netherlands

Females evaluate ambiguous facial expression – morphed between happy and fearful – faces as more fearful when exposed to fear sweat as compared to control odor (Zhou & Chen, 2009). We investigated the *specificity* of this effect, i.e. whether processing of *fearful* faces is affected specifically by *fear* sweat. In **Study 1**, we investigated the influence of the sensory properties of sweat in general, such that females may have a general tendency to disambiguate towards fearfulness in the context of any sweat odor. Thirty females evaluated ambiguous faces as either happy or fearful when exposed to fearful sweat, sports sweat or control pad. Using intensity evaluations of the sweat as covariates in the analysis, initially significant differences between fearful sweat and sport sweat in the facial evaluation task (p = .029) remained significant (p = .031). In **Study 2** we investigated whether facial expressions are evaluated as fearful also in the context of sweat associated with another negatively valenced emotion: disgust. Twenty-four females performed the same task when exposed to fear sweat, disgust sweat or control pad. Significant differences emerged at one of the central morph levels, with the difference between fear and control significant at p = .014, between fear and disgust at p = .033, but ns between disgust and control pad. However, only the difference between fear and control remained after covariance-analysis leaving the difference between fear and disgust sweat insignificant. **Conclusion:** Evidence so far supports the notion of specific effects for *emotional* sweat versus *non-emotional* sweat, but *not* that of specificity *between* two negatively valenced emotional states. Acknowledgements: Supported by NWO-Vidi 452-03-334 to M.A.M.S. and TNO

#P301

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Human Pheromones, Epigenetics, Physiology, and the Development of Animal Behavior

James V Kohl[1], Linda C Kelahan[2], Heather Hoffmann[2]
[1]Stone Independent Research, Inc. Phoenix, NY, USA, [2]Knox College/Psychology Galesberg, IL, USA

Androsterone, as used here, smells like fresh sweat. It is an individual human male-specific and somewhat primate-specific part of a mixture of axillary chemical secretions that contain androstenol, which influences levels of luteinizing hormone (LH) and mood in women. LH is a hormonal measure of diet-dependent sexual maturity and fertility, which is influenced by mammalian pheromones. Mammalian conditioning paradigms suggest that androstenol conditions hormonal effects in females, which may be unconsciously associated with behavioral affects of androsterone in women. We evaluated individual video-taped fifteen-minute interactions of fourteen women with fertile phase levels of LH during a cooperative task. During the task, our male accomplice wore either a standardized androstenol / androsterone mixture diluted in propylene glycol, or just the diluent — with sandalwood odor added to keep him blind to his condition. When he was wearing the mixture compared to when he wore the diluent, women were more likely to make eye contact ($t(12) = 3.43$, $p = .01$; IRR: $r = .964$, $p = .01$). They also laughed more ($t(12) = 5.20$, $p < .01$; IRR: $r = .810$, $p = .01$), and they subsequently rated themselves as being more attracted to him ($t(12) = 2.786$, $p = .016$). Our results combine the known effects of androstenol on LH and on mood with a likely behavioral affect of androsterone. They also address contrarian opinions and extend to human females a eusocial insect model for the epigenetic effects of diet and of pheromones on hormone-mediated gene expression during behavioral development. Our mixture characterizes species-specific human pheromones, their epigenetic effects on physiology, and their affect on behavior. Our results are consistent with a validated, unaltered, decades-old, across-species concept of pheromones.

#P302

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

The Scent of Age: Can Humans Determine Age Based on Body Odor Perception Alone?

Susanna D. Mitro[1], Amy R. Gordon[2,3], Johan N. Lundstrom[2,3,4]
[1]Swarthmore College Swarthmore, PA, USA, [2]Monell Chemical Senses Center Philadelphia, PA, USA, [3]Dept. of Clinical Neuroscience, Karolinska Institute Stockholm, Sweden, [4]Dept. of Psychology, University of Pennsylvania Philadelphia, PA, USA

Scientific and anecdotal evidence suggest that our body odor changes as we grow older. It has been demonstrated that rodents are skilled at discriminating between body odors from young or old individuals, an effect attributed to differences in the chemical composition of fatty acids in an aged individual's body odor. Recent data demonstrates that similar age-related fatty acid changes also occur in the body odor of aged humans. In light of this, we tested whether humans, like rodents, can extract age-related information from body odors alone. Body odors were sampled from three age groups [Young (20-30), Middle-aged (45-55), Old (75-95)] using nursing pads sewn into the armpits of t-shirts. Body odor donors bathed using odor-free soap each night, washed their bedding with odor free detergent, and slept in the shirts for five nights. Super-donor stimuli were then created by combining the body odor from four individual participants to eliminate effects based on individual odor differences and to adhere to previous experiments in rodents. Participants' ($n = 32$; mean age 24) ratings of intensity and pleasantness of the body odor were significantly affected by the age of the body odor donor. Additionally, there was an interaction between the sex of the body odor donor and the sex of the rater for the perceptual ratings. Preliminary analysis demonstrates that female raters can discriminate between ages of female donors based only on their body odor in a two-alternative forced choice test. However, subjects were unable to accurately assign a correct age to each body odor using a sorting task, a visual analogue scale, or a verbal report. These preliminary findings indicate that humans can discriminate age based solely on body odor in a side-by-side comparison but with little to no conscious awareness.
Acknowledgements: Supported by a grant from the National Institute on Deafness and other Communication Disorders (NIDCD R03DC009869) awarded to JNL.

#P303

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Sniffing out the gender: Human steroids subconsciously modulate gender perception of biological motion in a sexual dimorphic manner

Wen Zhou[1], Xiaoying Yang[1], Yi Jiang[1], Sheng He[2]
[1]Institute of Psychology, Chinese Academy of Sciences Beijing, China, [2]Department of Psychology, University of Minnesota Minneapolis, MN, USA

Recent studies have pointed to the existence of human sex pheromone, with particular interest on two human steroids: androsta-4,16,-dien-3-one and estra-1,3,5(10),16-tetraen-3-ol. The current study takes a critical step to test their qualification by examining whether the two substances produce sex specific effects on gender perception. Using point-light walkers whose gender is digitally morphed from male to female, we show that smelling androstadienone systematically biases females, but not males, towards perceiving the walkers as more masculine, whereas smelling estratetraenol systematically biases males, but not females, towards perceiving the walkers as more feminine, as compared with the control clove oil carrier solution. These effects are obtained despite that the olfactory stimuli are perceptually indiscriminable. Our results provide the first direct evidence that the two human steroids communicate opposite sexual information that is differently decoded by the two sex groups, and demonstrate that human visual gender perception draws on subconscious chemosensory biological cues, an effect that has been hitherto unsuspected. Acknowledgements: Knowledge Innovation Program of the Chinese Academy of Sciences grants KSCX2-YW-R-250, KSCX2-YW-R-248, and 09CX192019.

#P304

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Evidence that the Putative Human Pheromone Androstadienone Modulates Women Intrasexual Competition: the Case of Face Processing

Valentina Parma[1], Roberto Tirindelli[2], Angelo Bisazza[1], Stefano Massaccesi[1], Umberto Castiello[1]
[1]University of Padova Padova, Italy, [2]University of Parma Parma, Italy

Evidence suggests that specific compounds found in human body secretions influence the behavior and physiology of other individuals. Amongst these odorant, the most intensively studied is the steroidal compound androstadienone (4,16-androstadien-3-one), a chemical which is known to constitute a sexual signal that conveys to women information about male mate quality. An issue which remains untested is whether the effects of androstadienone on such function vary depending on women's menstrual cycle phase. Here we asked heterosexual women exposed to either androstadienone or a control compound to view stimuli including females' faces, males' faces and familiar objects while their eye movements were recorded. The results indicate that women at high conception risk spent more time viewing females' than males' faces irrespective of the administered compound. Women at a low conception risk exhibited a preference for female faces only when exposed to androstadienone. These data are discussed in light of the possible effects that androstadienone might have in triggering women intra-sexual competition via a possible influence on endocrine balance.

#P305

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

Is Androstadienone a Social Odor in Older Adults? Thresholds and Attention to Emotional Stimuli

David W. Kern, Martha K. McClintock
Department of Comparative Human Development and Institute of Mind and Biology, University of Chicago Chicago, IL, USA

Δ 4,16-androstadien-3-one (androstadienone, AND) is found in all human bodily fluids and axillary hair. In a young adult population, AND odor thresholds are bimodally distributed, affected by reproductive status, and are not associated with overall olfactory acuity for physical odors. In young adults, AND is a particularly important example of a social odor. AND alters behavior and neuroendocrine function at high concentrations as an odorant, as well as at nanomolar concentrations, below odor detection. Although much research has focused on its function in a sexual context, recent evidence indicates a broader social function. Recently, we have demonstrated that subliminal exposure to AND heightens attention to emotional stimuli in young adults. The current research tests odor thresholds and behavioral effects in older adults who are nearing the end of their reproductive lifespans. Olfactory thresholds for 30 older adults were determined by administering well-validated staircase threshold tests using a social odorant (AND) and a standard physical odorant (n-butanol). Behavioral effects of exposure to AND on attention to emotional information were tested by subliminally presenting pairs of faces, one neutral and the other happy or angry, and then measuring the latency to fixate on a visible target either contralateral or ipsilateral to the emotional face (the dot-probe protocol used in our lab to demonstrate subliminal effects of AND in young adults). We will investigate the relationship between the threshold distributions for each odorant, the effect of AND on perception of subliminal emotional faces, as well as the relationship between AND thresholds and perception of subliminal emotional faces in this older sample. Acknowledgements: The National Social Life, Health, and Aging Project Wave II (R37 AG030481), Gianinno Graduate Research Fund

#P306

POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING

The effect of menstrual cycle phase and oral contraceptive use on odor hedonics

Cathryn A. Griffiths[1,2], Jelena Djordjevic[1]
[1]Montreal Neurological Institute, Cognitive Neuroscience Unit Montreal, QC, Canada, [2]McGill University, Department of Psychology Montreal, QC, Canada

Few studies have examined the variations of perceived odor pleasantness as a function of menstrual cycle phase. The purpose of this study is to examine the effects of menstrual cycle phase on hedonic ratings of a large number of odors in a large sample of women. The experiment was a mixed-model design, with between-subjects factors of group (2 levels: oral contraceptive [OC, n=20] users and spontaneously ovulating [SO, n=20] women) and a within-subjects factor of testing time (3 levels: days 1-6 [menstruation], days 12-18 [ovulation or mid-cycle for OC users], and days 18-33 [luteal phase]. In each testing session, subjects completed the Profile of Mood States (POMS; McNair et al., 1971), the Sniffin' Sticks (Hummel et al., 1997) n-butanol threshold test, and rated 5 food odors (orange oil, cinnamon, meat pate, rum, coffee), 5 environment odors (rose, pine, lavender, nicotine, guiaicol), 5 cosmetic odors (body lotion, men and women's deodorant, perfume, shampoo), and androstenone for pleasantness, intensity, familiarity, and edibility on a 10-cm visual analog scale. An analysis of variance revealed no significant main effects or interactions on the POMS and n-butanol threshold. Most ratings did not show effects of testing time or group, but there were notable exceptions. Pleasantness of rum was rated significantly higher during the ovulatory compared to the menstrual phase by SO women. Familiarity of men's deodorant was rated significantly higher by SO women compared to OC users. SO women rated guiaicol as significantly more edible in the luteal phase than OC users did. These results suggest that both menstrual cycle phase and oral contraceptive use can have an effect on the perception of certain odors, pointing to a compelling avenue for future research. Acknowledgements: This research was supported by an NSERC discovery grant (awarded to J.D.) and an NSERC Summer Research Award (awarded to C.A.G).

