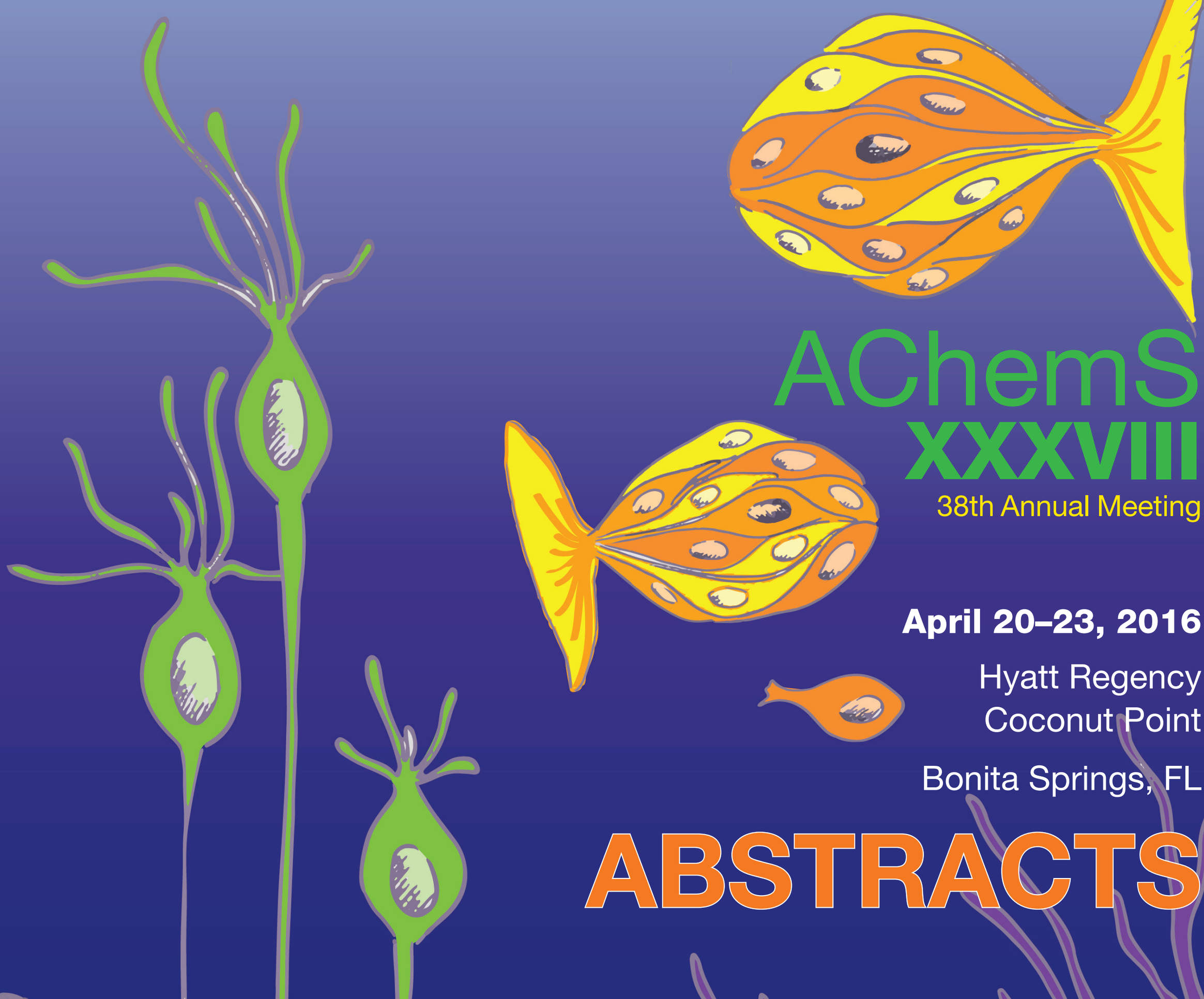

AChemS XXXVIII

38th Annual Meeting

April 20–23, 2016

Hyatt Regency
Coconut Point

Bonita Springs, FL

ABSTRACTS

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ORAL ABSTRACTS

#O1

GIVAUDAN LECTURE
WEDNESDAY, 6 – 7 PM

The Human Oral Microbiome

Ann Griffen
College of Dentistry Division of Pediatric Dentistry and Division of Biosciences, The Ohio State University

New and powerful technologies have driven rapid advances in understanding the human microbiome. We'll overview these and then take a deeper look at the oral microbiome. Oral communities show high fidelity to their niche, dysbiosis leading to disease is common, and the natural history of their acquisition is being elucidated.

#O2

Symposium Overview

Diego Restrepo
University of Colorado Anschutz Medical Campus, Aurora, CO, United States

Olfactory dysfunction after traumatic brain injury (TBI) has a significant effect on the quality of life. Reduced appreciation of food and drinks; loss of employment that depends on an intact sense of smell (e.g. for a Chef); and increased danger from environmental hazards such as gas and spoiled food are significant consequences of post-TBI olfactory deficits that affect quality of life. Because of this, it is key to understand the mechanisms of olfactory dysfunction in TBI. However, recent studies indicate that the study of olfaction in TBI patients is also relevant because it may provide a means to diagnose future central nervous system complications in mild TBI patients. This symposium is designed to bring together experts in basic science, clinical and translational studies of human olfaction and TBI to catalyze discussions on the study of olfactory dysfunction in TBI.

Funding Acknowledgements: NIH DC014253.

FCOI Declarations: None.

#O3

SYMPOSIUM: OLFACTORY DYSFUNCTION IN TRAUMATIC BRAIN INJURY

THURSDAY, 10:30 AM – 12:30 PM

Excitatory/Inhibitory Synaptic Imbalance and Dietary Therapy Following Traumatic Brain Injury

Akiva Cohen[1], Brian Johnson[2], Jaclynn Elkind[3], Hannah Metheny[2], Kaitlin Folweiler[4], Christopher Palmer[4]

[1]Department of Anesthesiology Perelman School of Medicine, University of Pennsylvania, Philadelphia, PA, United States, [2]Children's Hospital of Philadelphia, Philadelphia, PA, United States, [3]Children's Hospital of Philadelphia, Philadelphia, PA, United States, [4]Neuroscience Graduate Group, Perelman School of Medicine, University of Pennsylvania, Philadelphia, PA, United States

Traumatic Brain Injury (TBI) afflicts up to 1.5 million people in the United States each year and even mild TBI can lead to a vast array of long-lasting neurological impairments including deficits in learning and memory and a reduction in seizure threshold. The hippocampus is critically involved in both of these phenomena, and highly susceptible to damage from traumatic brain injury. Optimal brain function requires a delicate balance between excitatory and inhibitory neurotransmission (E/I balance). In TBI, as in epilepsy and other CNS disorders, E/I balance is disrupted. Currently, no therapy exists to mitigate or treat the underlying causes of cognitive impairments suffered by TBI patients. To model mild TBI (mTBI) in mice, we employed lateral fluid percussion injury (LFPI). LFPI is a commonly used rodent model of brain injury that reproduces many key features of human TBI including neuronal cell loss, gliosis, ionic perturbation and memory deficits (Dixon et al., 1987, McIntosh, 1987, 1989, Smith et al., 1991). One week following LFPI we conducted a hippocampus-dependent memory test and investigated electrophysiological alterations in hippocampal activity. We report that LFPI causes hippocampal-dependent memory impairment and regional hippocampal imbalances in excitatory and inhibitory synaptic transmission. In particular, area CA1 demonstrates a decrease in net synaptic efficacy while the dentate gyrus demonstrates an increase in net synaptic efficacy. Furthermore, our laboratory has shown that dietary supplementation with branched chain amino acids (BCAAs) leucine, isoleucine, and valine initiated 48 hours after LFPI and maintained for 5 days restores net synaptic efficacy in the mouse hippocampus and re-instates hippocampal dependent cognitive function at 7 days following LFPI.

Funding Acknowledgements: NIH grants: R37HD059288 and R01NS069629.

FCOI Declarations: None.

#O4

SYMPOSIUM: OLFACTORY DYSFUNCTION IN TRAUMATIC BRAIN INJURY

THURSDAY, 10:30 AM – 12:30 PM

How to Fix a Broken Nose: Olfactory Stem Cells and Regeneration

James E. Schwob
Tufts University School of Medicine/DMCB, Boston, MA, United States

Head trauma and traumatic brain injury are known to cause olfactory dysfunction in humans. The pathological changes in the olfactory periphery suggest that the communication between nose and brain is disrupted at the cribriform plate or by damage to the olfactory bulb/olfactory stalk, which is seen when the injury is very severe. Damage to the olfactory nerve admits the possibility of re-establishing axonal connectivity between epithelium and bulb. However, the abbreviated lifespan of olfactory sensory neurons (OSNs) and the enhancement of neurogenesis following disconnection are not benign. Protracted accelerated neuronal turnover following bulbectomy is associated with neurogenic exhaustion of the olfactory periphery - a condition in which neurons and globose basal cells (GBCs) have disappeared from the olfactory epithelium leaving behind sustentacular cells (Sus) and horizontal basal cells (HBCs). In this setting the HBCs remain dormant, despite the cessation of neurogenesis. We have been investigating neurogenic exhaustion using a mouse genetic model, among others, that abbreviates OSN life-span by indirectly driving the A subunit of Diptheria toxin via the expression of the OMP gene as OSNs mature. At a relatively young age (4-6 months), the composition of the GBC population shifts and then neurons and GBCs disappear, resulting in a pathology closely mimicking that seen in humans. HBCs remain dormant as the expression of the transcription factor p63, the master regulator of HBC activation, remains high. Because the reduction in p63 levels is necessary and sufficient to push the HBCs out of quiescence, targeting p63 may be an effective therapeutic strategy for regenerating GBCs and rejuvenating neuronal production to a neurogenically exhausted OE. To that end, we have been investigating the signal transduction pathways regulating p63 levels and found clear evidence that a Notch pathway-based, Sus-to-HBC cue exerts a central role. With the development of an *in vitro* model in which p63 down-regulation leads to HBC activation, the stage is being set for identifying small molecules to accomplish that end *in vivo*.

Funding Acknowledgements: NIH grants R01 DC002167 and R01 DC014217.

FCOI Declarations: None.

#O5

SYMPOSIUM: OLFACTORY DYSFUNCTION IN TRAUMATIC BRAIN INJURY

THURSDAY, 10:30 AM – 12:30 PM

Trauma-Related Olfactory Deficits: an Otolaryngology Perspective

Bradley J. Goldstein
University of Miami Miller School of Medicine, Department of Otolaryngology, Miami, FL, United States

Common causes of hyposmia or anosmia include active rhinosinusitis, post-viral olfactory disorder, and preceding head trauma. Olfactory losses due to head injuries, such as traumatic brain injury (TBI), remain a challenging problem to treat, despite the regenerative capacity of the peripheral olfactory system. A broader understanding of the potential pathogenic mechanisms involved in TBI from civilian accidents, sports injuries and from recent military conflicts provides new insights into this disorder. Otorhinolaryngologist involvement in the care of such patients can be helpful, providing endoscopic assessment of the nasal airway and olfactory cleft. Exclusion of other treatable causes of olfactory loss, olfactory functional testing, confirmation of head injury as the likely etiology for the loss, and expertise in obtaining olfactory mucosal biopsies for purpose of research studies can all be offered. In addition, the clinical sub-specialty of rhinology maintains an active interest in translational research aimed at sensorineural olfactory disorders, including post-TBI loss. Recent and ongoing studies regarding novel treatment options will be discussed here. Finally, a consideration of future therapeutic targets will be reviewed.

Funding Acknowledgements: NIH K08DC013556.

FCOI Declarations: None.

#O6

SYMPOSIUM: OLFACTORY DYSFUNCTION IN TRAUMATIC BRAIN INJURY

THURSDAY, 10:30 AM – 12:30 PM

Using the Olfactory System to Study Traumatic Brain injury

Leonardo Belluscio, Zhishang Zhou
National Insititutes of Health/NINDS, Bethesda, MD, United States

Olfactory dysfunction is an early indicator of traumatic brain injury (TBI) with patients presenting a variety of olfactory phenotypes. We previously established an in vivo mouse model to study the affects of TBI on olfactory function as a consequence of olfactory bulb (OB) damage. Using a direct Olfactory Bulb Impact (OBI) we reported a clear loss of olfactory function that was partially restored following a 30-day recovery period. To determine if damage to higher brain regions results in a similar loss we applied a Controlled Cortical impact (CCI) to the anterior cortex using UBI7 transgenic mice and assessed olfactory function. In UBI7 mice, all the olfactory sensory neurons (OSNs) express low levels of the I7 receptor in addition to their endogenous receptors. Thus, with multi-electrode recordings we could detect and compare OB responses to Octanal, an I7 receptor ligand, in both CCI and control UBI7 mice. Here we show that following CCI all mice produced clear and reliable odor-induced activation of all OB layers. Interestingly, we revealed that the CCI mice showed activation at lower Octanal concentrations and with prolonged activity compared to control mice, suggesting a possible increase in odor sensitivity in CCI mice. We tested other odors, Acetophenone, Amyl-acetate, Butanal and Propanal, and again found that CCI mice exhibited higher probability of activation than controls consistent with increased sensitivity but also implying a decrease in odor selectivity. Finally, using a buried-food assay we tested behavior and found that CCI mice could indeed locate food faster than controls further supporting enhancement of olfactory sensitivity. Together these data demonstrate a varied olfactory response to TBI that may be specific to the portion of olfactory circuitry that is disrupted and therefore provide a potential means to help identify areas of damage.