#P307 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Fragrance change impacted interactions of close female friends

Melissa Bart, Robin Freyberg
Stern College for Women, Department of Psychology New York, NY, USA

The current study examined whether disrupting the olfactory environment would impact the interaction of pairs of close friends through examining questionnaires and psychophysiology. One hundred pairs of female undergraduate close friends participated in two fifteen minute interactions. The first session was established as a baseline interaction for all participants. In the second session for half of the pairs, one member of the dyad was given a different perfume to wear; for the other half one participant was given a different watch to wear as a non-olfactory control condition. In questionnaires, disrupting the olfactory environment impacted fragrance-wearers only. Specifically, a repeated measures ANOVA revealed there was a session x fragrance condition interaction, $F(1, 46) = 4.01$, $p = .05$. Only participants in the unfamiliar fragrance condition reported lower levels of enjoyment in the second session compared to the first, $t(22) = 3.22$, $p = .004$. Further, perceptions of closeness decreased for both participants when the olfactory environment was disrupted, $F(1, 46) = 7.06$, $p = .011$ such that all participants reported lower levels of closeness following the second session than the first. Analyses of psychophysiology data revealed that there was a trend for a session x condition interaction for beats per minute (BPM), $F(1, 72) = 3.74$, $p = .057$. In the fragrance conditions, participants continuing their regular fragrance routine exhibited a decrease in BPM which was not observed in the unfamiliar fragrance condition. No changes in questionnaires or psychophysiology were observed in the non-olfactory control condition. Such findings suggest that exposure to the unfamiliar fragrance during the second session dynamically and rapidly affected close relationships.

#P308 **POSTER SESSION VI: OLFACTION: PERIPHERY; OLFACTORY CNS; PSYCHOPHYSICS; HUMAN CHEMICAL SIGNALING**

Body Odor Origin and Its Effects on Emotional Processing

Amy R. Gordon[1,2], Mats J. Olsson[1], Johan N. Lundstrom[1,2,3]
[1]Karolinska Institute, Department of Clinical Neuroscience Stockholm, Sweden, [2]Monell Chemical Senses Center Philadelphia, PA, USA, [3]University of Pennsylvania, Department of Psychology Philadelphia, PA, USA

It was recently demonstrated that smelling a body odor originating from an unknown individual (Stranger) elicits increased activity within the amygdala (Lundstrom et al., 2008), a cerebral structure known to process fear-relevant stimuli. In this study, we hypothesized that smelling a Stranger's body odor (ST) would modulate the speed of reaction and autonomic responses to fear-relevant, but not to non fear-relevant, visual stimuli relative to smelling oneself (SE) or a perceptually similar but fake body odor (Fake). To eliminate any effects based on individuals' odors, super-donor ST stimuli were created by combining the body odors of four different Stranger participants. The behavioral task involved speeded detection of either an angry face (fear-relevant) or happy face (non fear-relevant) hidden within an array of neutral faces. Twenty-two participants were presented with body odors intra-nasally by a computer-controlled olfactometer while performing the detection task. Measures of evoked autonomic responses were also acquired. Preliminary analyses demonstrate that participants reacted significantly faster to fear-relevant than to non fear-relevant stimuli ($p<.001$), indicating effective visual stimuli, but there was no main effect of ST odor relative to Fake odor on reaction time to either visual stimulus. Unexpectedly, however, SE odor attenuated reaction time to the non fear-relevant stimuli relative to ST odor in an interaction effect analysis, an effect that was replicated in a separate behavioral experiment. Autonomic measures and two separate behavioral experiments will be presented. Notwithstanding, preliminary analyses indicate that there are no strong behavioral effects of exposure to the scent of a Stranger but that there may be minor positive effects of exposure to one's own scent.
Acknowledgements: Supported by a grant from the National Institute on Deafness and other Communication Disorders (NIDCD R03DC009869) awarded to JNL.

#P309 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Expression and ecological significance of the gustatory receptor gene family of two divergent Daphnia species – D. pulex and D. magna

D. Carolina Penalva-Arana, Michael Lynch
Indiana University/Biology Bloomington, IN, USA

Our study investigated the expression of the gustatory receptor (GR) gene family of two divergent *Daphnia* species, *D. pulex* and *D. magna*. We began by annotating the GR genes of the newly sequenced *D. magna* genome. *D. pulex* has 58 GR genes while an exhaustive search of the *D. magna* genome reveals only 38 GR genes. Gene number differences may be attributed to differences in chromosome number as well as extensive gene duplication in the *Daphnia* subgenera. 27 *D. magna* GR genes have direct orthologs in *D. pulex*, and there is only one *D. magna* subclade that has undergone species-specific expansion. In *D. pulex*, qPCR expression data reveals that 53 of 58 genes are expressed in both sexes, but males express most genes at a higher level than females. One gene, *DapuGr*40, has a 424-fold expression increase in males. We hypothesize that *DapuGr*40 may bind the highly elusive *Daphnia* female pheromone, and finding its ligand will reveal the nature and identity of crustacean pheromones. In *D. magna* 36 of 38 GR genes are expressed across both sexes. Curiously in *D. magna*, two GRs appear to be female specific, but none appear to be male-specific. Gene expression across two distinct embryonic stages reveals that only 43 genes are expressed in *D. pulex* embryos. Of these, 2 GR genes, *DapuGr*24 and 48, are expressed higher in embryos than in adults. In *D. magna* current data indicates that only 24 GR genes are expressed in embryos. Alternative splicing was observed in both embryonic and adult stages and will require further investigation. We conclude by hypothesizing that GR genes with high expression during embryonic development are ecologically relevant and essential for guiding phenotypic plasticity under varying environmental conditions and require further investigation. Acknowledgements: NIH GM871342

#P310 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Comparative Analysis of the Chemoreceptor Gene Family of *Daphnia pulex* and *Daphnia magna* Reveals Intron Gains and Extensive Gene Duplications in *D. pulex*

Richard N Keith, Michael Lynch, D. Carolina Penalva-Arana
Indiana University Bloomington, IN, USA

In 2009, Penalva-Arana et al. published on the gustatory receptor (GR) gene family of *D. pulex.* Recently the Daphnia Genome Consortium has sequenced the genome of *D. magna*. These two species belong to two distinct subgenera, *Daphnia* (*D. pulex*) and *Ctenodaphnia* (*D. magna*), having a divergence of ~10 million years. The objective of our study was to utilize these gene models to manually annotate the GR genes in the *D. magna* genome, and uncover the evolutionary trajectory of this gene family across the two divergent species. The 58 GRs found in *D. pulex* span across 19 scaffolds, and our *D. magna* annotation reveals only 38 intact genes present across 12 scaffolds and 2 contigs. Gene structure and synteny are well-conserved in orthologs, but a high degree of duplicate gene movement and intron gains are observed in *D. pulex*. We hypothesize that GR gene number could differ between *D. magna* and *D. pulex* because of incomplete sequencing, but is most likely due to gene duplications that occurred in the *Daphnia* subgenera. Using the codeml package in PAML, we are currently identifying sites under selection across orthologs and gene duplicates in these two *Daphnia* species. A further investigation into the partially sequenced genome of *D. obtusa*, a species closely related to *D. pulex,* is also currently underway. Synteny and gene structure is conserved in *D. obtusa* and *D. pulex* suggesting gene duplication events may be shared by all species in the *Daphnia* subgenera. These differences in gene duplicates may allow different *Daphnia* species, such as those in the *Daphnia* subgenera, to better adapt to changing and diverse environmental conditions, and further investigation is required on the function and expression of these gene duplicates. Acknowledgements: NIH GM871342

#P311 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

***Daphnia* Clonal Variation of Sex-Biased Chemoreceptor Genes Across Sexual and Asexual Lineages**

Robert A. Sommer, Michael Lynch, D Carolina Penalva-Arana
Indiana University/Biology Bloomington, IN, USA

With a recently sequenced genome, the microcrustacean *Daphnia pulex* has become a model organism in evolutionary and population biology. *Daphnia* rely on gustation to detect basic environmental cues involved in feeding, reproduction, and predator avoidance. In this study, we sequenced one unbiased and several sex-biased gustatory receptor (GR) genes in multiple sexual and asexual *Daphnia* populations to understand population level variation and selection on sex-biased genes in lineages with distinct breeding systems. Of the sequenced GR genes, one male-biased GR has shown distinct phylogenetic clustering of asexual and sexual clones. These male-biased GR genes experience relaxed purifying selection in asexuals compared to the sexuals, while female-biased and unbiased GR genes have similar selection coefficients. We have found a protein altering two-nucleotide indel in *DapuGR28* specific to a handful of populations dispersed throughout North America. We are determining the functionality of the final gene product and the alternative splice version by sequencing cDNA from representative lines. We hypothesize that male-biased GR genes will experience relaxed purifying selection in asexual populations where males are not produced. Female-biased GR genes and unbiased GR genes are expected to experience similar selection coefficients in asexual and sexual populations. Our research will elucidate the variation in GR genes across *D. pulex* populations but also reveal selection pressures undergone by populations having different modes of sexual reproduction. Acknowledgements: NIH GM871342

#P312 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Gα genes expressed in fish taste receptor cells

Makoto Ohmoto[1], Shinji Okada[1], Shugo Nakamura[2], Keiko Abe[1], Ichiro Matsumoto[1,3]
[1]Department of Applied Biological Chemistry, The University of Tokyo Tokyo, Japan, [2]Department of Biotechnology, The University of Tokyo Tokyo, Japan, [3]Monell Chemical Senses Center Philadelphia, PA, USA

Teleost fish and mammals share common mechanisms in the peripheral gustatory system, while they have diverged independently during the course of evolution of vertebrates. T1Rs and T2Rs function as receptors for amino acids and so-called bitter substances, respectively, and the cells expressing these G protein-coupled receptors (GPCRs) have common signaling molecules including PLCβ- 2 and TRPM5, which are crucial for the reception of sweet, umami, and bitter tastes in mammals. Taking into consideration that the molecular features and cellular functions of taste receptor cells (TRCs) are conserved well between teleost fish and mammals, it is reasonable to hypothesize that gustducin ortholog, which is also important for sweet, umami, and bitter taste reception in mammals, is expressed in the TRCs in teleost fish. However, no gustducin ortholog was found in the public database of teleost genomes, and zfGia and zfG14 were expressed in zebrafish TRCs. Their expression was mutually exclusive and consequently divided zfPLCβ- 2-expressing TRCs into two subsets. Interestingly, known taste receptors were expressed only in a subset of zfGia-expressing TRCs. Identification of mutually exclusive expression of Gα subunits and biased expression of known taste receptors in zebrafish indicates that the PLCβ- 2-expressing TRCs were more diversified in teleost fish than in mammals, and suggests that unidentified GPCRs would be expressed in PLCβ- 2-expressing TRCs in fish and function as taste receptors. In silico comprehensive searches for T1Rs and T2Rs in teleost fish genomes conducted by several research groups including our group have revealed almost all T1R and T2R genes. The unidentified GPCRs other than T1Rs and T2Rs would be expressed in PLCβ- 2-expressing TRCs in fish. Acknowledgements: This work was supported in part by Grants-in-Aid for Scientific Research from the Ministry of Education, Culture, Sports, Science, and Technology of Japan.

#P313 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Characterization of TNF-α and Its Receptor Expression in Specific Taste Cell Populations

Pu Feng, Agnes Kim, Daniel Sauers, Hong Wang
Monell Chemical Senses Center Philadelphia, PA, USA

A healthy taste system is important for the maintenance of proper nutrition and overall quality of life, and inflammation has been associated with abnormal taste function. We have previously determined the distribution of immune cells in normal human gustatory tissues and activation of the interferon signaling pathways in the murine taste epithelium challenged with inflammatory stimuli. To further elucidate the roles of inflammatory cytokines in taste tissues, we studied the expression of several cytokines in specific taste cell populations using immunohistochemistry. We found that a variety of cytokines, such as IL-6, IFN-γ, and TNF-α, were expressed by different types of taste cells in circumvallate and foliate papillae. Here we report the presence and distribution of TNF-α and its receptors in taste cells. We found TNF-α was preferentially expressed in type-II taste cells determined by PLC-β2 immunostaining, but was not detected in type-III cells determined by NCAM and SNAP-25 staining. However, one of its receptors, TNFR2, was expressed in a wide spectrum of taste cells including type-II and type-III cells. This expression pattern of TNF-α and TNFR in the taste tissue strongly suggests that TNF-α produced by type-II taste cells could interact with taste cells through both autocrine and paracrine mechanisms. As no regular TNF-α-producing cells (macrophages, T-cells, and other immune cells) were found in taste buds of normal healthy animals, the high endogenous levels of TNF-α within taste buds might have some important physiological roles, possibly related to turn-over, proliferation, differentiation, and programmed cell death. The roles of this taste cell-produced TNF-α are currently under investigation in acute and chronic inflammatory animal models in our lab.
Acknowledgements: This study is supported by NIH/NIDCD grant DC010012.

#P314 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Acetylcholine's role in peripheral taste

Robin Dando[1], Gennady Dvoryanchikov[1], Stephen D. Roper[1,2]
[1]Department of Physiology and Biophysics, University of Miami Miller School of Medicine Miami, FL, USA, [2]Program in Neuroscience, University of Miami Miller School of Medicine Miami, FL, USA

The expression of acetylcholinesterase, the catabolic enzyme for acetylcholine (ACh) in taste tissue was reported nearly 50 years ago (Rakhawy & Bourne, 1964). Thus, a role for ACh in taste transduction has long been postulated. ACh-evoked Ca^{2+} mobilization in taste buds has been observed (Ogura, 2002), however the nature of ACh's role in taste transduction is yet to be clarified. Muscarinic and nicotinic pathways in taste buds have been proposed. Using Ca^{2+} imaging and ATP biosensors, we find that ACh (1μM) stimulates Ca^{2+} mobilization in mouse circumvallate papillae taste cells. In addition, ACh triggers secretion of the afferent taste neurotransmitter ATP from taste cells. Moreover, taste-evoked Ca^{2+} signals and neurotransmitter secretion are augmented by ACh, suggesting that ACh acts as a positive neuromodulator in the taste bud. Interestingly, atropine (5μM) reduces taste-evoked ATP secretion. This suggests that taste stimulation elicits concurrent secretion of ATP and ACh, with ACh acting as an autocrine positive feedback signal. Indeed, using ACh biosensors we find that taste buds secrete ACh during taste stimulation. Finally, genetic deletion of M1 and M3 ACh receptors (M1/M3 knockout), two receptors previously implicated in taste tissue, removes any atropine sensitivity to taste responses. There is also a general reduction of taste-evoked ATP release in knockout animals. Collectively, our findings suggest an endogenous neuromodulatory role (positive autocrine feedback) for ACh in peripheral taste.
Acknowledgements: Supported by NIH/NIDCD grants 5R01DC000374 and 5R01DC007630 to SDR.

#P315 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Characterization of Voltage-gated Calcium Channels in Mouse Taste Receptor Cells

Michelle R Rebello, Kathryn F Medler
Dept of Biological Sciences, University at Buffalo Buffalo, NY, USA

There are functionally different populations of mammalian taste receptor cells that use distinct mechanisms to generate evoked-calcium signals. Type II taste cells depend on calcium release from internal stores while Type III taste cells use calcium influx through voltage gated calcium channels (VGCCs). There is also a population of dual-responsive taste cells that express VGCCs and respond to bitter, sweet or umami taste stimuli by releasing calcium from internal stores (Hacker et al, 2008). Currently, the expression patterns of different VGCC isoforms are not well characterized for mouse taste receptor cells. Several isoforms have been molecularly identified (DeFazio et al. 2006) and Roberts et al. (2009) reported that distinct sub-populations of Type III cells express different VGCCs. We have also found that within the populations of taste cells expressing VGCCs, some VGCCs functionally associate with ryanodine receptors while others do not (Rebello and Medler 2010). These studies indicate a significant variability in the expression of the VGCCs within taste receptor cells. The goal of this study is to characterize the functional diversity of VGCCs in mouse taste cells. Using calcium imaging and pharmacological blockers, we are characterizing the VGCC isoforms and their functional contribution to the calcium influx signal. We find that the calcium influx signals in taste cells are due to the presence of multiple VGCCs and their varied distribution contributes to the functional diversity of taste cells.
Acknowledgements: This work is supported by NSF 0917893 to KM.

#P316 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Imaging cAMP changes in taste cells using a FRET-based reporter

Mani V Kurian[1], Nirupa Chaudhari[1,2]
[1]Department of Physiology and Biophysics, University of Miami Miller School of Medicine Miami, FL, USA, [2]Program in Neurosciences, University of Miami Miller School of Medicine Miami, FL, USA

The second messenger, cAMP, is implicated in taste transduction. There is also evidence of cross talk between Ca^{2+} and cAMP signaling pathways in taste cells. To understand the significance of cAMP for taste signaling, we used a Fluorescence Resonance Energy Transfer (FRET)-based imaging method to examine changes in cAMP levels in real-time and with cellular resolution. We used a genetically-encoded cAMP reporter with Protein Kinase A subunits fused to fluorescent proteins, CFP and YFP. To express the reporter in taste cells, we used two approaches - transgenic and biolistic transfection. First, we generated a transgenic mouse in which the cAMP reporter is inducible in the presence of doxycycline and tetracycline-transactivator (rtTA). Although the cAMP reporter was expressed in taste cells, its titer was insufficient for functional imaging. Second,we biolistically transfected *ex-vivo* slice cultures of mouse vallate and foliate papillae and palate epithelium. 16-24 hours later, the cAMP reporter was expressed robustly in many taste and non-taste epithelial cells. To assess the functionality of the reporter, we exposed the tissue to 20 µM forskolin, an adenylate cyclase activator, and/or to 100 µM IBMX, a phosphodiesterase (PDE) inhibitor. Of all transfected cells in taste epithelia, 17% (28 of 166) responded with an elevation of cAMP. This is consistent with the presence of adenylate cyclases in only a subset of cells in these epithelia. Further, we found that 2% (3 of 166) of all transfected cells responded to a bitter taste mix (10 µM cycloheximide + 1 mM denatonium) with a decrease in cAMP levels, and this response was blocked by IBMX, suggesting that bitter may activate PDEs. We are exploring the taste cell types and potential cross-talk for these cAMP responses. Acknowledgements: Supported by NIH/NIDCD R01DC6021 (NC)

#P317 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Overexpression of recombinant gurmarin secreted by the yeast *Pichia pastoris*

Maud Sigoillot, Nicolas Poirier, Loïc Briand
Centre des Sciences du Goût et de l'Alimentation, UMR6265 CNRS, UMR1324 INRA, Université de Bourgogne Dijon, France

Gurmarin is a polypeptide (35 amino acid residues) known to selectively inhibit responses to sweet substances in rodents without affecting responses to other basic taste stimuli, such as NaCl, HCl, and quinine. Recent studies have proposed that gurmarin may interact with T1R2/T1R3 sweet taste receptor. To further understand the molecular basis of gurmarin inhibition, a large amount of purified gurmarin is suitable for biochemical and structural studies. Here we report the heterologous expression of gurmarin using the methylotrophic yeast *Pichia pastoris*. Gurmarin was secreted into the extracellular medium using the alpha-factor preprosequence peptide of *Saccharomyces cerevisiae*. We found that gurmarin regularly accumulated in the culture medium reaching 50 mg per liter of culture over an expression period of 4 days. The recombinant gurmarin was purified using reversed phase HPLC followed by a cation-exchange chromatography. The molecular mass of the purified gurmarin was shown by MALDI-TOF mass spectrometry to correspond to that expected for fully reduced gurmarin. Circular dichroism and 1D NMR spectroscopy revealed that recombinant gurmarin is properly folded and has secondary and tertiary structures. The efficient production of recombinant gurmarin in *Pichia pastoris* should allow mutational analysis and labeling of sweetness-suppressing peptide with stable isotopes for NMR investigations. These approaches will be useful to study interactions between gurmarin and rodent sweet taste receptor. Acknowledgements: This work was supported by INRA and Burgundy council (Région Bourgogne) grant to M.S.

#P318 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Microwave Processing of Gustatory Tissues for Immunocytochemistry

Amanda E. Bond[1,2], John C. Kinnamon[1,2]
[1]University of Denver/Department of Biological Sciences Denver, CO, USA, [2]Rocky Mountain Taste & Smell Center Aurora, CO, USA

In our laboratory we use immunohistochemistry to study sensory transduction pathways in taste cells. We are attempting to elucidate how taste receptor cells communicate with nerve fibers and/or adjacent taste cells. Previously, we have used conventional immunohistochemistry techniques for our studies. Recently, we have begun to use microwave fixation and processing for taste buds on rat circumvallate papillae. Using the Ted Pella Biowave Microwave Processor we compared classical immunohistochemical tissue processing with microwave processing for the colocalization of several biochemical pathway markers and the nuclear stain, Sytox. The results of our study indicate that microwave fixation is better than conventional immunohistochemistry in terms of ease and speed of the procedures as well as the quality of the results. Microwave tissue processing reduces the amount of time for an experiment from three days to less than one day. In sum, microwave tissue processing of gustatory tissues is faster and superior to conventional immunohistochemical tissue processing. Acknowledgements: NIH DC000285

#P319 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

High-throughput tests of variation in olfactory behavior in a natural population of *Drosophila melanogaster*

Elizabeth B. Brown, Stephanie M. Rollmann, John E. Layne
University of Cincinnati Cincinnati, OH, USA

Olfactory signals are used by many animals to gain information about their environment. Individual variation in behavioral responses to these cues are influenced both by numerous genetic and environmental factors. We have designed a high-throughput behavioral assay system that differs from traditional olfactory behavioral assays in that it allows for fine scale assessment of temporal and spatial location of Drosophila in response to chemical cues. Video tracking revealed significant variation in responses to 2,3 butanedione (a volatile compound present in fermenting fruit) between 162 inbred Drosophila melanogaster lines from a single natural population. These responses were distinct from general activity patterns. These results next allow for studies of the specific genetic mechanisms underlying behavior, and how sequence variants that arise during the evolution of these genes can contribute to individual variation in behavior and give rise to subtle shifts in olfactory perception.

#P320 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Variation in Odor Sensitivity and Behavior in Response to Host Shift in Drosophila mojavensis

Priya Date, Alicia Schwieterman, John E. Layne, Stephanie M. Rollmann
University of Cincinnati. Department of Biology Cincinnati, OH, USA

Olfactory perception is a primary means by which many animals gain information about their surrounding environment, for instance recognizing appropriate food and oviposition sites or mediating social interactions. Divergence in olfactory preferences for host plants may result in differential adaptation of populations to alternative habitats. Here, we examine the evolution of the olfactory system among cactophilic *Drosophila mojavensis* populations that oviposit on different host plants in different regions of the species' range. The volatiles released from fermenting cactus rots are cues by which flies navigate to their appropriate host cacti. Electrophysiological evidence demonstrates significant inter-population differences in sensitivity to four odorants present in fermenting cacti. Olfactory trap assays revealed significant variation in behavioral responses to the same four odorants in response to low odorant concentrations. Specifically, flies showed differential attraction to acids, which may be a key cue in distinguishing different cacti. These results demonstrate that changes in peripheral olfactory system mediate preference for different host plants among populations and may represent host specific adaptation. Acknowledgements: Wieman/Wendel/Benedcit Award

#P321 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Blend blindness during pheromone orientation in the European corn borer

Zsolt Karpati[1], Marco Tasin[2], Charoula Christopoulou[3], Teun Dekker[3]
[1]Max Planck Institute for Chemical Ecology Department of Evolutionary Neuroethology Jena, Germany, [2]Research and Innovation Centre, Edmund Mach Foundation-IASMA S. Michele all'Adige, Italy, [3]Department of Plant Protection Biology, Swedish University of Agricultural Sciences Alnarp, Sweden

The European corn borer with two pheromone strains (Z, E), is a good model species of reproductive isolation based on chemosensation. The Z-strain males respond to a ratio of 97:3 of Z11-tetradecenyl acetate and E11-tetradecenyl acetate, while the E-strain prefers almost the opposite ratio (1:99). Male pheromone preference is a major factor contributing to reproductive isolation of the two strains as wind tunnel studies demonstrate that cross-strain attraction is nearly zero. Yet, hybrids can be found where the two strains occur in sympatry, which is surprising considering male blend selectivity. We conjectured that in sympatry a male moth may be confronted with overlapping plumes of different composition, a situation we simulated in the wind tunnel. We found that the specificity of the males' decreases when plumes of their own strain are interlaced with other plumes, in other words they land on the other strain's source. The experiments also indicate that selectivity differs between the various behavioural steps leading to source finding, i.e., males are not equally sensitive to the blend ratios in the different stages in the orientation process. Detailed flight analyses with swapping pheromone plumes demonstrate that males may not be as sensitive to the blend ratio once locked onto the plume. Taken together the results indicate that in the field hybridization rates increase as a function of population density. Possible causes of the observed 'blend blindness' are discussed. Acknowledgements: Acknowledgements: This project was supported by Carl Trygger Stiftelse CTS 06:94 and 07:77 grants, the ICE3 Linnaeus grant to the division of Chemical Ecology, FORMAS grant 2007-1491, and a Marie Curie IEF (255193) fellowship.