Funding Acknowledgements: NINDS Intramural Research Program; NIH/USUHS Center for Neuroscience and Regenerative Medicine.

FCOI Declarations: None.

#O7

Symposium Overview

Joel D. Mainland
Monell Chemical Senses Center, Philadelphia, PA, United States

Humans consume an enormous variety of foods, each containing hundreds of volatile molecules. In contrast, most olfactory coding studies are performed with monomolecular odors. How can we identify which components are essential to the odor and make sense of these complex mixtures? Although reproducing the flavor encoded by this vast array of odorant combinations appears daunting at first glance, recent evidence suggests that reproducing the odors of most foods can be accomplished using only a few hundred building blocks. In this symposium we will examine how analytical chemistry combined with sensory panels has simplified the volatile landscape of food, and how this understanding can lead to the creation of new flavor combinations to excite the palette.

Funding Acknowledgements: R01-DC013339.

FCOI Declarations: Receive support from Ajinomoto Co., Inc.

#O8

From Chemicals to Odor Images

Terry E. Acree[1], Géraldine Prévost[2], Manon Gros[1]
[1]Cornell University Food Science, Ithaca, NY, United States, [2]Cornell University/Food Science, Ithaca, NY, United States

In the 1970s the study of food odor turned from the analysis of volatile components to the detection of odor-active components using bioassays. Most of these were variants of Gas Chromatography- Olfactometry and GCO-Dilution Analysis (Acree, *et al.* 1976, 1984, Ulrich and Grosch, 1987). In the next 20 years more than 300 dominant odorants was identified in foods and other natural products (Arn and Acree, 1997). This knowledge was enormously useful in determining the causes of inappropriate odors or taints in foods but we still could not predict the odor images produced by **mixture** from the sensory properties of the pure compounds alone. Unfortunately, it is mixtures that produce almost all "ecologically" important odor images. Many studies of odorant mixtures indicated that as little as 3 odorants in a are all that can be recognized in a complex mixture [Laing, 1986]. Recently, the analysis of published data on food odors indicated that less than 250 odorants contribute anything to the aroma of all foods [Dunkle, 2014]. Furthermore, it has been estimated that as little as 10 key odorants could yield a billion odor qualities [Buschdid, 2014]. This would imply that it is the ratio of a small number of key odorants (KO) that create food odor images and the "Laing Effect" may be direct evidence of this simplicity. We built an olfactometer to deliver defined compositions with minimal stimulus exposure (< 100ms) in trials lasting less than 5 seconds. Using the key odorants found in commercial potato chips, methanthiol, methional, 3-ethyl-2,5- dimethylpyrazine, we studied potato chip odor image formation. These three components smell individually like 'rotten cabbage', 'potato' and 'toast' are 50 times more potent than all the other chip components. Using different ratios of these components we measured the frequency 'potato chip' was chosen as opposed to the 'component images' [Acree 2014, 2015]. In summary, the results indicate that the interactions are very specific to the odorant pair and range from additive to intensely suppressive. We will discuss these results as they affect our understanding of flavor chemistry and perception.

Funding Acknowledgements: Cornell University Funds.

FCOI Declarations: None.

#O9

Deciphering Food Odor and Taste Codes by Means of a Sensomics Approach

Thomas F. Hofmann
Technical University of Munich, Germany, Freising, Germany

The hedonic evaluation of food flavors is due to the high discriminatory power of the olfactory and gustatory system arising from ~380 odorant and ~30 taste receptors. In contradiction to traditional views, the sheer unlimited variations in food flavors have recently been shown by the so-called Sensomics approach to be due to a "combinatorial chemosensory code" comprising a surprisingly small center group of 3 - 40 key food odorants per item, out of ~230 KFOs out of the 10.000 food-born volatiles, and 10 - 40 key food tastants and taste modulators per item, out of the several 100.000 non-volatile food constituents. Chemosensory recombinants of 3 - 40 key odorants and 15 - 40 key tastants were found to be necessary and sufficient for "synthesizing" the authentic percept of a specific food flavor. Olfaction will be shown to be considered a "constructive sense" that consults a large repertoire of odorant receptors to sense a subset of disparate key volatiles per food item out of a comparatively small population of ~230 key food odorants and create new single odor qualities in our brain that are not represented by any of the single volatiles alone. In contrast, the small number of only 30 taste receptors is designed to sense a vast number of chemically diverse non-volatiles, among which 15-40 per food item are "analytically", that means without any further combinatorial processing, translated into the perception of the five basic taste modalities bitter, sweet, sour, salty, and umami. This complementarity of taste and olfaction might have the evolutionary benefit of being able to detect a wide range of odors, favoring the hedonic behavior to open up new food sources and to develop food preferences, while the "analytical" sense of taste helps to meet different nutritional requirements by detecting attractive sweet carbohydrates (energy source), salty sodium ions (sodium homeostasis), and umami-tasting amino acids (essential amino acids), and to guard against consuming harmful chemicals, many of which taste bitter to humans, as well as excess acids that would negatively affect the maintenance of the body's acid-base balance.

Funding Acknowledgements: Technical University funds.

FCOI Declarations: None.

#O10

SYMPOSIUM: DECONSTRUCTING FOOD

THURSDAY, 10:30 AM – 12:30 PM

The Flavour Network: An Introduction to Computational Gastronomy

Sebastian E. Ahnert
TCM Group, Cavendish Laboratory, University of Cambridge, Cambridge, United Kingdom

The cultural diversity of culinary practice, as illustrated by the variety of regional cuisines, raises the question of whether there are any general patterns that determine the ingredient combinations used in food today or principles that transcend individual tastes and recipes. We introduce a flavor network that captures the flavor compounds shared by culinary ingredients. Western cuisines show a tendency to use ingredient pairs that share many flavor compounds, supporting the so-called food-pairing hypothesis. By contrast, East Asian cuisines tend to avoid compound sharing ingredients. Given the increasing availability of information on food preparation, our data-driven investigation opens new avenues towards a systematic understanding of culinary practice. In light of this we also discuss a variety of datasets on food ingredients and flavour compounds, including chef-curated flavour pairings, aroma compound concentrations, olfactory detection thresholds, and olfactory receptor responses. These datasets can be combined using large-scale data analysis in order to provide a deeper understanding of the impact that shared aroma compounds can have on perceived ingredient compatibility.

Funding Acknowledgements: Royal Society.

FCOI Declarations: Founder and Director, FlavourByte Ltd.

#O11

SYMPOSIUM: DECONSTRUCTING FOOD

THURSDAY, 10:30 AM – 12:30 PM

Volatiles in Fruits can Enhance Sweetness Independently of Sugars

Linda M. Bartoshuk
University of Florida/Food Science and Human Nutrition, Gainesville, FL, United States

Flavor results from the central integration of taste and retronasal olfaction. Although the rules of this integration are far from understood, in the past forty years around a dozen volatiles have been identified that enhance sweet; however the effects were often very small. We discovered serendipitously that many more volatiles contribute to fruit sweetness than were previously suspected. Our group grew 79 heirloom tomatoes (genetically diverse); they were subjected to chemical analysis (sugars, acids, 66 volatiles) and psychophysical analysis (sweet, sour, bitter, salty, umami, tomato flavor, liking using the sensory and hedonic versions of the gLMS). Multiple regression identified volatiles that contributed significantly to sweetness independent of sugars. Similar studies with strawberries, oranges and blueberries identified more than 80 sweet-enhancing volatiles. The magnitudes of the sweetness contributions were quantified by manipulating the volatile content of individual varieties using techniques that alter volatiles but leave other constituents unchanged. These include using mutant tomatoes (devoid of carotenoid volatiles), chilling tomatoes (which reduces some volatiles) and exposing strawberries to blue light (which increases some volatiles). In these experiments the altered fruit were compared to the unaltered controls to quantify the sweetness of specific volatiles. Taken together, these results suggest that small effects of individual volatiles are summing to provide a substantial contribution of sweetness to some fruits. Finally, sweet-enhancing volatiles significantly increased (nearly doubled) the sweetness of a 2 percent sucrose solution to which they were added showing potential for the use of volatiles as sources of sweetness in foods and beverages.

Funding Acknowledgements: NIDCD R21DC013751.

FCOI Declarations: None.

#O12

Symposium Overview

Sanne Boesveldt
Wageningen University, Division of Human Nutrition, Wageningen, Netherlands

Few people appreciate the range of information provided by the sense of smell, while it forms a major part of many of life's pleasurable experiences, whether eating a meal, a walk in the countryside, or intimacy with one's partner. Hence, losing the sense of smell — Anosmia — can have a severe impact on the lives of those who suffer from it. Currently, there is limited treatments or cure available. The symposium will give an overview of current knowledge and status regarding diagnostics and prognosis in the ENT clinic, how this affects patient's quality of life, and present and discuss recent exciting developments in fundamental research with the aim of treating certain anosmias (i.e. olfactory epithelial stem cell regeneration, and gene therapeutic approaches to restore olfactory loss).

Funding Acknowledgements: No funding support.

FCOI Declarations: None.

#O13

The Impact of Olfactory Disorders on Quality of Life and Emotional Wellbeing

Duncan Boak
Fifth Sense, London, United Kingdom

Chemosensory disorders can have a huge impact on patients' quality of life, something that the medical profession and indeed the wider public are largely unaware of. Whilst it is easier to understand the impact that losing the sense of smell has on the enjoyment of food and drink, other effects such as relationship difficulties and reduced interest in socialising are less well understood. Duncan Boak is the founder of Fifth Sense, the UK-based charity for people affected by smell and taste disorders. Drawing on his own experience, in addition to published research based on a survey of nearly 500 Fifth Sense members, he will talk about the hidden consequences of smell and taste disorders and why further research into better treating such conditions is essential.

Funding Acknowledgements: I run a charitable organisation which is funded, at present, entirely by donations.

FCOI Declarations: None.

#O14

CLINICAL SYMPOSIUM: ANOSMIA - THE PATIENT, THE CLINIC, THE CURE?

THURSDAY, 1:30 – 3 PM

ENT Perspective in Sinonasal Anosmia - Diagnosis, Treatment and Prognosis

Antje Welge-Lüssen
Dept. of Otorhinolaryngology, University Hospital Basel, Basel, Switzerland

Olfactory disorders are common complaints in patients presenting to ear, nose and throat physicians. This is reflected by approximately 200000 patients / year in the USA and 79000 patients / year in central Europe seeking medical advice from ENT specialists. Olfactory disorders are classified according to the etiology. Sinonasal disorders are disorders in which the cause of the disorder lies inside the nose and can further be differentiated into inflammatory (chronic rhinosinusitis) or non-inflammatory (tumor, stenosis) disorders. They are considered to be the most common ones. A meticulous history helps in identifying sinonasal disorders which usually develop gradually and are often described as being fluctuating. In case of an underlying inflammation nasal discharge, postnasal drip or nasal breathing problems are encountered. Endoscopic endonasal examination is mandatory and might reveal septal deviation, mucus, pus or polyps or a combination of these and is also mandatory to rule out a tumor. Very often the olfactory cleft is obstructed and cannot be visualized endoscopically. After psychophysical olfactory testing additional imaging such as CT (computer tomography) or MRT (magnetic-resonance- tomography) is only recommended if history and clinical findings do not correspond. Initial treatment consists of steroids which in most cases are prescribed systemically but might also be applied topically in a head down forward position. Surgical therapy (endoscopic functional sinus surgery) is performed in cases of chronic rhinosinusitis failing to respond to conservative treatment only. Markers to predict the effect of either conservative or surgical therapies on olfaction are lacking so far. Often olfactory function improves temporarily only since the underlying disease (such as chronic rhinosinusitis) is yet difficult to cure.

Funding Acknowledgements: Funded by university funds (Dept. Otorhinolaryngology).

FCOI Declarations: None.

#O15

CLINICAL SYMPOSIUM: ANOSMIA - THE PATIENT, THE CLINIC, THE CURE?

THURSDAY, 1:30 – 3 PM

Illuminating Cellular Diversity and Mechanisms of Regeneration in the Olfactory Epithelium Stem Cell Niche

John Ngai[1], Russell Fletcher[2], Levi Gadye[3], Michael Sanchez[4], Diya Das[4], Yoon-Gi Choi[5], Ariane Baudhuin[4], Davide Risso[6], Kelly Street[6], Allon Wagner[7], Michael Cole[7], Sandrine Dudoit[6], Elizabeth Purdom[8], Nir Yosef[7]

[1]Dept of MCB and Helen Wills Neuroscience Institute University of California, Berkeley, CA, United States, [2]Department of Molecular and Cell Biology University of California, Berkeley, CA, United States, [3]Helen Wills Neuroscience Institute University of California, Berkeley, CA, United States, [4]Department of Molecular and Cell Biology University of California, Berkeley, CA, United States, [5]Functional Genomics Laboratory University of California, Berkeley, CA, United States, [6]Division of Biostatistics University of California, Berkeley, CA, United States, [7]Department of EECS University of California, Berkeley, CA, United States, [8]Department of Statistics University of California, Berkeley, CA, United States

Adult tissue stem cells of the olfactory epithelium maintain olfactory function over the lifetime of the animal by supporting tissue homeostasis as well as regeneration in response to injury. This stem cell niche is heterogeneous and includes horizontal basal cells or HBCs (quiescent, reserve stem cells) and globose basal cells or GBCs (proliferating progenitor cells). We are using a suite of approaches - including conditional knockouts of regulatory factors, clonal analysis in vivo and single cell transcriptome profiling - to define the cell types in the olfactory epithelium stem cell niche and the genes that regulate their progression through the olfactory lineage. Together our studies provide a model for understanding the mechanisms regulating adult neural stem cells and lay the groundwork for the future development of treatments to ameliorate certain anosmias and degeneration in the nervous system more generally.

Funding Acknowledgements: NIDCD: R01 DC007235, NIA: K01 AG045344, NIMH: U01 MH105979, NHGRI: T32 HG000047.

FCOI Declarations: None.

#O16

Gene Therapeutic Strategies for Congenital Anosmias: Translating Basic Science and Pre-clinical Work

Jeffrey R. Martens
University of Florida, Department of Pharmacology & Therapeutics, Gainesville, FL, United States

Alterations in cilia formation or function underlie a growing class of pleiotropic disorders termed ciliopathies. The genetic basis of ciliopathies is remarkably complex, with an incomplete but expanding list of more than 89 loci implicated in various disorders. Olfactory dysfunction is a clinical manifestation of ciliopathies and may underlie a significant fraction of the anosmia affecting at least 2.5 million people in the U.S. alone. Current treatment of ciliopathies is limited to symptomatic therapy. Recently, we reported that using gene therapy odor detection can be restored to animals with a hypomorphic mutation in the gene encoding for the ciliary protein IFT88 that results in the loss of cilia on differentiated olfactory sensory neurons (OSNs). This suggests that ectopic gene delivery, *in vivo,* may provide a viable approach to treating olfactory dysfunction, resulting from ciliopathies. Using olfactory specific IFT88 null mice, we now show that gene therapeutic rescue of cilia in the olfactory periphery can prevent neural reorganization in the olfactory bulb and rescue odor guided behavior an important step towards restoring odor perception. Importantly, gene therapeutic rescue of olfactory disruption has been extended to other ciliopathies including Bardet-Biedl syndrome, which represents a treatable patient population. Also we now incorporate clinically approved AAV vectors, which for the first time demonstrate gene transduction and rescue in the olfactory system. In addition, our new data indicate primary cilia on olfactory basal stem cells control neural regeneration and that alterations in the regenerative properties of the OE are a previously unrecognized phenotype of ciliopathies. This highlights cilia pleiotropy within an individual organ system-which influences the cells we target and the approaches used for therapeutic rescue.

Funding Acknowledgements: NIH NIDCD RO1DC009606.

FCOI Declarations: None.

#O17

Taste Pathways and Tastant Selectivity in Tree Shrews *(Scandentia, Tupaia Belangeri)*

Erin E. Maher[1], Suvarnambiga Kannan[1], McKenzie Prillaman[1], Saloni Singh[2], Sussana Owusu-Ansah[2], Isabel Ho[2], Heywood Petry[3], Alev Erisir[2]
[1]University of Virginia, Department of Psychology, Charlottesville, VA, United States, [2]University Of Virginia, Department of Psychology, Charlottesville, VA, United States, [3]University of Louisville, Department of Psychological and Brain Sciences, Louisville, KY, United States

The tree shrews, the closest phylogenetic relative to primates, are used to study sensory system organization and function, psychosocial stress and anesthetics. Having evolved primate-specific sweet-taste receptors, and as our preliminary work demonstrated, a direct solitariothalamic pathway as phenotypical in primates, a tree shrew model provides novel opportunities to study taste perception, processing, pathway development, plasticity and evolution. Additionally, the tree shrew genome has been mapped, making it suitable for use with genetic methods. In this study, we have characterized taste pathways and the tastant selectivity behavior of *T. Belangeri*, and evaluated its use as a model system for primate gustation. We used the two-bottle preference paradigm with sucrose, aspartame, maltose, quinine, malic acid, tryptophan, and umami to further identify tree shrew tastant selectivity characteristics that are primate-like. Anatomical features of tree shrew taste pathways are also more similar to primates than to rodents: Cytoarchitectural borders of the nucleus of the solitary tract, as studied with myelin and Nissl stains, are differentiated anteriorly at the dorsal aspect of the trigeminal tract, and extend 1.5mm posteriomedially before the nucleus borders the 4th ventricle. Gustatory thalamic sensory nucleus (VPMp) is a 500x400x200mm structure with well differentiated borders from surrounding thalamic nuclei. Unlike the thalamic taste nucleus in rodents, VPMp in tree shrew displays ultrastructural properties that are common in LGN, MGN and VPN, including triadic arrangements and glomeruli. Injection of a retrograde tracer in the VPMp led to visualization of cells in the anteriolateral NTS indicating that tree shrew taste information reaches the thalamus via a direct solitariothalamic projection. Overall, our data support the validity of adding gustation to the growing list of primate systems the tree shrew can be used for as a model organism.

Funding Acknowledgements: UVa CHARGE/NSF ADVANCE Grant # HRD1209197.

FCOI Declarations: None.

#O18

Lhx2 Determines Odorant Receptor Expression Frequency in Mature Olfactory Sensory Neurons

Tim McClintock, Guangfan Zhang, William Titlow
University of Kentucky Department of Physiology, Lexington, KY, United States

Homeodomain-like sites are functional elements of odorant receptor (OR) gene control regions. Lhx2 and Emx2 are the most likely transcription factors acting at these elements because they bind proximal promoters in vitro; and in the case of Lhx2, OR enhancers in vivo as well. To avoid the loss of mature olfactory sensory neurons (OSNs) that confounds the assessment of germ line knockouts of *Lhx2* and *Emx2*, we did targeted gene deletion in immature and mature mouse OSNs. Loss of Lhx2 in immature OSNs decreases 676 OR mRNAs. These decreases are due to reduced frequencies of expression rather than reduced amounts of OR mRNA per OSN. Deletion of *Emx2* gives much smaller effects and more often results in increased expression frequencies of ORs. Deletion of both *Lhx2* and *Emx2* in immature OSNs has an additive, rather than synergistic, effect; reducing the frequency of expression of 755 OR mRNAs. Conditional deletion of *Lhx2* in mature OSNs is very similar to deletion in immature OSNs, decreasing the frequency of expression of 765 ORs, but also causing increased expression frequencies of several ORs. Of 1,098 OR mRNAs measured, only 44 are insensitive to the loss of Lhx2. Lhx2 controls expression of nearly all OR genes via a mechanism that not only helps determine which OR gene is chosen for expression in each immature OSN, but also is necessary to maintain expression of the chosen OR. We hypothesize that Emx2 acts differently, probably by helping to control the availability of OR genes for expression.

Funding Acknowledgements: NIH R01 DC007194 and R21 DC013343.

FCOI Declarations: Equity in Odorcept, LLC.

#O19

Odorant Receptor Expression is Perturbed in Mice Following Recovery from Genetically-Mediated Lesion

Jessica H. Brann[1], Xinmin Zhang[2], Eleonora F. Spinazzi[3], Claire Franklin[1], Paulina Lis[1], Natasha Kharas[3], Clara Altomare[3], Stuart J. Firestein[3]
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A repository of basal neural stem cells in the olfactory epithelium (OE) generates excitatory projection neurons that extend long axons from the OE to the olfactory bulb. Each sensory neuron expresses a single odorant receptor (OR), a single gene choice out of ~1200 OR genes in mice, conferring an identity required for odorant detection and appropriate targeting of its axon to the OB. For more than 30 years, it has been known that these stem cells generate sensory neurons, but their ability to faithfully recapitulate the OE after lesion is relatively unexplored. In particular, aging may alter OR expression, implying OR gene choice in newborn neurons may become restricted. Here we probe the ability of the stem cell to generate a diverse array of sensory neurons expressing the appropriate repertoire of odorant receptors in aged animals. To this end we generated a line of mice, iDTR^{+}OMP$^{Cre/+}$, whereby a Cre-mediated excision of a STOP cassette renders mature neurons sensitive to diphtheria toxin (DT) via activation of the DT receptor. This method permits a specific and reversible ablation of mature (OMP-expressing) neurons upon DT administration but without damage to potential synaptic targets in the OB or to other cell types found in the OE. We administered either DT or saline to male mice of several age groups (2-18 months) for six days. RNAs were harvested 30 days following ablation, to allow for complete degeneration and subsequent recovery of the OE. Results reveal that age does not affect the cohort of OR genes expressed following recovery from lesion in DT injected mice. In addition we observe that age does not affect OR expression in saline injected mice. However, the OR repertoire does significantly change following ablation and these effects are observed at all ages tested. These results provide evidence that the regenerative potential of the neuronal stem cell is not altered by age per se, as a wide array of sensory neurons are generated. However, lesion induced ablation of a large number of sensory neurons disrupts the typical OR expression patterns observed in intact mice.

Funding Acknowledgements: R01 DC012567 to SJF and startup funds to JHB from Loyola University Chicago.

FCOI Declarations: None.

ORAL ABSTRACTS

#O20

ORAL SESSION I
THURSDAY, 3 – 5 PM

Gene Therapeutic Restoration of Olfactory Cilia and Odor Detection in Bardet-Biedl Syndrome

Cedric R. Uytingco[1], Corey L. Williams[1], Art Zimmerman[1], Warren W. Green[1], Lian Zhang[1], Jeremy C. McIntyre[2], Val C. Sheffield[3], Jeffrey R. Martens[1]
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Bardet-Biedl syndrome (BBS) is a heterogeneous pleiotropic disorder featuring obesity, renal cystic dysplasia, polydactyly, visual deficits, olfactory dysfunction and anosmia. BBS symptoms arise from defective cilia morphology and/or function in various cell/tissue types throughout the body. Although the genetic etiology of BBS is mostly elucidated, our understanding of BBS protein function is incomplete and clinical treatment is limited to symptomatic therapy, underscoring the need to develop targeted approaches. Toward this end, we assessed the utility of gene therapy to treat olfactory deficits in BBS mutant mice. Despite having relatively healthy olfactory epithelium, BBS mutant mice exhibit reduced ciliation of the olfactory epithelium as well as reduced odor-evoked responses. Our analysis revealed that both *Bbs1* and *Bbs4* mutant mice possess truncated olfactory sensory neuron (OSN) cilia. Along with defects in cilia morphology, afflicted OSNs exhibit altered intraflagellar transport (IFT) of proteins including those of the BBSome complex. In the absence of *Bbs4*, other BBSome proteins enter mutant OSN cilia but are unable to efficiently traffic with IFT. In contrast, ciliary localization of BBSome proteins is fully ablated in *Bbs1* mutant OSNs. In functional rescue experiments, gene therapeutic treatment of BBS mutants with adenovirus-delivered wild-type BBS genes corrected BBSome trafficking defects in olfactory cilia, increased cilia length and number in the olfactory epithelium, and improved odor detection. As a first step toward developing a clinically relevant treatment for BBS-associated anosmia and other gene-associated olfactory dysfunctions, we also restored ciliation and odor detection in *Bbs1* mutants using adeno-associated virus as the gene delivery vehicle. Together, our data demonstrate that ciliogenesis can be induced in differentiated cells of BBS mutants and highlight the potential of gene therapy as a viable option for treating anosmia in BBS patients.

Funding Acknowledgements: NIH/NIDCD R01 - DC - 009606 (JRM).

FCOI Declarations: None.