#P322 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Concentrated sodium chloride solutions stimulate feeding in *Anopheles gambiae* mosquitoes

Jae Kwak[1], Nuwar Ahmed[1], Natasha Rivers[1], Paul A. S. Breslin[1,2]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]Rutgers University/Nutritional Sciences New Brunswick, NJ, USA

Sodium chloride is an essential micro-nutrient for both vertebrate and invertebrate physiology. In general, animals are attracted to NaCl at low concentrations, but avoid it at moderate to high concentrations (> 150mM). The concentration-dependent acceptance/rejection behavior is attributed to the salty taste and the differential receptor activation. Unexpectedly, we observed that *Anopheles gambiae*, the major vector of malaria transmission, found NaCl appetitive at high concentrations. To confirm and extend our preliminary observations, we compared sucrose intake

in *Drosophila melanogaster* and in *Anopheles gambiae* while stimulating the tarsal taste receptors with either vehicle (water) or NaCl solutions. *D. melanogaster* ingested more sucrose when their tarsi were stimulated with water than when stimulated with either 100 or 200 mM NaCl, whereas *A. gambiae* ingested more when their tarsi were stimulated with the hypertonic NaCl. Furthermore, about 80% of the tested mosquitoes consumed sucrose while their tarsi were stimulated with 1M NaCl. This suggests that *Anopheles gambiae* find high salt appetitive, which distinguish them from other omnivores, including *Drosophila*. Attraction to the taste of high salt concentrations on skin would be advantageous for *Anopheles gambiae*, an anthropophilic, tropical species, since it is normally exposed to evaporated human sweat on skin while taking blood meals. Acknowledgements: Funded in part by a grant from the Gates Foundation, Grand Challenges Explorations to PASB.

#P323 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Insect Chemical Defense Compounds as Mammalian Irritants

Paige M Richards, Wayne L Silver
Wake Forest University, Department of Biology Winston-Salem, NC, USA

Insects release a variety of chemicals to avoid predation or protect their territory. It was recently proposed that these defensive compounds may have been selected to elicit vertebrate chemesthesis (stimulation of the somatosensory system by chemical irritants). In the current study, we are testing a number of insect defense compounds (including 6-methyl-5-hepten-2-one, benzoquinone, tetradecane, trans-2-hexenal, and trans 2-hexen-1-ol) to determine if they are irritating to rats. In mammals, nasal and facial chemesthesis is primarily mediated by the trigeminal nerve. We determined if the tested stimuli activated the trigeminal nerve by recording from the ethmoid nerve, a sub-branch of the trigeminal nerve, when stimuli were perfused through the rat's nasal cavity. As activation of the trigeminal nerve is known to decrease respiratory frequency, we also monitored respiration in the rats both before and after stimuli were introduced. By combining these two methods, we determined that 6-methyl-5-hepten-2-one, benzoquinone, trans-2-hexenal, and trans 2-hexen-1-ol do elicit chemesthesis by activating the trigeminal nerve. Exposure to tetradecane did not result in activation of the ethmoid branch of the trigeminal nerve nor did exposure elicit respiratory depression in rats. Our next step is to determine the receptor targets of these compounds using calcium imaging of both primary trigeminal neuron cultures and heterologous expression systems. Additional compounds will be tested in the future. Acknowledgements: Institutional

#P324 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

The Effect of Odor on Reaction Time

Gurprit S. Bains[1], Alan R. Hirsch[1], Sally Freels[2], Syed A. Hassan[1], Navdeep Lail[1], Sunith Vijaykumar[1], Hossameddin Keshlaf[1]
[1]Smell & Taste Treatment & Research Foundation Chicago, IL, USA, [2]University of Illinois at Chicago School of Public Heatlth Chicago, IL, USA

Objective: Contrary to Millot et al. (2002) findings that odors, independent of hedonics reduced reaction time, we hypothesize that this response would be hedonically dependent.
Methods: Twenty one subjects, ages 18-61, chosen using a convenience paradigm, participated in this IRB approved study. All underwent the QSIT and 3 epochs of 8 trials of reaction time testing, using the Eckner (2010) method whereby a Queen Square Reflex Hammer like device is dropped and the distance before grasping is measured. Average reaction time (ms) = 1000*sqrt(2*distance/980). Each subject served as their own control while wearing unodorized 3M Aseptex surgical masks as compared to masks impregnated with jasmine (Custom Essence) or charcoal mesquite meat (IFF) odor. Order of mask was presented randomly. Effects of order, hedonics, gender, and odor were assessed and independently analyzed using ANOVA.
Results: When exposed to jasmine, reaction time (207.3 ms) was faster than blank mask (220.9 ms) (p=.0014) as well as pleasant odors (211.4 ms) compared to blank mask (223.9 ms) (p=.0015). Effect of Reaction time when exposed to charcoal mesquite meat odor (227.7 ms) (p=.2752) and unpleasant odors (230.8 ms) (p=.2138) is not significant. An effect of order of presentation was demonstrated with each series of presentations from 226.6 ms to 218.3 ms to 211.1 ms, with a significant difference from the 1st to the 3rd series (p=.0123). Men had a faster reaction time than women (213.9 ms vs. 230.6 ms) (p=.0478), but the pattern of response to different odors was the same for both genders (p=.8554). **Conclusion:** Jasmine may be useful in enhancing athletic performance and facilitating physical therapy and rehabilitation. Further investigation is warranted.
Acknowledgements: none

#P325 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Manipulating the Consolidation of Odor-Cued Memories during Sleep

Katherina K. Y. Hauner, Phyllis C. Zee, Jay A. Gottfried
Northwestern University Chicago, IL, USA

Recent research has demonstrated that newly learned episodic memories can be selectively enhanced during sleep, via the re-delivery of night-time olfactory cues that were introduced during initial daytime learning (Rasch et al., 2007). The present study examined whether non-episodic, emotional memories could also be selectively enhanced using similar methods. Specifically, olfactory contextual cues were used to enhance the consolidation of classically conditioned fear memories. Critically, the decision to use odor stimuli as memory modulators was based on the intimate anatomical and functional connections between olfactory and limbic systems—olfactory sensory neurons are only two synapses

removed from brain areas involved in emotion and learning. Moreover, there is recent evidence to suggest that odors have privileged access to brain networks during sleep, and that episodic memories can be enhanced by re-presenting the same odor cues during sleep that were originally presented during daytime learning. As hypothesized, results indicated that odor cues originally paired with daytime fear learning (and re-presented during sleep) significantly changed fear responses after initial training, as compared to odor cues that had also been paired with fear learning but not re-presented during sleep. More generally, these results also support the use of odor as both a contextual cue for fear learning (indicated by odor-specific fear responses) as well as a practical, unobtrusive night-time stimulus (indicated by participants' failure to awaken during nighttime odor delivery). Follow-up fMRI research examining the neural correlates of these processes is currently underway and will also be discussed. Acknowledgements: The research in this presentation is supported by the National Institutes of Health [NICDC 1 R01 DC 010014, NINDS 5 T32 NS 047987].

#P326 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Chemosensory induced arousals during human sleep - effects of a pure olfactory stimulus and artificial smoke

Boris A. Stuck[1], Franziska Lenz[1], Jann Baja[1], J. Ulrich Sommer[1], Raphael M. Herr[2], Clemens Heiser[1]
[1]University Hospital Mannheim, Department of Otorhinolaryngology Mannheim, Germany, [2]Mannheim Institute of Public Health, Social and Preventive Medicine Mannheim, Germany

Study Objectives: The central processing of olfactory stimuli is different compared to other sensory systems. Negatively engaged olfactory stimuli are followed by no arousals in women. It is still unclear whether a positive engaged olfactory stimulus or stimuli with high significance or stimulation of male subjects increase arousal frequency. Methods: 11 young healthy normosmic volunteers of both sexes were included in this prospective controlled trial. Intranasal chemosensory stimulation was performed during sleep with an olfactometer. PEA (phenylethylalcohol, a pleasant smell like rose) in 3 concentrations (10%, 20% and 40% v/v) and an artificial smoke (0.05% v/v) using different stimulus durations were used and compared to odorless control stimulation. Arousal reactions in relation to chemosensory stimulation during sleep were assessed with the help of overnight sleep recordings during 22 nights of testing. Results: Stimulation with PEA in both sexes did not lead to an increase in arousal frequency compared to odorless control, even in the highest concentrations. There was only a tendency for an increase in arousal frequency with artificial smoke when using long stimulation periods, not reaching statistical significance. Conclusions: Positive olfactory stimuli do not lead to arousals in both sexes, which is accordance to the results of a negative stimulus used in women. Even a strong and significant olfactory stimulus such as smoke does not seem to elicit arousals, which could be relevant with regard to house burns. Overall, the results confirm the exclusive role of the olfactory system in sensory physiology. Acknowledgements: The study was supported by a grand of the German Research Foundation (Deutsche Forschungsgemeinschaft STU 488/2-1)

#P327 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Behavioral benefits and cortical responses to multisensory stimulation

Eva C. Alden[1], Jessica Albrecht[1,2], Valentin A. Schriever[3], Johan N. Lundstrom[1,4,5]
[1]Monell Chemical Senses Center, Cognitive Neuroimaging Laboratory Philadelphia, PA, USA, [2]RWTH Aachen, Department of Diagnostic and Interventional Neuroradiology Aachen, Germany, [3]University of Gottingen, Department of Neurophysiology and Cellular Biophysics Goettingen, Germany, [4]University of Pennsylvania, Department of Psychology Philadelphia, PA, USA, [5]Karolinska Institute, Department of Clinical Neuroscience Stockholm, Sweden

Studying one sensory modality at a time is insufficient to further our understanding of our everyday perceptual experience, since we rarely experience pure unimodal sensory stimuli in isolation. To assess the benefits of multisensory stimulation, behavioral responses and event-related potentials (ERP) were measured in two separate studies. We used olfactory, auditory, and visual cues originating from common semantic objects to create unimodal auditory (A) and visual (V) stimuli, bimodal olfactory-auditory (OA), olfactory-visual (OV), auditory-visual (AV), and trimodal olfactory-visual-auditory (OVA) stimulus combinations. Two distinct olfactory stimuli were used, a pleasant coffee odor and an unpleasant fish odor. To enhance the effects of multisensory stimulation, weak auditory and visual stimuli were used. Masked videos and sounds were set to an 80% detection rate on an individual level. In Study 1 we measured reaction time to the various masked sensory stimulus combinations. Participants were instructed to respond as quickly as possible when they identified the object. The more sensory modalities stimulated, the faster and more accurate participants identified the objects. In Study 2, participants were shown the same masked stimuli and asked to identify the stimulus presented, while we recorded electrocortical responses using a 32-channel active-electrode EEG system. Data from Study 2 are currently being analyzed and will be presented. These data support the central tenet of sensory integration, which states that multisensory stimulation facilitates object identification. Acknowledgements: Supported by start-up funds from the Monell Chemical Senses Center awarded to JNL and a DAAD postdoctoral fellowship D/08/40252 awarded to JA.

#P328 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Effects of Odorant Administration on Consumer Product Selection and Expected Value

Sarah Mogan, Megan Foutty, Bryan Raudenbush
Wheeling Jesuit University Wheeling, WV, USA

Prior research has shown the effectiveness of product-scent matching on influencing consumer purchases and perceived value of in-store products. However, there is no evidence to support whether this effect can also be extended to online shopping. In the present study, scents were matched to three consumer products in a slideshow of ten products. The scents included Froot Loops (Froot Loops cereal), leather (a leather coat), and coffee (coffee beans). Participants were randomly placed in either the control

group (no scent) or one of the three scent conditions, and asked to rate their desire to purchase each product, the dollar amount they would be willing to pay for each product, and their perceived quality of each product. No significant difference was found in any of the three scent groups, Froot Loops, leather, or coffee. This implies that participants were not influenced by the scent of the product in accordance with their desire to purchase, the amount of money they were willing pay, or their perceived quality of the product. These results are contrary to past research; however, there are a number of differences between this and similarly conducted studies, primarily with this being based "on-line" rather than "in-store." Thus, there is an obscure component about having a scent paired with products in-store that does not translate to on-line shopping.

#P329 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Effects of Peppermint Scent Administration on Cognitive Video Game Performance: A Physiological Explanation

Mark Sappington, Kristin McCombs, Andrea Bova, Bryan Raudenbush
Wheeling Jesuit University Wheeling, WV, USA

Past research has shown the positive effects of video game play while other research has shown the benefits of peppermint scent administration. The present study assessed the combination of video game play and peppermint scent administration on video game performance related to cognition, physiology, mood, and task load. Participants completed a baseline control condition and were then assigned to either repeat the control session or to participate in an experimental condition in which peppermint scent was delivered via nasal cannula at 3LPM. Participants played 3 Nintendo Wii Fit Plus games requiring cognitive and hand/eye reactions (Perfect 10, Snowball Fight, and Obstacle Course). Participants in the peppermint scent condition showed greater improvements, such as completing significantly more levels [t(14)=-2.95,p=.01], more hits [t(14)=-4.03,p=.001] and stars [t(14)=-4.00, p=.012], and distance completed [t(14)=-1.97, p=.08]. Further, participants in the peppermint scent condition reported decreased mental demand [t(14)=1.96, p=.070], perceived effort [t(14)=2.27, p=.039], and anxiety [t(14)=2.39, p=.031]. In terms of physiological data, control group participants had a significantly lower pulse change [t(15)=2.246, p=.04] and diastolic blood pressure change [t(15)=12.13, p=. 069]; whereas, participants in the peppermint scent condition experienced no significant difference in pulse, suggesting that the scent administration promoted greater physiological arousal, thus keeping participants more engaged in the testing process. Implications include the combination of video games and a physiologically arousing scent (specifically, peppermint) to further promote cognitive performance.

#P330 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

On the quest for a common chemical sensitivity

Johan N Lundstrom[1,2,3], Amy R Gordon[1,2], Johannes Frasnelli[4]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]University of Pennsylvania / Dept. of Psychology Philadelphia, PA, USA, [3]Karolinska Institutet / Dept of Clinical Neuroscience Stockholm, Sweden, [4]Université de Montréal / CERNEC Montreal, QC, Canada

The three chemical senses exhibit both differences, such as receptive mechanisms and physical location of the receptive structures, and common properties, such as chemical structure and neural processing sites. Based on their integrative function in flavor processing, we hypothesized that there would be shared properties in sensitivity among the senses. To determine potential commonalities between the detection thresholds which might indicate a general chemical sensitivity variable, we performed an unbiased and exploratory principal component analysis on five z-transformed, 16-step detection thresholds measured in 70 women: two olfactory thresholds (n-butanol, peanut oil), two gustatory thresholds (sucrose, quinine), and one intranasal trigeminal detection threshold (l-menthol). Extracting only the components loading highest (more than the mean eigenvalue) rendered a two-component solution where Component 1 accounted for 29.4% and Component 2 accounted for 26.9% of the total variance. The two oral detection threshold tests loaded high on Component 1 whereas the three intranasal detection threshold tests all loaded high on Component 2, indicating a nose-mouth axis. However, a subsequent principal component analysis restricting extraction to a three component solution rendered a subdivision into the three chemical senses. Further, Spearman rank correlation tests indicated that there was no correlation between the senses in respect of sensitivity. In conclusion, chemical sensitivity seems to be related to anatomical location (nose and mouth) indicating either that the two tasks are utilizing separate behavior schemata or that there is a general nasal sensitivity and general taste (oral) sensitivity. Further, our data do not support a general, modality independent, chemical sensitivity.

#P331 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Multilevel Analysis of Variance in Taste Sensitivity Scores Across Time in Healthy Older Adults

Emily S. Bower[1], Claire Murphy[1,2]
[1]San Diego State University San Diego, CA, USA, [2]University of California San Diego, CA, USA

Determining a standardized taste sensitivity test with age-appropriate cutoff scores is essential for diagnosing taste sensitivity abnormality. Sucrose taste detection tests are simple and cost-effective, but more information is needed regarding sources of variance in test results. The current study analyzed repeated, annual observations of taste sensitivity in healthy older adults (50-80 years) using an absolute threshold test for sucrose to explore possible sources of variance in this age group. A series of 14 concentrations of sucrose prepared in quarter-log steps (.56 M to .00032 M) were dissolved in distilled water. Participants

were presented with two unmarked polyethylene cups containing 10 ml of a sucrose solution and of distilled water, respectively. A threshold was determined using a forced-choice procedure beginning with the weakest sucrose solution and proceeding to the next strongest concentration until the participant chose correctly 5 times in a row. Hierarchical linear modeling was used to analyze annual observations of taste threshold (Level 1) nested within individuals (Level 2). The intercept-only model revealed that variance in taste threshold existed at both levels, although more variance existed in Level 1. Gender and age, added as predictor variables at Level 2 and Level 1 respectively, did not significantly explain the variance. However, visit number added as a predictor at Level 1 explained 74% of the variance and was positive and statistically significant, indicating that taste thresholds increased with successive observations. This finding may be due to a behavioral component such as attention, thus further research is needed to explore other possible sources of variance at the repeated-measures level. Acknowledgements: Supported by NIH Grant R01 AG04085 to C.M. We thank the members of the UCSD Shiley-Marcos Alzheimer's Disease Research Center (ADRC), and the support of the NIH Grant #P50 AG05131 to the ADRC.

#P332 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Taste Intensity in the Beaver Dam Offspring Study

Mary E Fischer[1], Karen J Cruickshanks[1,2], Carla R Schubert[1], Alex Pinto[1], Guan-Hua Huang[3], Barbara E K Klein[1], Ronald Klein[1], Derek J Snyder[4]
[1]University of Wisconsin, Department of Ophthalmology & Visual Sciences Madison, WI, USA, [2]University of Wisconsin, Department of Population Health Sciences Madison, WI, USA, [3]National Chiao Tung University, Institute of Statistics Hsinchu, Taiwan, [4]San Diego State University, Department of Psychology San Diego, CA, USA

Factors associated with the perception of saltiness, sweetness, sourness, and bitterness were examined in the Beaver Dam Offspring Study. Taste intensity was measured using filter paper disks and a general labeled magnitude scale (gLMS) with a range of 0-100. Subjects were classified as perceiving each taste intensity as strong or greater or less than strong. There were 2374 subjects (mean age=48.8, range=21-84 years) and the percentage perceiving the intensity as strong or greater was: salt-30%, sweet-16%, sour-49%, and bitter-74%. Approximately 22% (n=526) of the subjects perceived only 1 of the 4 tastes as strong or greater while 9% (n=207) perceived all 4 tastes as strong or greater. Of the 480 subjects who perceived all 4 as less than strong, 25% (n=120) rated 6-*n*-propylthiouracil (PROP) intensity as strong or greater. Strong taste intensity was observed significantly more often in older subjects (4 tastes combined, adjusted model with age, sex and education (adj): Odds Ratio(OR)$_{adj}$=1.1, 95% Confidence Interval(CI)=1.0,1.2), females (OR$_{adj}$=2.6, 95% CI=1.8,3.7), the less educated (OR$_{high\ school\ vs\ college\ graduate,adj}$=2.2, 95% CI=1.4,3.4) and those with olfactory impairment (OR$_{adj}$=3.1, 95% CI=1.4,7.1). A perception of strong salt intensity was more common in those with allergies (OR$_{adj}$=1.2, 95% CI=1.0,1.5). Strong bitter intensity was observed more often among those with allergies (OR$_{adj}$=1.3,95% CI=1.1,1.6) and those taking anti-anxiety drugs (OR$_{adj}$=2.2, 95% CI=1.1,4.3). Strong sour intensity was more common among subjects on anti-hyperlipidemics (OR$_{adj}$=1.6, 95% CI=1.2,2.0). Adjustment for perceived PROP intensity did not appreciably alter the results. Age, sex, and education were associated with the 4 tastes individually and combined but associations with other factors differed by tastant. Acknowledgements: The project described was supported by R01AG021917 from the National Institute on Aging, National Eye Institute, and National Institute on Deafness and Other Communication Disorders.

#P333 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Interactions Between Limonin and Nomilin, Two Bitter Compounds of Orange Juice

Sharon Dea, Anne Plotto, Elizabeth A. Baldwin
USDA-ARS Winter Haven, FL, USA

As a preliminary step to understand and characterize which metabolites are responsible for the bitter off-favor of Huanglongbing (HLB) infected fruit, the thresholds of limonin, nomilin, and their combination in a sugar and acid matrix, as well as in healthy 'Valencia' orange juice were determined by taste panels. Food grade limonin and nomilin were added alone or in combination to a simple (sucrose and citric acid) or complex (sucrose, glucose, fructose, citric and malic acid) matrix, or were added directly into orange juice. Thresholds were determined by 18 to 23 trained panelists using a three-alternative forced choice method (ASTM: E-679). In the simple matrix, the threshold of limonin was lower than nomilin. The synergetic effect of limonin and nomilin was significant in decreasing their individual thresholds. Interestingly, the thresholds of limonin and nomilin were lower in orange juice compared to the thresholds measured in the complex matrix. The threshold concentrations of limonin and nomilin when added to healthy 'Valencia' orange juice were higher than the concentrations of those compounds measured in juice made with symptomatic HLB fruit, which was perceived bitter by a taste panel. Possibly, the lower sugar and higher acid content of HLB fruit decreased the threshold of those bitter compounds. Acknowledgements: The Citrus Research and Development Foundation

#P334 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Better Tomatoes Through Psychophysics

Linda M. Bartoshuk[1], Adilia Blandon[1], Peter L. Bliss[1], David G. Clark[1], Thomas A. Colquhoun[1], Harry J. Klee[1], Howard K. Moskowitz[1,2], Charles A. Sims[1], David W. Smith[1], Derek J. Snyder[1,3], Denise M. Tieman[1]
[1]University of Florida Plant Innovation Program Gainesville, FL, USA, [2]Moskowitz Jacobs Inc. White Plains, NY, USA, [3]San Diego State University San Diego, CA,

The primary taste/flavor constituents of tomatoes are sugars, acids, possibly umami and about 30 volatile organic compounds. The concentrations that maximize palatability are not known. Using the gLMS (general Labeled Magnitude Scale) and the hedonic gLMS, subjects (N=137) rated the following in each of 50 varieties of tomatoes: sensory - sweet, sour, salt, bitter, umami, tomato flavor and sensations desired in an "ideal" tomato;

hedonic - liking/disliking, best/worst tomato ever tasted and favorite/least favorite foods. Finally, non-food items emphasized that the scales assess sensory and hedonic experiences of all kinds. Correlations between overall liking and the concentration of each constituent showed the expected positive correlations between sugars and liking. For about half of the volatiles, concentration was positively correlated with liking; concentration of one volatile correlated negatively and the rest did not correlate. For sweet and tomato flavor, the ideal values significantly exceeded the observed values. This suggests one way to improve tomato palatability: remove the volatile with the negative correlation and increase concentrations of volatiles showing positive correlations. Because of links between taste and retronasal olfaction, these increases might serve to intensify perceived sweetness. Since the scales used permit valid comparisons across groups of people, we examined group differences for the ideal tomatoes by gender, race, ability to taste, and weight. Those most fond of tomatoes tended to be supertasters and wanted higher sensory intensities; females wanted more sweetness in their ideal tomatoes. For sweet, sour and tomato flavor, non-white subjects wanted greater intensities, but for umami, they did not. Ideal tomatoes did not differ for obese and thin subjects. Acknowledgements: We thank NIDCD for support via grants DC283, DC8613 and DC8620, the Florida Agricultural Experiment Station and the UF Research Foundation.