#O21

Coordinated Guidance of Presynaptic and Postsynaptic Processes by Wnt5, Derailed/Ryk and Van Gogh in Drosophila melanogaster

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While the mechanisms of axon guidance are well understood, those of dendritic targeting have remained obscure. Our goal is to elucidate the mechanisms of dendritic targeting using the *Drosophila* antennal lobe (AL) as a model. In the fly AL, the dendrites of ~50 unique sets of Projection Neurons (PNs) form synapses with the axons of their respective presynaptic partners, the olfactory sensory neurons (OSNs), in ~50 distinct glomeruli. We previously showed that the PN dendrites, which are the first to arrive in the AL, undergo a 30˚ rotation upon the ingrowth of OSN axons, to attain their final glomerular positions. Loss of Wnt5, a non-canonical Wnt expressed in the dorsolateral AL, abolished the dendritic rotation, suggesting that Wnt5 acts as a guidance cue for the dendrites. Indeed, overexpression of *wnt5* strongly repels the DA1, VA1d and DC3 PN dendrites. Moreover, the PN dendrites express the Derailed/Ryk receptor, which antagonizes Wnt5 signaling, allowing the dendrites to migrate up the Wnt5 gradient. We recently found that mutation in the *Van Gogh (vang)* gene, which encodes a four-pass transmembrane protein, also prevented the rotation of the PN dendrites (61.4˚ ± 2.6˚, n=22, versus 30.1˚ ± 1.0˚, n=22, in the wild type, $p<0.0001$), suggesting that *vang* mediates *wnt5* signaling. Accordingly, the *wnt5; vang* double-homozygotes exhibited a *wnt5*-like phenotype, and *wnt5* failed to repel the DA1, VA1d and DC3 PN dendrites in the absence of *vang*, suggesting that *vang* acts downstream of *wnt5*. Surprisingly, cell-type specific gene knockdown and rescue experiments showed that *vang* functions in the OSNs instead of the PNs. Collectively, our data show that, in addition to Wnt5, PN dendrites require instructive inputs from the OSN axons for proper targeting. We propose a model in which the non-canonical Wnt5 protein signals through Derailed/Ryk and Vang to coordinately regulate the targeting of the PN dendrites and OSN axons respectively. Our ongoing experiments will shed light on the cellular and molecular mechanisms by which coordinated targeting of presynaptic and postsynaptic processes in the nervous system is controlled.

Funding Acknowledgements: NIH/NIDCD 2R15DC010916.

FCOI Declarations: None.

#O22

β-catenin is Required for Taste Bud Homeostasis and Maintenance of Behavioral Taste Perception in Adult Mice

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Taste buds are multicellular structures comprising three cell types: Type I - glial-like, Type II - detect sweet, bitter and umami, and Type III - sense sour. Regardless of type, taste cells are continuously replaced throughout life and this renewal is required for taste function. Wnt/β-catenin signaling is a key regulator of homeostasis in many adult tissues. We have shown that increasing β-catenin function in cytokeratin (K) 5+ epithelial progenitors, which normally produce taste cells and non- taste lingual epithelium, is sufficient to alter progenitor output, biasing them to generate primarily Type I cells (Gaillard et al. PLOS Genet 2015 11(5):e1005208). Here we test if β-catenin is required for taste bud homeostasis via conditional deletion of β-catenin function in K5+ progenitors. Over a period of 7 weeks, progressively fewer and smaller K8+ taste buds are found in fungiform (FFP) and circumvallate (CVP) papillae of β-catenin loss-of-function (LOF) mutants. In fact, by 7 weeks, FFP taste buds are entirely absent, while many, albeit smaller buds are still resident in the CVP. All taste cell types are similarly reduced in mutants, as assessed by immunomarkers of Type I, II and III cells, NTPDase2, PLCβ2 and SNAP25, respectively. To explore if and how this reduction in taste cells affects taste behavior, control and mutant mice were tested in a brief-access lickometer paradigm. Interestingly, at 4 weeks, mutant mice could no longer distinguish sweet from water, while bitter detection was comparable in mutants and controls. LOF mice only showed reduced sensitivity to bitter at 7 weeks. These behavioral data are consistent with published reports where the anterior FFP field is more sensitive to sweet, and the posterior CVP is more attuned to bitter (Ninomiya et al. 2000 *J Nutr 130*: *950S*; Sako et al. 2000 *Physiol Behav 71*:*193*). Thus, earlier loss of FFP taste buds corresponds to an early decrement in sweet taste, while a delayed reduction in CVP taste buds correlates with later reduced bitter sensitivity. In sum, we show that β-catenin in lingual progenitors is essential for homeostasis of taste buds and taste perception in adult mice.

Funding Acknowledgements: This work was supported by NIDCD grants R01 DC008373, ARRA DC008373-03S1 and R01 DC012383 to LAB, a South West Affiliate Award from the American Heart Association to DG, P30 Core Grant DC004657 to Diego Restrepo at the Rocky Mountain Taste and Smell Center, and NIH R01 DE024570 to SEM.

FCOI Declarations: None.

#O23

Inhibition of Neurophysiological and Behavioral Responses to Sweet Stimuli by Gastrointestinal LPS

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The gut bacterial environment is shaped by diet and contributes to obesity. The taste system also plays a role in obesity, as it influences food choice and nutrition. However, little is known about the interaction between gut bacteria and taste function. We previously found that lipopolysaccharide (LPS), purified from bacterial cell walls, inhibits neural responses to sweet stimuli. Mice that ingested LPS during a single overnight period displayed reduced chorda tympani nerve (CT) responses to sucrose and decreased expression of the sweet taste receptor subunits in taste buds. Here, we test: (1) whether a more clinically-relevant detoxified form of LPS (dLPS) inhibits neurophysiological responses to sweet tastants; (2) modulation of sweet taste preference by LPS; and (3) potential mechanisms upstream from taste buds. Gavage with dLPS (14 μg/g b.w.) robustly suppressed neural responses to sucrose, glucose, saccharin and acesulfame K in C57BL/6J mice. Moreover, LPS ingestion decreased licking behavior to sucrose, and more prominently to saccharin, in brief-access testing in rats. Neurophysiological and behavioral changes were sweet-selective. The effect of ingested or gavaged LPS is not likely mediated by inflammation; LPS is undetectable in plasma and circulating cytokine levels remain at baseline levels. Instead, our preliminary results indicate that the anorexigenic hormone, leptin, inhibits sweet taste sensitivity in taste buds. We propose a model whereby dLPS stimulates the release of endogenous leptin, activating leptin signaling in taste receptor cells and suppressing sweet taste input to the brain. Introducing a non-toxic bacterial component to the gut may offer a novel, selective approach to controlling sweet taste preference and consumption.

Funding Acknowledgements: NIH (NIDCD) 5R01DC005811 and Medical College of Georgia Pilot Study Research Program 00072 to LM.

FCOI Declarations: None.

#O24

The Correspondence of the Nasal Microbiome and Normal Olfactory Function

Johanna L. Reichert[1], Kaisa Koskinen[2], Christine Moissl-Eichinger[2], Stefan Hoier[3], Veronika Schöpf[1]
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The human body is populated by trillions of microorganisms with tremendous effects on health, disease and behavior. The microbiome changes during the course of life, and in particular older persons possess a distinct, diversity-reduced microbiome, which seems to cause a variety of symptoms. Our microbiome is like a personal fingerprint, as besides the key- and core microbes, our microbial partners are individual. The present pilot study aimed to explore the association of the composition of the nasal microbiome with the smelling ability of healthy human volunteers. On the same day, we first assessed olfactory function with the TDI test using the SniffinSticks battery (Burghart Instruments, Wedel, Germany). Subsequently, microbiome samples from the olfactory mucosa were taken by an ENT doctor using nasal swabs. Microbial composition was assessed by means of DNA sequencing. Our first results with $N = 10$ normosmic, healthy subjects (age *mean* = 22.2, *SD* = 3.4; 8 female, TDI *mean* = 35.9, *SD* = 3.5) point to a tendential relation between two bacteria geni (*Burgholderia* and *Pseudomonadales*) and overall olfactory function (total TDI) of the subjects. Also, we observed differences in microbial community structure between groups of participants with higher vs. lower odor thresholds and odor identification scores. Our results highlight the importance of further investigation of the nasal microbiome and its association with olfactory function. Future research is warranted to reveal whether the nasal microbiome could be an effective biomarker for smelling function and elucidate if the microbiome might offer new therapeutic possibilities for olfactory disorders.

Funding Acknowledgements: FWF (P23205-B09), BioTechMed Graz.

FCOI Declarations: None.

#O25

Multimodal Responses in the Olfactory Bulb

Lindsey A. Czarnecki, Cynthia D. Fast, John P. McGann
Behavioral & Systems Neuroscience Section, Psychology Dept., Rutgers University, Piscataway, NJ, United States

Mammals spontaneously learn contingencies among sensory stimuli including across sensory modalities. Stimulus recognition is faster and more accurate when cross-modal cues are congruent with previous experience (e.g. Gottfried & Dolan, 2003). This suggests that information from multiple sensory modalities could converge in the brain early in sensory processing. In the olfactory system, the olfactory bulb glomerulus receives heavy anatomical "top-down" projections from brain regions that might contain such information. Using wide-field *in vivo* imaging of awake head- fixed mice expressing GCaMP in cells expressing the *Gad2* gene (as in Wachowiak et al. 2013), we see that activity in GABAergic interneurons in the olfactory bulb can be evoked not only by odors but also by lights, tones, and whisker deflections. Anesthesia eliminates responses to non-olfactory stimuli, but responses to odors remain intact and appear qualitatively similar to the responses in the awake mouse. Tracheotomy eliminated the olfactory responses but not the responses to other stimulus modalities, demonstrating that these signals are not driven by respiratory changes. To test whether these stimuli convey predictive information to the olfactory system, mice were presented with a sequence of 13 trials in which a light and tone predicted the presentation of an odor. On the 14th trial, the odor was omitted and the response evoked by the odor-predictive stimuli was observed. On this *odor-omitted* trial, the magnitude of GCaMP signals was not significantly different from the previous odor-present trial, but the pattern of activity was diffuse and not obviously glomerular. These data show that non-olfactory stimuli as well as multimodally-derived expectations about an odor can drive activity in olfactory bulb *Gad2*-PG interneurons and these responses are not airflow dependent.

Funding Acknowledgements: This work was supported by MH101293 and DC013090 to JPM.

FCOI Declarations: None.

#O26

The Effect of Anesthetics on Taste Responses

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Various anesthetics are used commonly in the field to record taste responses from small mammals. We previously found that pentobarbital affects taste transduction by inhibiting 5-HT_3 receptors raising the concern that different anesthetics may have masked effects on taste responses. Pentobarbital, urethane, and ketamine-xylazine are the most common anesthetics used but each has a different mechanism of action. To test whether these various anesthetics influence taste responses in mice, we recorded the chorda tympani nerve responses to different tastants applied on the tongue. Our results show that gustatory nerve responses to all taste qualities are significantly reduced under pentobarbital anesthesia compared to urethane or ketamine-xylazine. Then, to determine if the observed reductions in chorda tympani activity were the result of reduced afferent neuron response to neurotransmitters, we measured responses to ATP and serotonin in isolated geniculate ganglion neurons. After loading with Fura-2-AM, geniculate ganglion neurons showed significantly smaller responses to serotonin but not ATP in the presence of pentobarbital, whereas responses were unaffected by urethane or ketamine-xylazine. While both urethane and pentobarbital bind and potentiate the action of $GABA_A$ receptors, pentobarbital, at anesthetic doses, inhibits 5-HT_3 receptors. In taste buds, 5-HT_3 receptors are expressed on nerve fibers innervating Type III taste cells and previous results suggest that they play a role in transmission of taste information to the nervous system. Our current results provide experimental evidence for a deleterious effect of pentobarbital on taste responses and suggest that the choice of anesthetic should be carefully considered in future studies.

Funding Acknowledgements: Funded by NIH grants R01 DC012555 to SCK, R01DC012931 to TEF, and P30 DC04657.

FCOI Declarations: None.

#O27

ORAL SESSION II
THURSDAY, 3 – 5 PM

Sweet Dopamine: Striatal D2 Receptor Binding and Age are Associated Differentially with Sucrose Preferences in Obesity

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Alterations in dopaminergic circuitry plays a critical role in food reward and ingestive behavior and may contribute to susceptibility to obesity or hinder weight loss efforts. Sweets, the most primitive food reward, release dopamine, and both sucrose preferences and striatal D_2 receptors (D2R) decline with age. However, the relationships among these variables are not well understood. Thus, the aims of this study were to determine a) whether striatal D2R relate to individual differences in sweet preferences, and b) whether the relationships between D2R, age, and sucrose preferences differ between people with and without obesity.19 subjects without obesity (15 female; BMI 22.6 ±2.5 kg/m2; age 28.3 ±5.4y) and 22 subjects with obesity (19 female; BMI 40.3±5.0; age 31.2±6.3) participated in the study. We assessed: 1) sucrose preferences (Monell forced-choice paired-comparison tracking procedure), 2) perception of sweetness intensity of 3%, 12% and 36% w/v sucrose (general Labeled Magnitude Scale), and striatal D2R binding potential (D2R BP_{ND}) using PET with a D2 receptor- selective and radioligand not sensitive to endogenous dopamine, (*N*-[11C] methyl)-benperidol. Groups had similar sucrose preferences, perceived sweetness intensity and striatal D2R BP_{ND} (all $p>0.25$) and D2R BP_{ND} declined with age ($r=-0.45$, $P=0.002$). However, both striatal D2R BP_{ND} (partial $r= -0.69$, $P=0.002$) and age (partial $r= -0.70$, $P=0.001$) correlated with sucrose preferences in normal-weight subjects, such that they together explained 52% of individual variance in sucrose preference ($F=10.7$; $P=0.001$). In contrast, these associations were absent in subjects with obesity (D2R BP_{ND} partial $r= 0.23$ and age partial $r= 0.01$).Our data suggest that the relationships between D2R, age and sweet preferences are disrupted in obesity.

Funding Acknowledgements: This project was supported by the National Institutes of Health (NIH) (DK085575, DK05634, DK56341, UL1 RR024992 and DK 020579).

FCOI Declarations: None.

#O28

Separate Circuitries Encode the Gustatory and Nutritional Values of Sugar

Ivan E. De Araujo[1], Luis Tellez[2], Wenfei Han[2], Xiabing Zhang[2], Tatiana Ferreira[2], Sara J. Shammah- Lagnado[2], Anthony van den Pol[2]
[1]The John B Pierce Laboratory, New Haven, CT, United States, [2]The J.B. Pierce Laboratory, New Haven, CT, United States

Sugar exerts its reinforcing effects via both gustatory and post-ingestive pathways. It is however unknown if sweetness and nutritional signals recruit separate brain pathways to drive ingestion. We show in mice that separate basal ganglia circuitries mediate the gustatory and nutritional actions of sugar. We found that, during sugar intake, suppressing hedonic value inhibited dopamine release in ventral but not dorsal striatum, whereas suppressing nutritional value inhibited dopamine release in dorsal but not ventral striatum. Cell-specific ablation of dopamine- excitable cells in dorsal, but not ventral, striatum inhibited sugar's ability to drive the ingestion of unpalatable bitter solutions. Conversely, optogenetic stimulation of dopamine-excitable cells in dorsal, but not ventral, striatum substituted for sugar in its ability to drive the ingestion of unpalatable bitter solutions. Our data demonstrate that *i.* sugar's sweetness and nutritional value are encoded by separate basal ganglia subcircuits; *ii.* This neuroanatomical arrangement allows organisms to prioritize energy seeking over taste quality.

Funding Acknowledgements: NIH R01DC014859.

FCOI Declarations: None.

#O29

SYMPOSIUM: CHEMOSENSORY PROCESSING IN AMYGDALA SUBNUCLEI: SUBSTRATES FOR AFFECTIVE DECISIONS?

FRIDAY, 10:30 AM – 12:30 PM

Symposium Overview

Michael Meredith
Program in Neuroscience and Dept. Biological Science, Tallahassee, FL, United States

This symposium addresses the proposition that the amygdala is a primary location for the interpretation of chemosensory signals - and is responsible for the engagement of appropriate behavioral and physiological circuits, subject to cognitive and hormonal modulation. The amygdala assigns affective value to sensory information and has direct main and accessory olfactory input to medial and cortical nuclei, indirectly to basolateral amygdala. Symposium chair, Meredith will briefly outline the background for selective chemosensory responses in amygdala subregions and cell populations. The invited speakers, using diverse model systems and experimental techniques, bring together new evidence on chemosensory processing in the amygdala and examples of amygdala modulation producing related changes in chemosignal response and behavior.

Funding Acknowledgements: NIDCD DC0051813, FSU support.

FCOI Declarations: None.

#O30

SYMPOSIUM: CHEMOSENSORY PROCESSING IN AMYGDALA SUBNUCLEI: SUBSTRATES FOR AFFECTIVE DECISIONS?

FRIDAY, 10:30 AM – 12:30 PM

Sexually Dimorphic Encoding of Social Stimuli by the Medial Amygdala

Joseph F. Bergan
University of Massachusetts, Amherst, MA, United States

The vomeronasal system plays an essential role for the recognition of social cues and for orchestrating social behaviors. The medial amygdala is the primary target of chemosensory input from the accessory olfactory bulb and is positioned just upstream of hypothalamic centers dedicated to defensive and social behaviors. Thus, the medial amygdala occupies a central position in the vomeronasal pathway. To data, few electrophysiological studies of sensory processing in the medial amygdala have been conducted. Using multichannel electrophysiological recordings, we simultaneously observed dozens of medial amygdala neurons from a single animal as they responded to chemosensory stimuli. This technique allows us to observe the moment-to-moment encoding of social information by neurons in the medial amygdala, as well as, in other nuclei of the social behavior network. We characterized neural responses to social stimuli, including predators, pups, competitors, and mates. Perhaps most interesting, we found that sensory responses in the medial amygdala of adults differ between individuals depending on sex, age, and genetic strain. Sexual dimorphisms in neural responses were absent in the medial amygdala of juvenile animals (before sexually dimorphic behaviors develop), and required sex steroids during perinatal development for its later expression in adults. Together, these results suggest the medial amygdala encodes social cues sparsely and acts as a 'switch' to select an appropriate social behavior depending on an animal's sex and age.

Funding Acknowledgements: Supported by the University of Massachusetts, NIH NIDCD, & HHMI.

FCOI Declarations: None.

#O31

SYMPOSIUM: CHEMOSENSORY PROCESSING IN AMYGDALA SUBNUCLEI: SUBSTRATES FOR AFFECTIVE DECISIONS?

FRIDAY, 10:30 AM – 12:30 PM

A Medial-amygdala/ Intercalated-nucleus Circuit for Modulation of Amygdala Response to Chemosensory Signals and its Subsequent Output

Don Tucker Award Finalist

Lindsey M. Biggs

Florida State University, Dept of Biological Sciences, Program of Neuroscience, Tallahassee, FL, United States

The medial amygdala (Me) is involved in processing chemosensory signals used by many species to convey social information, however the underlying circuitry has not been well studied. The vomeronasal and main olfactory systems project to Me which responds differentially to different conspecific and heterospecific stimuli, then projects to downstream hypothalamic nuclei responsible for appropriate behavioral responses. My current research focuses on the functional connections between three regions essential to this processing: anterior Me (MeA), the main chemosensory recipient area; posterior Me (MeP), a major output area; and the main intercalated nucleus (mICN) which may modulate Me activity. The mICN is one of several mainly GABA-ir nuclei interspersed throughout the amygdala and previous immediate early gene studies have shown a negative correlation between mICN and MeP responses after exposure to some chemosignals, suggesting mICN may selectively inhibit MeP. Using whole cell patch-clamp electrophysiology in hamster coronal and horizontal brain slices, I have shown functional excitatory and inhibitory connections from MeA to MeP, predominantly excitatory connections from MeA to mICN and functional inhibitory connections from mICN to MeP. These results are consistent with a role for mICN in chemosignal processing and suggest MeA may also modulate MeP activity via excitatory input to mICN. Additional regulation of this circuit via dopamine (DA) and infralimbic (IL) cortex input may also be involved in producing behavioral responses. The MeA/MeP/mICN triangular circuitry shares intriguing similarities with the Basolateral/paracapsular ICN/Central triangular circuit involved in regulation of fear conditioning and extinction in the amygdala. In both, the main nuclei communicate directly and indirectly via distinct adjacent intercalated nuclei, which also receive modulatory DA and IL input. These similarities suggest a possible modular organization within the amygdala for evaluation of different sensory stimuli.

Funding Acknowledgements: NIDCD grants R01-DC005813, T32-DC000044 and funding from Florida State University.

FCOI Declarations: None.

#O32

SYMPOSIUM: CHEMOSENSORY PROCESSING IN AMYGDALA SUBNUCLEI: SUBSTRATES FOR AFFECTIVE DECISIONS?

FRIDAY, 10:30 AM – 12:30 PM

Biological Olfactory Processing in the Amygdala and Reproductive Behavior: The Sheep as a Model

Matthieu Keller
Physiologie de la Reproduction & des Comportements, UMR INRA/CNRS/University of Tours, Nouzilly, France

One noticeable characteristics of maternal behavior in sheep, is the rapid formation of a selective bond with the lamb during the first hours following parturition. Indeed, sheep mothers show an immediate attraction for the lamb following birth and this attraction is induced by the intracerebroventricular release of oxytocin due to parturition. This attraction also provides the opportunity to learn the individual olfactory characteristics of their lamb and leads to a restrictive acceptance of the familiar lamb at the udder, while any other lamb trying to suck will be then rejected. Knowing the role of main and accessory olfactory inputs to medial (MeA) and cortical (CoA) nuclei for the expression of affiliative behavior in other species, we explored the involvement of these structures in the establishment of maternal bonding in ewe. First, we established the anatomical connections between the main and accessory olfactory bulbs and the CoA and Mea in sheep by injections of anterograde or retrograde neuronal tracers. Then, by using immediate-early gene expression, we delineated the brain regions, involved in the processing of lamb odor from other brains regions activated through more general processes related to parturition. We then probed the functional involvement of some of these structures, including the medial, cortical and basolateral nuclei of the amygdala by using temporary and reversible inactivation through stereotaxic injection of lidocaine. As a whole, we showed the involvement of both the CoA and MeA in maternal bonding in sheep. Current works are now focused on the use of magnetic resonance imaging tools to study the neuroendocrine and olfactory networks involved in the establishment of maternal bonding in ewes.

Funding Acknowledgement: This work was funded by the French national research agency (ANR) and the Centre Val-de-Loire regional council.

FCOI Declarations: None.

#O33

SYMPOSIUM: CHEMOSENSORY PROCESSING IN AMYGDALA SUBNUCLEI: SUBSTRATES FOR AFFECTIVE DECISIONS?

FRIDAY, 10:30 AM – 12:30 PM

Odors Associated with Infant Trauma Rescue Depressive-like Adult Behavior via Changes in Amygdala

Regina M. Sullivan
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Rat pups have a sensitive period for learning attachment to the caregiver, and learn the maternal odor regardless of the quality of care received. Classical conditioning underlie this learning, with odor-reward pairing causing an odor preference but also a new maternal odor that supports pups social behavior with the mother and nipple attachment. One unusual characteristic of this learning is that the reward can be presumably pleasant (milk, warmth, tactile stimulation) or noxious (shock, tail pinch). This odor remains important throughout life and continues to guide ecologically relevant behaviors, including reproduction (Fillion & Blass, 1986). Paradoxically, the infant odor, if learned via an abusive mother or shock pairings, will rescue depressive-like behaviors caused by the early life trauma (Sevelenges et al., 2011; Rincon-Cortes et al., 2015). Here we explored the enduring olfactory memory induced by infant odor-shock conditioning. Pups were conditioned daily from postnatal (PN) days 8-12, which pups preferred in infancy but also rescued later-life depressive-like behaviors as characterized by the Forced Swim Test (FST), sucrose consumption and social behavior. Here we explore how the learned infant odor alters amygdala function and interacts with the adult depressive-like behaviors. We found that the presence of the infant odor altered amygdala function and rescued adult depressive-like behavior. A causal role of glucocorticoid/5-HT in the later-life rescue was demonstrated by blocking amygdala 5-HT, which prevented the odor-mediated rescue, whereas increasing amygdala 5-HT and blocking CORT mimicked the odor rescue effect in the FST. Taken together, these results suggest that the infant odor has properties reminiscent of safety signals, which can be acquired in infancy, modulate the adult amygdala LFP and rescue adult depressive-like behavior by modulating amygdala 5HT and glucocorticoids.

Funding Acknowledgements: NIH DC009910, MH091451, HD083217.

FCOI Declarations: None.

#O34

SYMPOSIUM: STRUCTURAL INSIGHTS INTO CHEMOSENSORY RECEPTORS

FRIDAY, 10:30 AM – 12:30 PM

Symposium Overview

Simone Weyand
Biochemistry, Cambridge, United Kingdom

The biology of vertebrate and invertebrate chemosensory receptors has been extensively studied during recent years, but to date their structures still remain unresolved at the atomic level. However, with complementary techniques involving computation, mutational analysis, biochemical and biophysical characterization a holistic understanding of the structure-function relations of these receptors has been approached. Therefore, the objective of the proposed symposium is to bring key experts together who present their latest results at the forefront of understanding the function of taste and olfactory receptors as well as transient receptor potential channels of vertebrates and insects.