#P335 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Toothpaste and Orange Juice: The Teetotalers Artichoke Effect

Alan R. Hirsch[1], Gurprit S. Bains[1], Sally A. Freels[2], Lovpreet S. Mangat[1]
[1]Smell & Taste Treatment & Research Foundation Chicago, IL, USA, [2]University of Illinois at Chicago School of Public Health Chicago, IL, USA

Objectives: The artichoke effect is where the taste of dry wine is sweetened by the previous consumption of artichoke. Bartoshuk (1972) suggests this is due to the potassium salts of cholinergic acid and cynarin; potassium salts inhibiting sweet receptors. A more pedestrian example is seen with the taste of orange juice altered by toothpaste which also contains potassium salts. The effect of temperature on this artichoke effect has not been described. **Methods:** 9 subjects (5 male, 4 female), average age 36 years (range 26-61), on a visual analog scale of sweetness and bitterness, rated unsweetened 100% orange juice at refrigerated and room temperatures and immediately after brushing with Colgate toothpaste for 2 minutes.
Results: Average ratings on a one to ten scale were as follows:

	Before Brushing	After Brushing	Change	Paired t-test (p)
Sweetness/room temperature	5.44	3.78	-1.67	0.0001
Sweetness/refrigerated	4.44	3.56	-0.88	0.05
Bitterness/room temperature	4.44	6.11	+1.67	0.0004
Bitterness/refrigerated	4.56	6.11	+1.56	0.04

Conclusions: The artichoke effect persisted, but with less intensity at lower temperature. This appears to be due to the reduction in baseline sweetness and increase in baseline bitterness when cold, similar to the temperature effect on taste perception of wine (Ross and Weller, 2008). The practical implications of these results may be found in the culinary arts or meal planning in the obese, anorexic, geriatric or diabetic populations.
Acknowledgements: None

#P336 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Age-related Changes in the Bitterness of 6-n-propylthiouracil (PROP) and Food Preferences in an Isolated Population in Northwest Italy

Beverly J Tepper[1], Yvonne Koelliker[1], Carmela Lanzara[2], Nicola Pirastu[2], Paolo Gasparini[2], Cinzia Sala[3], Daniela Toniolo[3]
[1]Rutgers University, Food Science Department New Brunswick, NJ, USA, [2]University of Trieste, Department of Reproductive and Developmental Sciences, IRCCS Burlo Garofolo Trieste, Italy, [3]San Raffaele Scientifirc Institute, Division of Genetics and Cell Biology Milan, Italy

The ability to taste the bitterness of PROP is considered a stable and reliable human trait. Studies have shown that PROP non-tasters have higher preferences for many bitter, strong tasting and fatty foods than tasters. However, most studies have been conducted in young subjects. Thus, data are lacking in aging populations. The objectives of this study were to examine age-related changes in PROP taste intensity and determined if these changes weakened the expected associations between PROP status and food preferences. We studied 589 healthy, adults (18-96 yrs of age; 343 females; 246 males) residing in the isolated community of Val Borbera located in Northwest Italy. Half of the participants were >50 yrs of age. Participants rated the intensity of PROP- and NaCl–impregnated filter papers using the LMS; they were then classified as PROP tasters or non-tasters according to the method of Zhao et al. (2003). We also collected hedonic ratings (9-pt scale) for 66 familiar foods by questionnaire. PROP ratings declined slowly and consistently with age in both genders; NaCl ratings showed a similar pattern of decline (all p-values <0.001). Consistent with previous data, females preferred vegetables, fruits and sweet-fat foods, whereas males preferred alcohol and meats. Among males <50 yrs of age, non-tasters liked bitter vegetables (chicory, cabbage, radicchio and cauliflower) more than tasters ($p<0.05$). In contrast, among females <50 yrs of age, tasters liked gorgonzola and fontina cheeses, ice cream and milk chocolate more than non-tasters ($p<0.05$). After age 50, no PROP-related differences in liking ratings were found. These data suggest that declines in PROP taste intensity may be part of the normal aging process, and that the influence of PROP status on food preferences also diminishes with age.

#P337 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Association Between PROP-Sensitive Taster Status and Health Factors Including Smoking and Body Fat Percentage in Adult Men and Women

Sara M Shanahan[1,2], Nicole L Garneau[2]
[1]University of Denver Denver, CO, USA, [2]Denver Museum of Nature & Science Denver, CO, USA

Little is known about the relationship between phenotypic taster status and health factors including smoking status and percent body fat. Data generated from the study of this relationship could be used to aid people in thinking more critically about their diet and lifestyle factors. We were interested in determining if an association exists between body fat percentages and different phenotypic taster statuses, and to determine the effect of smoking on those phenotypic taster statuses. The data derived for this study was obtained from participants in the Genetics of Taste study going on in the Expedition Health exhibit at the Denver Museum of Nature & Science. The subjects provided information concerning their smoking status as part of a questionnaire, a TANITA body composition scale was used to calculate percent body fat, and finally a 6-n-propylthiouracil taste test was administered. This study revealed a number of results, including a significant difference in percent body fat among those who reported non-taster status and those who reported supertaster status. Also, women who reported themselves to be smokers and were also non-tasters were found to smoke for more years than women who smoked and were tasters. This preliminary data suggests that taster status does play a role in overall health and lifestyle choices. Acknowledgements: R25 RR025066-02

#P338 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Variation in bitter taste perception between moral vegetarians and non-vegetarians

Amy S. Teller[1], Hillary J. Wiener[2], Linda Bartoshuk[3], Susan E. Marino[4]
[1]Carleton College/Environmental Studies Northfield, MN, USA, [2]Carleton College/Psychology Northfield, MN, USA, [3]University of Florida/Smell and Taste Center Gainesville, FL, USA, [4]University of Minnesota/Ctr for Clinical & Cognitive Neuropharmacology Minneapolis, MN, USA

Previous research suggests that PROP non-tasters eat more vegetables than do PROP tasters, but this has not been evaluated in eating subgroups such as vegetarians. The present study compared females who are vegetarian for moral reasons (N=29) to females who are not vegetarians (N=28); subjects were students at a small midwestern college. Subjects completed a survey on vegetable consumption, rated vegetable bitterness intensity (gLMS), and rated liking/disliking of a variety of foods as well as selected non-food experiences (hedonic gLMS). As expected, the vegetarians consumed significantly more vegetables than did the non-vegetarians. PROP papers were used to determine taster status. Vegetarians rated PROP as significantly less bitter than did non-vegetarians (37 and 60, respectively). Given this difference, it is perhaps not surprising that both the average bitterness of all of the vegetables surveyed and the "most bitter vegetable ever tasted" were significantly less bitter to the vegetarians. This suggests that vegetarians' increased vegetable consumption may not just be due to their lack of other eating options but also because vegetables taste less bitter to them. However, this relationship did not appear to be mediated by liking. Hedonic ratings of food items and nonfood affective experiences did not differ for the two groups with two exceptions: steak and seeing a snake. Vegetarians reported significantly less liking for steak than did meat eaters, but unlike non-vegetarians, they showed mild liking for seeing a snake. Despite the lack of hedonic differences between vegetarian and non-vegetarians, the differences in perception of bitterness may partially explain why some people become vegetarians and others choose to express their environmental or moral concerns in other ways. Acknowledgements: none

#P339 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

***sAC* SNP Associates with Human Sweet, Bitter and Umami Sensations and Hedonics**

Shristi Rawal[1], Margaret R Wallace[2], Linda M Bartoshuk[3], Valerie B Duffy[1]
[1]Allied Health Sciences, University of Connecticut Storrs, CT, USA, [2]Molecular Genetics and Microbiology, University of Florida Gainesville, FL, USA, [3]Community Dentistry, University of Florida Gainesville, FL, USA

Adenylate Cyclase (AC) enzymes are important in generating and regulating intracellular '-5'-cyclic adenosine monophosphate (cAMP), the second messenger with suggested role in taste transduction. Changes in cAMP levels are associated with sweet, bitter, sour and umami stimulation. AC has high activity in chemosensory systems and various isoforms are expressed in taste cells. AC activity also increases in response to calcium, another second messenger involved in taste transduction. The gene for AC isoform 10 (soluble form, sAC) resides on human chromosome 1 and is expressed in multiple tissues (e.g., brain, lung, GI tract). We tested if *sAC* gene was associated with human variation in taste perception. DNA samples from 92 healthy adults were isolated from whole bloods and genotyped for *sAC* SNP (rs2071921) by TaqMan. Our genotype frequencies (18% homozygous for C allele, 47% heterozygous, 35% homozygous for T allele) were similar to reference frequencies for European-Americans (22%, 47%, and 32%, respectively). Using the sensory and hedonic forms of the general Labeled Magnitude Scale, subjects rated the intensity of: liking and taste quality of sampled foods/beverages, prototypical tastants swallowed with the whole mouth. Controlling for age, sex and intensity of tones as a cross-modal standard, TT adults reported greater intensity of 1 M sucrose and 1 mM quinine, greater saltiness from soy sauce and concurrent reduction in liking, greater sweetness and concurrent greater liking for milk chocolate, and greater liking of sampled bitter vegetables. The general effect size was significant, but not large. These results support a minor role of the *sAC* in response to sweet, bitter and possibly glutamate taste stimuli in solution and food systems in humans. Acknowledgements: Funded by USDA/Hatch and NIH DC008613

#P340 POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR

Effects of *hTAS2R38* Haplotype on Medicine Usage and Food Behaviors in Children

Sarah V. Lipchock, *Danielle R. Reed, Julie A. Mennella*
Monell Chemical Sense Center Philadelphia, PA, USA

Some children refuse to take bitter medicines or eat a variety of foods, whereas others comply readily. Likewise, not every person is equally sensitive to the taste of bitter compounds. The substantial degree of DNA sequence diversity present in taste genes may underlie individual differences in the behaviors of children due to taste. Although individual differences arise for a variety of reasons (e.g., age, experience, ethnicity), genetics play an essential role. To this end, we recruited a genetically diverse, large urban sample of children (N=472) ranging in age from 3 to 10 years. Genomic DNA was extracted from cheek cells and the A49P, V262A and I292V alleles of the TAS2R38 bitter receptor gene were genotyped using allele-specific probes and primers. Children and their mothers were asked about medication use and food habits. Children who were 3-5 years of age were significantly less likely to have taken a pill than those who were 5-10. However, for children who were homozygous for the taster alleles (PAV/PAV), those under the age of 5 were just as likely to have taken a pill as those who were 5-10 years of age. The aversion to bitter taste may be a motivating factor for children to find an alternative to liquid formulations. Children who were homozygous for the taster alleles (PAV/PAV) were also less likely to list a cruciferous vegetable as one of their favorites as compared to children of other genotypes. Cruciferous vegetables, like broccoli and collard greens, contain compounds that are specifically recognized by the TAS2R38 bitter receptor, so children who are homozygous for the taster alleles may find these vegetables much more unpleasant than those of other genotypes. Our studies indicate that genotype plays an important role in the behaviors of children that are affected by taste. Acknowledgements: This project was funded by NIH Grant HD 37119 and a grant from the Pennsylvania Department of Health. The Pennsylvania Department of Health specifically disclaims responsibility for any analyses, interpretations, or conclusions. Dr. Lipchock is a postdoctoral trainee on NIH grant T32-DC00014.