Funding Acknowledgements: not applicable.

FCOI Declarations: None.

#O35

Allosteric Modulators of the Human Sweet Taste Receptors

Guy Servant
Senomyx, San Diego, CA, United States

Ingredients that could boost the sweetness intensity of sucrose or HFCS without impacting temporal properties or off-tastes have been a major research focus of the flavor industry for decades. As a result, close to 2,500 patents have been filed or issued with claims describing ingredients having sweetness enhancement properties (SciFinder Patent Search). Still, most of these ingredients only marginally boost the sweetness intensity of different sugars and, in several cases, the effects reported can be explained by sub-threshold additivity rather than true synergy. More recently, bona fide and efficacious sweetness boosters have been identified using a biotechnology approach where sweet taste receptor cell based assays were developed and utilized to screen 100,000s different synthetic and natural samples. The optimized sweetener boosters can reduce the levels of sucrose, and sucralose and HFCS in products by 75%, 50% and 33% while preserving the sweetness intensity. Comparison of the effects of these boosters relative to other reported sweetness enhancers in the cell based assays reveal a clear difference in the apparent magnitude of synergy observed. Reported sweetness enhancers such as Reb-A, Reb-C, thaumatin, NHDC, trilobatin and cyclamate only produce little or no synergy in the assay, explaining their relatively poor effects in sensory studies. On the other hand, newly discovered sweetness boosters result is significant synergy with sucrose, sucralose and HFCS in the assay. Interestingly, the new boosters all share a similar mechanism of action on the sweet taste receptor, binding in close proximity to the sweetener binding site on the T1R2 extracellular venus flytrap (VFT) domain. On the other hand, reported sweetness enhancers such as NHDC, trilobatin, and cyclamate bind to the transmembrane (TM) domain of T1R3 and act mostly as agonists in the assays. These results suggest that unlike other class C GPCRs, the TM domain of the human sweet taste receptor is only loosely functionally coupled to the VFT domain to influence binding or activity of orthosteric ligands.

Funding Acknowledgements: Research funded by Senomyx.

FCOI Declarations: Guy Servant is a paid employee of Senomyx, Inc. GS has a personal financial interest in the form of stock ownership or options ownership of Senomyx, Inc, and is an inventor on several patents and patent applications in the area of cell-based assays for taste receptors and taste receptor modulators.

#O36

Structure, Function and Genetics of the Sweet Taste Receptor

Peihua Jiang
Monell Chemical Senses Center, Philadelphia, PA, United States

Mammalian sweet taste is mediated by a heteromer of T1R2 and T1R3 (T1r2 and T1r3 in non-human species), which belong to the Class C G protein-coupled receptor (GPCR) family. The expressed human T1R2/T1R3 receptor detects and responds to a wide variety of chemically and structurally diverse compounds that taste sweet to humans. Using chimeric human and mouse sweet taste receptors and other mutants, we and others had previously shown that there are multiple binding sites within the T1R2/T1R3 sweet taste receptor, enabling this single receptor to recognize many structurally and chemically distinct compounds. Evolutionarily, sweet taste is believed to predict the nutritional value of sugars and carbohydrates. For species that exclusively feed on meat, we showed that the sweet receptor genes *Tas1r2* and *Tas1r3* may become pseudogenized due to the relaxed selective pressure to maintain the receptor's integrity for carbohydrate detection. For ominvores and herbivores, the T1r2/T1r3 sweet receptor appears to be intact and functional. However, it remains to be fully determined whether the sensitivity and selectivity of the receptor differs from species to species as animals evolved to adapt to their unique feeding ecology. By behavioral taste tests, we show that a few species in Carnivora show distinct preferences for sugars and sweeteners. Using cell-based assays, we show that the giant panda T1r2/T1r3 receptor responds to sugars and sweeteners differently from the orthologous human and rodent receptors. Ongoing structural work at the atomic level may provide a detailed image of how the sweet receptor interacts with different ligands and how the receptors from different species differ in their sensitivity and specificity towards sugars and sweeteners.

Funding Acknowledgements: R01 DC010842 and Institutional Funds from the Monell Chemical Senses Center.

FCOI Declarations: None.

#O37

Predicting Human Odor Perception from Olfactory Receptor Function

Joel D. Mainland
Monell Chemical Senses Center, Philadelphia, PA, United States

There are a large number of single nucleotide polymorphisms and segregating pseudogenes in the human population. Some of these are loci at which functionally distinct alleles coexist in the population, forming a natural mutation of the gene in some individuals, but not others. We used a heterologous assay to determine how often genetic polymorphisms in odorant receptors alter receptor function, and found that 63% of the 27 odorant receptors we examined had polymorphisms that altered in vitro function. On average, two individuals have functional differences at over 30% of their odorant receptor alleles. To determine if these in vitro results are relevant to olfactory perception, we carried out a genotype/phenotype association study. Although humans have over 400 intact olfactory receptors, variation in a single receptor was significantly associated with changes in perception for 19 of the 68 tested odors (28%) ($p < 0.05$, with FDR correction). For these associated ORs, human behavior, in particular perceived intensity, correlates with *in vitro* receptor function. For example, human subjects with genetic variants of OR411 that reduce response to 2-ethylfenchol *in vitro* rated the intensity of the odor to be lower ($F(3,325) = 13.08$, $p < 0.001$) in comparison to subjects with a functional allele. In addition, we can begin to examine how genetic variation in multiple receptors combines to influence odor perception.

Funding Acknowledgements: R03-DC11373, R01- DC013339.

FCOI Declarations: Ajinomoto Co., Inc.

#O38

Cellular and Molecular Basis for Gustatory Detection of Food Texture and Toxins in Drosophila

Craig Montell, Yali V. Zhang, Chao Liu
University of California, Santa Barbara, Santa Barbara, CA, United States

Food texture, such as hardness and softness, has enormous affects on food preferences. However, the molecular and cellular identities of the mechanosensory receptors responsible for sensing the physical properties of food are unknown. We found that akin to mammals, the fruit fly, *Drosophila melanogaster*, preferred food with a specific texture, such as hardness. This taste discrimination depends on the fly homolog of the transmembrane channel-like (TMC) protein family. TMC is expressed in the primary gustatory organ, the labellum, and defines a previously unknown multidendritic neuron (md-L), which extends elaborate dendritic arbors innervating the bases of taste hairs (sensilla). We found that TMC is a cation channel, which is sufficient to sense stretching forces *in vivo* and *in vitro.* We propose that TMC and md-L neurons are long-sought-after molecular and cellular mechanoreceptors through which food mechanics is perceived and encoded by a taste organ. In a second project concerned with Drosophila taste, we made the surprising discovery that two members of the Drosophila rhodopsin gene family, *rh1* and *rh4*, are co-expressed in gustatory receptor neurons (GRNs), and enable flies to sense and avoid feeding on a plant-derived toxin, aristolochic acid. The chemosensory roles played by Rh1 and Rh4 are light-independent, and require only the opsin and not the chromophore. The two opsins enable the animals to detect low levels of the noxious chemical, by initiating an amplification cascade coupled to a heterotrimeric guanine nucleotide-binding protein (Gq), a phospholipase C and the Transient Receptor Potential channel, TRPA1. This study reveals a non-canonical role for opsins in chemosensation.

Funding Acknowledgements: NIDCD R01-DC007864.

FCOI Declarations: None.

#O.01

PRESIDENTIAL SYMPOSIUM
FRIDAY, 7 – 9 PM

Symposium Overview

Susan Travers
AChemS President, The Ohio State University, Columbus, OH, United States

The speakers in this symposium are deserving recipients of the AChemS awards. They will discuss findings arising from diverse technical and theoretical approaches which are united in representing discoveries of major significance in the chemical senses. These bodies of work include elegant molecular studies of the activity-dependent control of olfactory bulb dopaminergic phenotype, insightful evolutionary analyses of bitter receptor variability, the precise use of psychophysics for understanding food flavor and clever interrogations of behavioral and cognitive influences on olfactory bulb processing. Collectively, this symposium will illuminate important interactions between the environment and our chemosensory systems and in doing so reveal fundamental principles of their function.

#O39

PRESIDENTIAL SYMPOSIUM
FRIDAY, 7 – 9 PM

Recipient of the Barry Jacobs Memorial Award for Research in Psychophysics of Human Taste and Smell

Using Psychophysics to Understand Perception of Real Foods

John E. Hayes
Sensory Evaluation Center, The Pennsylvania State University, and Department of Food Science, The Pennsylvania State University, University Park, PA, USA

Thirty years ago, one of the first AChemS officers, Rose Marie Pangborn, was asked why the literature on taste interactions in real foods was so sparse and unsophisticated. Among the myriad reasons she identified for this, Pangborn indicated it was 'unusual for sensory analysts to use good psychophysics, and for psychophysicists to be well informed about the multiple functional properties of food'. While this concern is still relevant today, there are now a handful of researchers who are well grounded in both psychophysics and food science. Moreover, Pangborn noted both sensory analysts and psychophysicists are justifiably intimidated by complex stimuli where a change in one ingredient can alter multiple sensory attributes simultaneously. Understandably, this complexity contradicts the philosophy of simplification and control desired by any experimentalist – however, avoiding such complexity can also lead to reductionism. This talk discusses pitfalls associated with, and insights gained from, applying psychophysics to real foods, with salt, sweeteners and fat as examples. Wine, chocolate and dairy products will also be discussed. This type of integrated, non-reductionist approach is critical to successfully translate basic findings in chemosensory research to diet associated health outcomes and real world commercial applications.

#O40

PRESIDENTIAL SYMPOSIUM

FRIDAY, 7 – 9 PM

Recipient of the AChemS Young Investigator Award for Research in Olfaction

What is the Olfactory Bulb For?

John P. McGann
Behavioral & Systems Neuroscience Section, Psychology Department, Rutgers University, Piscataway, NJ, United States

The olfactory bulb is an evolutionarily-conserved, anatomically impressive structure that is physically separate from the rest of the brain and contains a distinctive array of glomeruli, projection neurons, and local interneurons. Decades of research have revealed much about the bulb's anatomy and physiology, especially the spatiotemporal patterns of neural activity that represent the identity of the external odorant. However, large lesions of the olfactory bulb do not prevent performance on basic laboratory odor discrimination tasks, and the core contributions of the olfactory bulb to overall olfactory function remain uncertain. Moreover, a growing body of evidence suggests that, despite its physical separation, the bulb does not actually function very independently from the rest of the brain, including receiving extensive centrifugal input from cortical regions, state-dependent modulation by the endocrine system and central neuromodulator systems, and behavioral modulation of sensory input through active sniffing. The bulb and its inputs from the nose are also increasingly appreciated to be highly labile, responding to short-term and long-term patterns of input with experience-dependent neuroplasticity. This talk will use recent findings from across the field to try to frame key questions about what the olfactory bulb is actually doing. The outlines of a new consensus appear to emphasize dynamic, task-relevant olfactory processing, the integration of real-time olfactory input with prior knowledge and expectations, and the combination of centrifugal and local circuitry in adaptive filtering of sensory input. This framework raises a number of new questions for future chemosensory research, and may have implications for the etiology of olfactory dysfunction and potential clinical therapies.

#O41

PRESIDENTIAL SYMPOSIUM

FRIDAY, 7 – 9 PM

Recipient of the Ajinomoto Award for Young Investigators in Gustation

Bitter Taste Perception: Genes, Toxins, and Evolution

Stephen Wooding
University of California Merced, Merced, CA, United States

Bitter taste sensitivity varies profoundly from person to person, resulting in differences in preference and consumption. These relationships have broad implications for health in populations today, but how did they first emerge? Mutational patterns in genes controlling bitter perception reveal the ancient origins of taste-health connections. Genes encoding bitter taste receptors (TAS2Rs), which are responsive to myriad noxious compounds in plants, harbor particularly strong signatures of fitness impacts including aggregations of mutants affecting receptor function, extensive population variability, and changes in gene number over time. Associations between TAS2R variation and species ecology shed further light on evolutionary trends, linking gene family diversity and animal diets. These patterns illustrate the depth and breadth of adaptive pressures on bitter perception, and highlight the notion that human diversity in perception today is the legacy of ancient evolutionary processes.

#O42

PRESIDENTIAL SYMPOSIUM

FRIDAY, 7 – 9 PM

Recipient of the Max Mozell Award for Outstanding Achievement in the Chemical Senses

A Journey Towards Understanding Molecular Mechanisms Underlying Synaptic Activity-Dependent Regulation of the Olfactory Bulb Dopamine Phenotype

Harriet Baker

Burke Medical Research Institute, White Plains, NY, The Feil Family Brain and Mind Research Institute of the Weill Cornell Medical College, New York, NY

A collaboration was initiated in 1981 to define the expression pattern of the catecholamine biosynthetic rate-limiting enzyme, *tyrosine hydroxylase (Th)*, in the olfactory bulb (OB) of rodents with lesions in the olfactory epithelium. Thus a career was initiated in olfaction that has lasted over 30 years. This initial collaborative work demonstrated that olfactory sensory neuron innervation was necessary to maintain *Th* expression levels in OB dopaminergic neurons and was among the first studies to show odor-dependent regulation of gene expression. Further work established that *Th* expression was sensitive to almost any perturbation of odorant-induced synaptic activity. Subsequent collaborative studies revealed many features of activity-dependent *Th* expression, including an alternative mechanism for mediating odorant signal transduction in the necklace glomeruli. Later studies concentrated on mechanisms that regulated *Th* transcription in OB dopaminergic neurons. Several transcription factors were identified that directly targeted *Th* cis-regulatory regions to modify expression levels in the OB. This later work culminated with the discovery of highly conserved regions in the *Th* proximal promoter coordinating a regulatory mechanism that included DNA secondary structures. The evolutionary conservation in these sites suggests they serve similar functions in regulating the DA phenotype in most vertebrate species. Understanding the molecular mechanisms regulating the other neurotransmitters in the olfactory bulb remains the task ahead.

#O43

Transduction of a Sense in the Gut

Polak Awardee
Diego V. Bohórquez[1], *Melanie M. Kaelberer*[2]
[1]*Duke University/Medicine and Neurobiology, Durham, NC, United States,* [2]*Duke University/Medicine, Durham, NC, United States*

Gastrointestinal chemosensation has been studied so far from an endocrine perspective. The reason is that enteroendocrine cells, the sensory epithelial cells of the gut, were thought to communicate with nerves indirectly, only through hormones, such as Peptide YY (PYY). However, enteroendocrine cells have the striking features of epithelial cell transducers: they are electrically excitable, fire action potentials, possess voltage-gated channels, express synaptic proteins; and recently, we reported that they are innervated by nerves in the small intestine and colon. Here, we explored the components and functional connectivity of this novel gut neuroepithelial circuit.

First, to define if peripheral nerves connect with enteroendocrine cells, we used the monosynaptic rabies virus B19G SADΔG-GFP, and developed a complementary transgenic mouse model, PyyCRE_tdTomato_rabG, to enable cell-specific spread of the virus. We found that when delivered in the lumen of the colon, there was visible GFP in mucosal and vagal nerves PyyCRE_tdTomato_rabG mice. These data show that colonic enteroendocrine cells are innervated by vagal nerve fibers. Second, to test neurotransmission in this neuroepithelial circuit, we developed an *in vitro* co-culture system using purified enteroendocrine cells and dissociated vagal nodose neurons. The two cell types connect in 12-36 hours and often remain viable for at least 5 days, showing that this neuroepithelial circuit can be isolated and recapitulated *in vitro*. Third, we then tested the possibility of afferent gut-to-brain transduction using whole cell electrophysiology. We discovered that a stimulus of 10mM of glucose applied to the enteroendocrine cell induces excitatory post-synaptic potentials and action potentials in the connected neuron. The same stimulus does not activate a nodose neuron by itself. These findings unveil a gut-brain neuroepithelial circuit with the ability to transduce a chemical sense.

Funding Acknowledgements: K01 DK-103832, AGA - Pilot Research Award, UNC-CGIBD Pilot Research Grant, and Duke Medicine - Gastroenterology.

FCOI Declarations: None.

#O44

NPY-like Regulation of Host-Seeking Suppression in Aedes aegypti Mosquitoes

Polak Awardee
Laura B. Duvall[1], *Leslie B. Vosshall*[2]
[1]*The Rockefeller University, Laboratory of Neurogenetics and Behavior, New York, NY, United States,* [2]*The Rockefeller University, Laboratory of Neurogenetics and Behavior, HHMI, New York, NY, United States*

Female *Aedes aegypti* mosquitoes, the primary dengue vector, require a blood meal from hosts to develop and lay eggs and are strongly attracted to human hosts when seeking a blood source. After a blood meal, attraction to host cues (such as CO_2 and human odor) is strongly suppressed. Host-seeking suppression is maintained until the female lays her eggs up to 4 days later, when host-seeking drive returns and she seeks another blood meal. The cycle of blood feeding and egg laying is critical to disease transmission. Our goal is to understand the mechanisms underlying the dramatic behavioral switch that occurs during host-seeking suppression. Although it is known that abdominal distension contributes to short-term suppression, less is known about the mechanisms that cause sustained behavioral inhibition. Neuropeptide Y (NPY)-related signaling pathways play evolutionarily conserved roles in feeding motivation and we hypothesize that they play a role in *Aedes*. We show evidence that a class of NPY-like receptors affects host-seeking suppression in mosquitoes following a blood meal. We identify pharmacological compounds, designed to target human NPY receptor, which modulate host-seeking in a behavioral screen. Using a cell based assay, we implicate specific *Aedes* NPY-like receptors as targets of the compounds identified in our behavioral screen. Ongoing work focuses on systematic deorphanization of the neuropeptide receptors in *Aedes* to identify novel interactions between neuropeptides and receptors as well as the generation of targeted mutations of these genes using CRISPR/Cas9 genome editing. Identification of the endogenous signaling underlying this dramatic switch in feeding behavior may inform novel strategies for vector control.

Funding Acknowledgements: This work was supported by a Rockefeller University Women & Science Fellowship, an American Philosophical Society Postdoctoral Fellowship in Biological Science to LBD and NIDCD grant R01DC014247 to LBV.

FCOI Declarations: None.

#O45

Podoplanin (PDPN) Plays Important Roles in the Development of Taste Organs in Mice

Polak Awardee
Nandakumar Venkatesan[1], Masako Toda[2], Lynda Bonewald[3], Yuji Mishina[2], Hong-Xiang Liu[1]
[1]University of Georgia, Athens, GA, United States, [2]University of Michigan, Ann Arbor, MI, United States, [3]University of Missouri - Kansas City, Kansas City, MO, United States

Podoplanin (PDPN) is an extensively glycosylated mucin type transmembrane protein and broadly expressed in major vital organs. Although the function of PDPN is largely unknown, it is reported that it plays an important role in the lung development and the ontogeny of nervous system, motility of dendritic cells and host response to brain injury and gliomas. In the present study we found the distribution of PDPN immunoreactivity in both epithelium and mesenchyme in the developing mouse tongue. In the tongue epithelium from E13.5 to E18.5, PDPN signals were progressively restricted to the basal epithelial cells and co-localized with Sox2. However, the PDPN immunoreactivity was not seen in the early immature taste bud cells where Sox2 is highly expressed. PDPN expression was also detected in the tongue mesenchyme and muscle cells. Further we characterized the regulatory roles of PDPN in the development of tongue and taste papillae using *Pdpn* null mutant (KO) model, that was validated by the absence of PDPN immunoreactivity and the abnormalities in the lung. At E15.5, the circumvallate and fungiform taste papillae in the mutant tongues were more elevated and had an enhanced expression of sonic hedgehog, a developing taste papilla marker, compared to the littermate control. However, the circumvallate and fungiform papillae were not fully developed in the E18.5-P1 mutant tongues. Also the lingual epithelial and muscle cells were highly disorganized in the mutant tongue tissues. No apparent changes were found in other tissues in the cranial region, including tooth, skull and skeletal muscles. Our data suggest a stage- and tissue-specific role of PDPN in the development of mouse tongue, taste papillae and the organization of lingual muscles.

Funding Acknowledgements: NIDCD NIH Grant R01DC012308 to HXL, NIDCR NIH R01DE020843 to YM.

FCOI Declarations: None.

#O46

Morphological Diversity of Taste Nerve Fibers in the Mouse Tongue

Polak Awardee
Tao Huang, Robin F. Krimm
Dept. Anatomical Sciences and Neurobiology, University of Louisville Medical School, Louisville, KY, United States

In most sensory systems, neurons were initially classified into different types based on their morphology. Nothing is known about the branching characteristics of individual taste neurons or whether they can be organized into distinct morphological types. A popular view of taste coding is that the neurons simply reflect the response characteristics of specific taste receptor cell types. If so, limited branching or morphological diversity would be required. To determine if this is the case, we traced the peripheral branching patterns of single taste neurons from where they enter the tongue to their terminations in the taste bud, using sparse cell genetic labeling. We analyzed total length, branch-points and ends, and the number of taste buds innervated. None of these traits were distributed normally, indicating that taste neurons exist as morphological subtypes. Based on the numbers of branch ends, 30 taste fibers were classified as sparsely- (S) (43.3%), moderately- (M) (33.3%), or heavily- (H) branched (23.3%). S fibers had the fewest branch ends (2.92±1.11) ranging from 1 to 5; H fibers had the most branch ends (15.4±2.99) ranging from 13 to 20. The 3 fiber types varied in the number of taste buds each innervated (F=5.71, p<0.01). Most S fibers innervated only 1 taste bud (mean = 1.54±0.66). The mean number of taste buds innervated by M (3.10±1.52) and H (3.57±2.23) fibers was greater than for S fibers (p<0.05), with some M and H fibers innervating 6-7 taste buds. Similarly, total length was different among the 3 fiber types (F=5.44, p=0.01); M (13.2±5.47 mm) and H (13.5±5.19 mm) fibers were longer than S fibers (7.98±2.72 mm). The average branches per taste bud was different across fiber types (F=9.80, p<0.001). H fibers had more branch ends (6.05±3.72) per taste bud than S (1.97±0.55) and M (3.21±1.41) fibers. Given this impressive diversity in the branching characteristics of individual taste nerve fibers, it is unlikely that all gustatory neurons simply reflect the input from one single taste receptor cell type. Instead, gustatory neurons exist as morphological subtypes, which likely vary in the specificity of taste response.

Funding Acknowledgements: NIH DC014857 and NIH DC007176.

FCOI Declarations: None.

#O47

Effective Connectivity of the Insular Gustatory Cortex

Polak Awardee
Emilia Iannilli, Paul Bovelet, Thomas Hummel
Interdisciplinary Center "Smell & Taste", Dep. of Otorhinolaryngology, TU Dresden, Dresden, Germany

Capsaicin is a component of chili peppers evoking burning and pungenct sensations due to the interaction with TrpV1 receptors located on the sensory neurons in the mucous membranes inside the mouth. Recent studies associate capsaicin ingestion with satiety [1], reduction of body fat [2], prevention of cancer [3], and enhancement of taste [4] to reduce daily sodium intake. In the present study we focused on the cross-modal interaction between nociception and taste using brain imaging. Specifically, our fMRI experiment aimed to differentiate taste-related networks from nociception- related networks and the network activated by the combined gustatory/nociceptive stimulation. Four different tastes conditions at iso-intensity level were presented independently to 24 healthy volunteers (mean age ± s.d. = 26 ± 3 years): capsaicin; NaCl; mixture of of NaCl and capsaicin; artificial saliva. The fMRI results were assessed at the whole brain level following the contrast of the stimulus masked by the activity revealed by the artificial saliva. We found differences and similarities in patterns associated with the three different conditions. Across the trigeminal nociceptive pathway, capsaicin stimulation evoked greater activation in thalamus, somatosensory areas, posterior insula and superior medial frontal cortex. One of the activations in the left insula overlapped with the taste induced activation. Effective connectivity analyses suggested a preferable link of the same areas with the somatosensory cortex during the capsaicin stimulation, with somatosensory cortex and amygdala following the mixture stimulation and with the lateral prefrontal cortex following the taste stimulation. Our results confirm the functional heterogeneity of the insular cortex. Specifically, inside the areas of the gustatory response, they suggest dynamic links to secondary cortices not only related with taste perception but more widely involving higher sensory networks possibly associated also with ingestion-related behavior. 1. van Avesaat M, Troost FJ, Westerterp-Plantenga MS, Helyes Z, Le Roux CW, Dekker J, Masclee AA, Keszthelyi D. *Capsaicin-induced satiety is associated with gastrointestinal distress but not with the release of satiety hormones.* Am J Clin Nutr. 2015 Dec 30. 2. Saito M. *Capsaicin and Related Food Ingredients Reducing Body Fat Through the Activation of TRP and Brown Fat Thermogenesis.* Adv Food Nutr Res. 2015;76:1-28. 3. Park JM, Lee HJ, Yoo JH, Ko WJ, Cho JY, Hahm KB. *Overview of gastrointestinal cancer prevention in Asia.* Best Pract Res Clin Gastroenterol. 2015 Dec;29(6):855-67. 4. Narukawa M, Sasaki S, Watanabe T. 2011. Effect of Capsaicin on Salt Taste Sensitivity in Humans. Food Sci. Technol. Res. 17: 167-170.