#P341 POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR

How the Sensory World of Children Differs from Adults: Sweets, Salt and Fat

Stacie S. Miller, *Susanna Finkbeiner, Aleida Silva-Garcia, Danielle R. Reed, Julie A. Mennella*
Monell Chemical Senses Center Philadelphia, PA, USA

Although modernization and industrialization of the food supply has produced many benefits, unanticipated consequences from eating diets rich in sugars, salt and fats have become increasingly commonplace. Thus, health organizations worldwide recommend that both adults and children limit their intake of salt and simple sugars. This recommendation may be particularly difficult for pediatric populations since findings from basic research strongly suggest that children prefer more intense sweet or salty tastes than adults. However, there is a paucity of information on the relationship between sweet and salty taste preferences during childhood and adulthood and whether these preferences relate, if at all, to the liking of creaminess in foods. To this end, we phenotyped 162, 5- to 10-year-old children and their mothers for the intensity of sucrose most preferred in water; of those 162 dyads, 84 were also phenotyped for preferences for creaminess and sweetness in pudding, and 99 were phenotyped for preferences for salt in a soup matrix and crackers and sweetness in jelly. Children preferred significantly higher levels of sweet and salt, but lower levels of creaminess, than did their mothers, thus suggesting that the age-related differences in taste preferences were not due solely to children preferring more intense stimuli. For both children and adults, the most preferred level of sucrose in water was significantly correlated with the most preferred level of salt in soup. We also found that the intensity of sweetness preferred in water was related to preferences for sweetness in pudding and jelly, and the intensity of saltiness preferred in soup was related to preference for saltiness in crackers, thus suggesting real-world significance of our measurements. Acknowledgements: The project described was funded, in part, by a grant from the Pennsylvania Department of Health and Award Number R01HD37119 from the Eunice Kennedy Shriver National Institute of Child Health and Human Development. The content is solely the responsibility of the authors and does not necessarily represent the official views of the Eunice Kennedy Shriver National Institute of Child Health and Human Development or the National Institutes of Health. The Pennsylvania Department of Health specifically disclaims responsibility for any analyses, interpretations, or conclusions.

#P342 POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR

Fish oil in the maternal diet: odorant transmission into human milk?

Stefanie Sandgruber[1], *Daniela Much[2], Ulrike Amann-Gassner[2], Hans Hauner[2], Andrea Buettner[1,3]*
[1]University of Erlangen, Food Chemistry Erlangen, Germany, [2]Else Kröner-Fresenius-Center for Nutritional Medicine, Klinikum rechts der Isar, Technical University of Munich Munich, Germany, [3]Fraunhofer Institute for Process Engineering and Packaging (IVV), Sensory Analytics Freising, Germany

Supplementation with fish oil products during pregnancy has become increasingly popular to provide long-chain n-3 PUFAs for brain and cardiovascular health function development. However, these preparations exhibit a considerable odour load. On the other hand, it is believed that maternal dietary odorants are transmitted into the milk, thereby representing the specific odour profiles of each food, and forming latter preferences for specific foods in the neonate. Therefore, the aim of the study was to investigate whether specific fish oil odour constituents translate into human milk. To achieve this goal, human sensory analyses, as well as qualitative and quantitative determination of fish oil odorants was both performed on the fish oil, as well as human milk obtained from mothers after fish oil supplementation in comparison to a control group using high resolution gas chromatography-olfactometry, two-dimensional HRGC-mass spectrometry, in combination with stable isotope dilution assays.

While the administered fish oil was characterized by intense odour notes, the sensory profile of human milk from mothers consuming this oil remained unmodified in relation to control human milk. Analytical data on fish aroma marker substances (hexanal, octanal, nonanal, decanal, (Z)-hex-3-enal, (E)-non-2-enal, (E)-dec-2-enal, (E,E)-deca-2,4-dienal, y-nonalactone, δ-decalactone, y-dodecalactone, oct-1-en-3-one) showed that no statistically significant modification occurred from fish oil intervention. This example demonstrates that food aromas do not necessarily modulate human milk aroma profiles, even if the respective food ingested is high in odour potency.
Acknowledgements: Financed by the German Federal Ministry of Education and Research (BMBF).

#P343 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Duration of Early Flavor Exposure: Impact on Acceptance of Savory Foods after Weaning
Julie A. Mennella, Sara M. Castor, Laura D. Lukasewycz, Gary K. Beauchamp
Monell Chemical Senses Center Philadelphia, PA, USA

Using as a model system the response to protein hydrolysate formula (PHF), we discovered the existence of a sensitive period, before four months of life, when exposure determines the hedonic tone of PHF flavors. The distinctive flavor of PHF is due, in part, to its amino acid content, and most strikingly from a sensory perspective is the abundance of the taste-active amino acid, glutamate, that occurs naturally in many foods (e.g., cheeses, broths) and imparts a savory taste (umami). The present study was designed to determine how the duration of PHF exposure during early life modulates the acceptance of foods that differ in MSG content after weaning. To this end, 2-week-old infants (N=47) were randomized into groups that differed in the number of months they were fed PHF: 0, 1, 3 or 8 months and CMF otherwise. When infants were 8 months of age, we measured their acceptance of plain broth (no MSG), broth containing 0.021 mol/l MSG, and a cheese spread containing Ragusano cheese, which is rich in umami and bitter peptides. Preliminary analyses revealed that 3 and 8 months of PHF exposure led to similar acceptance of the high MSG- containing broth relative to the plain broth; both groups were greater in acceptance than the no exposure group, thereby suggesting a dosing effect. Infants exposed to PHF for 8 months ate significantly more of the Ragusano cheese spread. This model system of research investigation sheds light on sources of individual differences in taste and perhaps cultural food preferences.
Acknowledgements: The project described was supported by Award Number R01HD37119 from the Eunice Kennedy Shriver National Institute of Child Health and Human Development. The content is solely the responsibility of the authors and does not necessarily represent the official views of the Eunice Kennedy Shriver National Institute of Child Health and Human Development or the National Institutes of Health.

#P344 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Food and Beverage Adventurousness and Taste Phenotype among Wine Experts and Wine Consumers
John E Hayes[1,2], Gary J Pickering[3,4,5]
[1]Sensory Evaluation Center, The Pennsylvania State University University Park, PA, USA, [2]Department of Food Science, The Pennsylvania State University University Park, PA, USA, [3]Department of Biological Sciences, Brock University St. Catharines, ON, Canada, [4]Cool Climate Oenology and Viticulture Institute, Brock University St. Catharines, ON, Canada, [5]Department of Psychology, Brock University St. Catharines, ON, Canada

Taste phenotypes have long been studied in relation to alcohol intake, dependence, and family history, with contradictory findings. However, on balance – with appropriate caveats about populations tested, outcomes measured and psychophysical methods used – an association between variation in taste responsiveness and some alcohol behaviors is supported. Recent work suggests supertasting (operationalized via propylthiouracil (PROP) bitterness) not only associates with heightened response but also with more acute discrimination between stimuli. Here, we explore relationships between food and beverage adventurousness and taste phenotype. A convenience sample of wine drinkers (n=330) were recruited in Ontario and phenotyped for PROP bitterness via filter paper disk. They also filled out a short questionnaire regarding willingness to try new foods, alcoholic beverages and wines as well as level of wine involvement, which was used to classify them as a wine expert (n=110) or wine consumer (n=220). In univariate logisitic models, food adventurousness predicted trying new wines and beverages but not expertise. Likewise, wine expertise predicted willingness to try new wines and beverages but not foods. In separate multivariate logistic models, willingness to try new wines and beverages was predicted by expertise and food adventurousness but not PROP. However, mean PROP bitterness was higher among wine experts than wine consumers, and the conditional distribution functions differed between experts and consumers. In contrast, PROP means and distributions did not differ with food adventurousness. These data suggest individuals may self-select for specific professions based on sensory ability (i.e., an active gene-environment correlation) but phenotype does not explain willingness to try new stimuli.

#P345 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Physiological Responses of Food Neophobics and Food Neophilics to Food and Non-food Stimuli
August Capiola, Bryan Raudenbush
Wheeling Jesuit University Wheeling, WV, USA

Individual differences in human food neophobia (the reluctance to try novel foods) and food neophilia (the overt willingness to try novel foods) influence the evaluation of tastes and odors, as well as the sampling of such stimuli. Past research also notes an association of food neophobia to PTC sensitivity, body weight, and cephalic phase salivary response. The present study assessed

physiological reactions of food neophobics and neophilics to pictures of food and non-food stimuli. Stimuli pictures were presented in random order on a computer screen for a period of 5 minutes. No significant differences were found between the groups in relation to non-food stimuli. However, pulse [F(1,21)=5.69, p=.03], GSR [F(1,21)=3.07, p=.09] and respirations [F(1,21)=4.43, p=.05] were significantly increased in food neophobics when presented pictures of food stimuli. Thus, further evidence is provided to support a physiological component at least partially responsible for differences noted between neophobics and neophilics in sensitivity, psychophysical ratings, and willingness to try personality. Such a component may also lead to differences in weight, nutrition, and overall health.

#P346 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Physiological and behavioral reactions to liked and disliked foods

RA de Wijk[1], V Kooijman[1], R Verhoeven[2], N. Holthuijzen[1], K de Graaf[1,3]
[1]AFSG, Consumer Science & Intelligent Systems Wageningen, Netherlands, [2]Social Sciences group, Behavioral Change program Nijmegen, Netherlands, [3]Division of Human Nutrition, Wageningen University Wageningen, Netherlands

Responses of the autonomic nervous system to liked and disliked foods were assessed in 16 children and 16 adults using a combination of physiological (galvanic skin response, heart rate, and skin temperature) and behavioural (facial expressions and speed of sampling) measures. First sight of disliked foods resulted in increased GSR (p=0.01), marginally increased skin temperature (p=0.08), and increased facial expressions of sadness, disgust, and angriness (p=0.05) compared to liked foods. The instruction to visually inspect the foods did not produce significant changes in the pattern of ANS responses. In contrast, the instruction to taste significantly affected the ANS patterns (p=0.04) as well as their interaction with the food's liking (p=0.07). The instruction to smell had even larger effects on the ANS patterns, with significant effects of the food's liking (p =0.01). The ANS patterns found for liked foods are associated with emotions of surprise but also disgust, whereas the patterns for disliked foods are associated with the negative emotions anger, disgust and sadness. The speed with which the foods were sampled was slowest after the instruction to visually inspect the foods (p<0.01) and was not affected by the food's liking.

#P347 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

The Relative Satiety Value of Chewing Gum in American Children

Jack Hirsch[1], Michele Soto[2], Alan Hirsch[2]
[1]Adlai Stevenson High School Lincolnshire, IL, USA, [2]Smell & Taste Treatment and Research Foundation Chicago, IL, USA

Introduction: Isocaloric foods have been categorized based on degree of satiety. Chewing gum without ingestion may similarly induce satiety. We sought to determine if the satiety index, a method of rating the satiety value of foods, would also apply to children chewing sugar-free commercially available gum. A second goal is to compare satiety value of these chewing gums to the standardized control, 110 calorie white bread.
Methods Ten boys and ten girls self-assessed the degree satiety on a visual analogue scale, before and after chewing gum for one minute, fifteen minutes, and thirty minutes. This was performed on Butternut Texas Toast white bread and 11 sugar-free gums: Extra Spearmint, Orbit Cinnamon, Orbit Bubblemint, Orbit Peppermint, Orbit Sweetmint, Trident Strawberry Twist, Trident Watermelon Twist, Trident Original Flavor, Orbit Wintermint, Orbit Spearmint, Wrigley's Solstice. The satiety index was computed for each, and the statistical significance determined for each was compared to the satiety index of white bread.
Conclusion Satiety of mastication of sugar-free gum children was able to be delineated in a hierarchal fashion using the satiety index generated by American children. No statistical significant difference ($p > 0.05$) was found between any of the gums and white bread. All gums tested induced the same satiety index as white bread, however, with less than 1/20th the calories. The results suggest chewing gum may have a role to promote satiety in children as part of a weight loss program. Further investigation is warranted.

#P348 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Lingual Tactile Acuity and Texture Preferences Among Children and their Mothers

Laura D. Lukasewycz[1,2], Julie A. Mennella[1]
[1]Monell Chemical Senses Center Philadelphia, PA, USA, [2]Food Science, Rutgers University New Brunswick, NJ, USA

Knowledge on the influence of texture perception on food choice is lacking in children, despite texture being one of the main drivers of food aversions. In 1999, Essick and colleagues developed a letter-recognition task to determine lingual tactile acuity in adults. In the present study, we used this method when testing 37, 7- to 10-year-old children (22 girls, 15 boys) and their mothers. In brief, each subject was asked to identify with the tip of his or her tongue raised alphabetical letters of varying height (2.5-8 mm) on Teflon strips. The up-down tracking procedure continued until the subject attained 8 reversals. To relate lingual tactile acuity to real-world significance, a forced-choice procedure was used to determine their liking of foods which differed in texture (e.g., smooth versus crunchy peanut butter). Preliminary analyses revealed that although children did not differ from adults in

lingual tactile acuity as measured by this task, they required more stimulus presentations to reach threshold criteria. Greater tactile acuity was associated with preferences for higher-textured foods among adults (p=0.018) but not children (p=0.52). Among all subjects, there was a positive association between age and preference for hard/crunchy foods (p=0.004). This method can be used to assess lingual tactile acuity in both children and adults. As with other aspects of food preferences, preferences for textured foods are influenced both by individual physiology and by increased food experience with age. Acknowledgements: The project described was supported by a grant from the Pennsylvania Department of Health, Monell Institutional Funds and by Award Number R01HD37119 from the Eunice Kennedy Shriver National Institute of Child Health and Human Development. The content is solely the responsibility of the authors and does not necessarily represent the official views of the Eunice Kennedy Shriver National Institute of Child Health and Human Development or the National Institutes of Health. The Pennsylvania Department of Health specifically disclaims responsibility for any analyses, interpretations, or conclusions. The research described was conducted in partial fulfillment of a Master's of Science degree at Rutgers University, NJ, USA.

#P349 **POSTER SESSION VII: TASTE: PERIPHERY; PSYCHOPHYSICS; CHEMICAL SIGNALS AND HUMAN BEHAVIOR**

Glucose transporters and KATP metabolic sensors are present in T1r3-expressing taste cells

Sunil Sukumaran[1], Karen K. Yee[1], Ramana Kotha[1], Timothy A. Gilbertson[2], Robert F. Margolskee[1]
[1]Monell Chemical Senses Center, Philadelphia, PA USA,
[2]Department of Biology Center for Advanced Nutrition, Utah State University, Logan UT, USA

Although T1r2+T1r3 is well established as the major receptor for sugars and non-caloric sweeteners, in mice there is evidence also of T1r-independent sweet taste, particularly so for sugars. Prior to the molecular cloning of the T1rs it had been proposed that sweet taste detection depended upon (a) activation of sugar-gated cation channels and/or (b) sugar binding to G protein coupled receptors to initiate second messenger cascades. By either mechanism, sugars would elicit depolarization of sweet-responsive taste cells which would transmit their signal to gustatory afferents. We examined the nature of T1r-independent sweet taste: our starting point was to determine if taste cells express glucose transporters and metabolic sensors that serve as sugar sensors in other tissues. Utilizing RT-PCR, qPCR, *in situ* hybridization, and immunohistochemistry we determined that several glucose transporters (GLUT2, GLUT4, GLUT8 and GLUT9), a sodium-glucose co-transporter (SGLT1), and two components of the K_{ATP} metabolic sensor (SUR1 and Kir 6.1) were expressed selectively in taste cells. Consistent with a role in sweet taste, GLUT4, SGLT1 and SUR1 were expressed preferentially in T1r3-positive taste cells. Electrophysiological recording determined that nearly 20% of the total outward current of mouse fungiform taste cells was comprised of K_{ATP} channels. Because the overwhelming majority of T1r3-expressing taste cells also express SUR1, and *vice versa*, it is likely that K_{ATP} channels constitute a major portion of K+ channels in the T1r3 subset of taste cells. Taste cell-expressed glucose sensors and K_{ATP} may serve as mediators of the T1r-independent sweet taste of sugars. Supported by grants from PepsiCo to RFM and the National Science Foundation DBI-0216310 to G. Beauchamp in support of Monell's Confocal Microscopy Facility.

Author Index

Bold indicates first/presenting author

Bold indicates first/presenting author

Bold indicates first/presenting author

Bold indicates first/presenting author

Bold indicates first/presenting author

Bold indicates first/presenting author

Notes

AChemS

Association for Chemoreception Sciences

PROGRAM AT A GLANCE

	WEDNESDAY, APRIL 13	THURSDAY, APRIL 14	FRIDAY, APRIL 15
	Registration 3:30 pm to 8:00 pm	**Registration** 7:00 am to 1:00 pm, 6:30 pm to 7:30 pm	**Registration** 7:30 am to 12:30 pm, 6:30 pm to 7:00 pm
8:00 am			**Platform Presentations: Olfaction** 8:00 AM - 10:00 AM *ISLAND BALLROOM*
8:15 am			
8:30 am			
8:45 am			
9:00 am		**Symposium: Optogenetics: Using Light to Study Smell** 9:00 AM - 11:35 AM *ISLAND BALLROOM*	
9:15 am			
9:30 am			
9:45 am			
10:00 am			
10:15 am			
10:30 am			
10:45 am			
11:00 am			**Symposium: New Frontiers in Chemesthesis** 10:15 AM - 12:15 PM *ISLAND BALLROOM*
11:15 am			
11:30 am			
11:45 am			
12:00 pm			
12:15 pm			
12:30 pm			
12:45 pm			
1:00 pm	**AChemS Executive Committee Meeting** 12:00 PM - 3:30 PM *SNOWY EGRET*		**AChem Business Meeting** 12:30 PM - 2:30 PM *ISLAND BALLROOM*
1:15 pm			
1:30 pm			
1:45 pm			
2:00 pm		**Industry Symposium** 1:00 PM - 4:00 PM *ISLAND BALLROOM*	**Break** 2:10 PM - 2:25 PM *GRAND PALM COLONNADE*
2:15 pm			
2:30 pm			
2:45 pm			
3:00 pm			**NIH Workshop: Funding Opportunities for the New Investigator** 3:00 PM - 5:00 PM *INDIAN/BIRD KEY*
3:15 pm			
3:30 pm			
3:45 pm			
4:00 pm			**Symposium: Expanding the Canonical View of Synaptic Processing in the Olfactory Bulb** 3:00 PM - 5:10 PM *ISLAND BALLROOM*
4:15 pm		**Industry Reception** *(Ticketed Event)* 4:15 PM - 6:00 PM *BRECK DECK NORTH*	
4:30 pm			
4:45 pm			
5:00 pm			**ChEMA Social** 5:00 PM - 7:00 PM *BRECK DECK NORTH*
5:15 pm			
5:30 pm			
5:45 pm			
6:00 pm	**Welcome Banquet** *(Ticketed Event)* 6:00 PM - 8:00 PM *BRECK DECK NORTH/NORTH BEACH*		
6:15 pm			
6:30 pm			
6:45 pm			
7:00 pm		**Refreshments Available** 7:00 PM - 7:30 PM *BANYAN BREEZEWAY*	**IFF Lecture** 7:00 PM - 8:00 PM *ISLAND BALLROOM*
7:15 pm			
7:30 pm			
7:45 pm			
8:00 pm	**Welcome/Awards Ceremony** 8:00 PM - 9:00 PM *ISLAND BALLROOM*	**Presidential Symposium: Olfaction in Translation** 7:30 PM - 9:35 PM *ISLAND BALLROOM*	**Break** 8:00 PM - 8:30 PM *BANYAN BREEZEWAY*
8:15 pm			
8:30 pm			
8:45 pm			
9:00 pm	**Givauden Lecture: The Neutral Mechanisms Underlying Touch-based Object Localization** 9:00 PM - 10:00 PM *ISLAND BALLROOM*		**Platform Presentations: Polak Young Investigator Award Winners** 8:30 PM - 10:00 PM *ISLAND BALLROOM*
9:15 pm			
9:30 pm			
9:45 pm			
10:00 pm			
10:15 pm			
10:30 pm			
10:45 pm			

THURSDAY, APRIL 14	FRIDAY, APRIL 15
POSTER SESSION I: Taste Psychophysics; Gustatory Receptors; Chemosensation and Disease 8:00 AM - 12:00 PM *BANYAN BREEZEWAY*	**POSTER SESSION III: Olfaction and Taste Development; Olfactory Psychopysics; Functional Imaging: Taste and Flavor** 8:00 AM - 12:00 PM *BANYAN BREEZEWAY*
POSTER SESSION II: Trigeminal and Chemesthesis; Chemical Signals and Animal Behavior; Taste: CNS 7:00 PM - 11:00 PM *BANYAN BREEZEWAY*	**POSTER SESSION IV: Olfaction: Receptors and Periphery; Olfactory CNS; Taste Modulation; Multimodal Perception** 7:00 PM - 11:00 PM *BANYAN BREEZEWAY*

Registration
7:30 am to 12:00 pm, 6:30 pm to 7:30 pm

Registration
7:00 am to 10:30 am

SATURDAY, APRIL 16

SUNDAY, APRIL 17

Symposium: Ionotropic Sensory Receptors
9:00 AM - 11:20 AM
ISLAND BALLROOM

POSTER SESSION V: Olfactory Modulation; Olfactory Development; Chemosensation and Disease; Functional Imaging
8:00 AM - 12:00 PM
BANYAN BREEZEWAY

Platform Presentations: Taste
8:00 AM - 10:00 AM
ISLAND BALLROOM

Refreshments Available
10:00 AM - 10:30 AM
BANYAN BREEZEWAY

POSTER SESSION VII: Taste: Periphery; Psychphysics; Chemical Signals and Human Behavior
8:00 AM - 12:00 PM
BANYAN BREEZEWAY

Clinical Luncheon: The Sentinel Function of the Chemical Senses in Health & Disease
12:00 PM - 2:00 PM
HORIZONS

Symposium: Odor-based Social Behavior in Mammals: Signals, Brain and Behavior
2:30 PM - 5:05 PM
ISLAND BALLROOM

Refreshments Available
7:00 PM - 7:30 PM
BANYAN BREEZEWAY

Symposium: Basic Tastes: Why Five?
7:30 PM - 9:30 PM
ISLAND BALLROOM

POSTER SESSION VI: Olfaction: Periphery; Olfactory CNS; Psychophysics; Human Chemical Signalling
7:00 PM - 11:00 PM
BANYAN BREEZEWAY

8:00 am
8:15 am
8:30 am
8:45 am
9:00 am
9:15 am
9:30 am
8:45 am
10:00 am
10:15 am
10:30 am
10:45 am
11:00 am
11:15 am
11:30 am
11:45 am
12:00 pm
12:15 pm
12:30 pm
12:45 pm
1:00 pm
1:15 pm
1:30 pm
1:45 pm
2:00 pm
2:15 pm
2:30 pm
2:45 pm
3:00 pm
3:15 pm
3:30 pm
3:45 pm
4:00 pm
4:15 pm
4:30 pm
4:45 pm
5:00 pm
5:15 pm
5:30 pm
5:45 pm
6:00 pm
6:15 pm
6:30 pm
6:45 pm
7:00 pm
7:15 pm
7:30 pm
7:45 pm
8:00 pm
8:15 pm
8:30 pm
8:45 pm
9:00 pm
9:15 pm
9:30 pm
9:45 pm
10:00 pm
10:15 pm
10:30 pm
10:45 pm

Notes

AChemS
Association for Chemoreception Sciences

34th Annual Meeting

April 25-29, 2012
Hyatt Huntington Beach, CA

35th Annual Meeting

April 17-21, 2013
Hyatt Huntington Beach, CA

Givaudan

the world's leading global flavor company;
helping cultivate our customers' market success
with forward thinking Science and Technology