Funding Acknowledgements: This research was funded by in-house research sources of the Department of Otorhinolaryngology of the TU Dresden, Germany.

FCOI Declarations: None.

#O48

The Interaction Effect of Estradiol Concentration and Intake Duration on Olfactory Performance Levels in Oral Contraceptive Users

Polak Awardee
Veronika Schöpf[1], *Iris Ohrenberger*[2], *Kathrin Kollndorfer*[2]
[1]*Institute of Psychology, University of Graz, Graz, Austria,* [2]*Dept of Biomedical Imaging and Image- guided Therapy, Medical University of Vienna, Vienna, Austria*

Previous studies indicated an effect of hormonal contraceptives on olfactory performance (Derntl *et al*, 2013). However, various studies provided contradictory results. One possible explanation of inconsistency in results may be the high variability in estradiol dose and duration of intake. Therefore, we aimed at investigating the impact of estradiol dose and duration of intake in oral contraceptive users on olfactory performance. Forty-seven females (age range 18-34 years) using oral contraceptives were included in this study. All subjects used either a low (0.02 µg estradiol; n=23) or high dosed (> 0.02 µg estradiol; n=24) oral contraceptive. Olfactory performance of all participants was assessed using the "Sniffin' Sticks" battery, including separate subtests for olfactory threshold, odor discrimination and odor identification. All subtests can be summed up for a measure of overall olfactory performance (TDI score). Results revealed an interaction of estradiol dose and duration of intake in the investigated study sample. TDI scores did not differ between high and low estradiol dose in subjects who used oral contraceptives for five years or less. In subjects with a longer duration of intake (6 years or longer), females using low- dose oral contraceptives achieved higher TDI scores than females using high-dose contraceptives. The results of the present study indicate that olfactory performance is not only influenced by oral contraceptive intake, but also by an interaction of estradiol dose and duration of oral contraceptive use. The findings of this investigation may be a first step in unravelling the complex interaction between hormone concentration and olfactory performance. References: Derntl B, Schöpf V, Kollndorfer K & Lanzenberger R (2013) Menstrual Cycle Phase and Duration of Oral Contraception Intake Affect Olfactory Perception. *Chem. Senses* **38:** 67-75

Funding Acknowledgements: Research was supported by a grant of the Austrian Science Fund (FWF; P23205-B09) to VS.

FCOI Declarations: None.

#O49

Chemosensory Model Systems

Christopher T. Simons[1], Wolfgang Meyerhof[2], Alan Spector[3], Punyatoya Mohapatra[4]
[1]The Ohio State University, Columbus, OH, United States, [2] Department of Molecular Genetics, German Institute of Human Nutrition Potsdam-Rehbruecke, Germany, [3]Department of Psychology, Florida State University, Tallahassee, FL, United States, [4]AFB International

Industrial and academic experimental designs often limit the use of humans in evaluating taste, smell and chemesthetic stimuli. As such, effective and convenient model systems of human chemosensory function are needed. Three chemosensory models that have been routinely used in both academic and industrial settings include cell-based high-throughput screening assays, animal behavioral models, and electrode-based systems including the electronic tongue or electronic nose. Whereas each of these models has proven utility in the study and evaluation of chemosensory function, none fully recapitulates the human mechanism or experience. In this year's Industry Workshop, participants will explore and discuss the underlying science and limitations associated with these typical chemosensory models. The structure of the workshop is to enable dialogue between academic and industry researchers regarding these important techniques and to explore their effectiveness and convenience as it relates to industrial research needs. A discussant for each of the three sub-theme's will present a short overview of the topic to the whole assembly and attendees will then break out into discussion groups of their choosing for the remaining period of time. During the discussion groups, conversations regarding the pros and cons of the techniques will be deliberated in more depth. Workshop discussants and moderators include Wolfgang Meyerhof, Ph.D., Department of Molecular Genetics, German Institute of Human Nutrition Potsdam-Rehbruecke, who will introduce the topic of cell-based screening assays; Alan Spector, Ph.D., Department of Psychology, Florida State University, who will discuss the topic of animal behavioral models; and Punyatoya Mohapatra, Ph.D., AFB International who will lead the discussion on electrode- based systems including the electronic tongue and electronic nose.

Funding Acknowledgements: None.

FCOI Declarations: Employee of AFB International PM.

ORAL ABSTRACTS

#O50

SPECIAL LECTURE
SATURDAY, 4 – 5 PM

Sometimes You Feel It, Sometimes You Don't

Christophe Laudamiel
DreamAir, New York City, NY

Mr Laudamiel will be talking about the frontstage and backstage of olfactory art, and scientific effects explored or perhaps to be explored related to that still infant discipline. An interactive speech showing on the one hand what outside sniffers and viewers have a hard time exploring within the mysterious metier of perfume composer, on the other hand what concepts Mr. Laudamiel has been exploring in his numerous installations around the world. Lecture material: just bring your nose and your brain, the rest will be provided.

#O51

Symposium Overview

Leslie B. Vosshall
HHMI-The Rockefeller University, New York, NY, United States

Olfaction is the least understood of the senses. Despite many centuries of thought about how smell "works," we still have no way to predict what a molecule will smell like. Given a chemical structure, the only way to determine what olfactory percept it gives is to smell it. This stimulus-percept problem was solved long ago for color vision and tone hearing. Solving the stimulus-percept problem for olfaction is more complicated because odor stimuli do not vary along a predictable axis. We do not know how many different smells exist, and we do not know how odors are arranged in perceptual space. The perception of an odor stimulus can be described in a number of distinct ways: how intense is it, how pleasant is it, what does it remind me of? The goal of the DREAM Olfaction Prediction Challenge is to use a crowd-sourced competition to develop models that can predict how a molecule smells from its physical and chemical features. A model that allows us to predict a smell from a molecule will provide fundamental insights into how odor chemicals are transformed into a smell percept in the brain. The talks in this symposium will present the psychophysical dataset that formed the foundation of the DREAM challenge as well as the models of the teams who won the challenge.

Funding Acknowledgements: Supported in part by grant # UL1 TR000043 from the National Center for Advancing Translational Sciences (NCATS, National Institutes of Health (NIH) Clinical and Translational Science Award (CTSA) program. LBV is an investigator of the Howard Hughes Medical Institute.

FCOI Declarations: I am a member of the Scientific Advisory Board of International Flavors & Fragrances Inc. (IFF), and receive compensation for this consulting activity.

#O52

Toward a Solution of the Stimulus Percept Problem in Olfaction

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How an odorous molecule is perceived by humans cannot currently be fully predicted based on features of the molecule. To improve current predictions, perceptual data for a larger number of diverse molecules is needed. Towards this goal, we tested the olfactory perception of 480 structurally and perceptually diverse molecules at two concentrations using a panel of 55 subjects. For each stimulus, we collected data on the perceived intensity and pleasantness and on how 20 different verbal descriptors are applied to the odors. We also asked subjects if they know what the smell "is" and how familiar they are with it. This dataset formed the basis of the DREAM Olfaction Prediction Challenge, the outcome of which will be presented in this symposium. Using this dataset, we replicated several previously reported correlations between perception and molecular features. The number of sulfur atoms in a molecule is highly correlated with the "garlic" descriptor, and large and structurally complex molecules are perceived to be more pleasant. The data we collected highlight perceptual variability in olfaction, and how subject familiarity with the odor affects how verbal descriptors are applied to these smells. Despite these complication, our dataset reveals robust correlations between molecular features and perceptual qualities, many of which have been described independently in different datasets. This shows that olfactory perception can be predicted at least to some degree based on stimulus features. A computationally more sophisticated analysis of this dataset will likely result in formal models that predict a considerable portion of molecules' perceptual qualities.

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#O53

Prediction of Personalized Olfaction Response

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Prediction of chemical smells from its physical and chemical features is a challenging task. The 2015 DREAM Olfaction Challenge included the sub-challenge 1 to build prediction models for olfactory response of each individual. To predict these personalized olfaction response, we used decision tree as a base-learner of this chemical structural data. We used decision tree because the dimension of the structure data is high with more than 4000 parameters and also the data matrix is sparse. Therefore, decision tree helped us to reduce the dimension as well as to determine the decision boundary between zeros and the rest of the values. The olfactory responses reported by different individuals were noisy; therefore, in order to capture the personalized features and stability for predictions, we used both personal as well as global response across all individuals. Where we used 0.2*individual score + 0.8* global average for each chemical as predictions but this parameter is flexible to achieve decent performance. The best performing algorithms of 2014 DREAM Broad Institute Gene Essentiality Challenge also applied similar technique. No external data was used for building models. This technique was the best-performing method of sub-challenge 1 for the prediction of olfactory response of each individual.

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FCOI Declarations: None.

#O54

From Shape to Smell: Predicting Odor Descriptors from Molecular Features

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The DREAM Olfaction Prediction Challenge asked participants to predict 21 different olfactory perceptual descriptor ratings (i.e. smell descriptors) of single molecules using a library of several thousands structural features of those molecules. One of the two sub-challenges asked participants to make predictions for the mean and variance of these ratings across subjects. Here I describe the winning submission to that sub-challenge. I trained Random Forest Regression models to predict descriptor ratings using the molecular features, using extensive cross- validation to optimize these models by size, structure, and feature selection. While this technique was effective for predicting the mean descriptor ratings, accurately predicting the variance across subjects required identifying and exploiting a relationship between the variance and the mean that followed from basic psychometric considerations. I also determined the complexity of each of these (intuitive) perceptual descriptors, as identified with the number of distinct molecular features required to successfully predict them. Finally, the remaining uncertainty in the problem is statistically dissected into data-limited and theory-limited components, informing whether more experiments or better models should be the priority for future research in this area.

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FCOI Declarations: None.

#O55

Personalized Predictions of Human Olfaction: A Community Effort to Accurately Infer Odor Perception from Molecular Structures

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It is currently not possible to fully predict how a person smells a molecule based solely on the structural features of the molecules generating the sensation. To improve current predictions, perceptual data for a larger number of diverse molecules is needed but also an adequate setup to evaluate the predictions. Towards this goal, we used a dataset of olfactory perception, obtained from a panel of 49 subjects exposed to 479 structurally and perceptually diverse molecules at two concentrations, to organize the DREAM olfaction prediction challenge. DREAM is a collaborative-competition project organized by IBM and Sage Bionetworks to blindly assess the performance on algorithms using unpublished datasets. For this challenge, 32 teams submitted algorithms solving two types of problems: Use the data from 338 odors and 49 subjects to build models that predict odor intensity, as well as odor valence and the matrix of 19 odor descriptors 1) for each of 49 subjects and 69 odors 2) for the average and standard deviation of all 49 subjects and 69 odors. We will here describe the diversity of models submitted, from linear models to classification trees and support vector machines, to their performance in individual and mean predictions topping 0.55 and 0.75 PCC. We will conclude on the surprising success obtained when performing blind predictions of individual/mean human olfaction perception using molecular structures and a well generated training set.

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FCOI Declarations: PM, GC, GS, AD, and RN are employees of IBM.

#O56

From Taste Buds To The Hindbrain: In Search Of A Rosetta Stone

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The question of how taste bud cells convey information on the identity of taste stimuli to the central nervous system has been examined and debated for decades. Early patch clamp recordings were limited by the inability to categorize the taste bud cells that responded to tastants. More recent electrophysiological and Ca2+ imaging studies, carried out in combination with cell type-selective fluorescent tags revealed that Type II taste bud cells, which express taste GPCR and their effectors, mostly respond to taste stimuli of only a single quality (e.g. bitter). In contrast, Type III cells in their native taste bud environment are broadly responsive to multiple taste stimuli. Further along the taste axis, single unit recordings on taste nerves showed that most afferent fibers yield maximum activity in response to a "best" stimulus. Nevertheless, many fibers are also stimulated by tastants of multiple qualities. Consensus has been lacking through the variety of stimuli, concentrations and preparations tested over the years.

We have used mice in which a genetically encoded Ca indicator, GCaMP3, is expressed in all taste neurons, to quantify the functional responses of gustatory afferent neurons in situ in the geniculate ganglia of anesthetized mice. The response properties of individual neurons in our study were stable at a given concentration, but were surprisingly plastic over the concentration range. At low stimulus concentrations, ≈70% of neurons were narrowly tuned, responding to a single type of stimulus. As stimulus concentration increased, geniculate ganglion neurons became more broadly tuned, responding to stimuli of two to four qualities. At the highest concentrations tested, but still within the behavioral response range, no more than 15% of neurons were narrowly tuned. Thus, the scheme by which the gustatory system encodes taste quality is more complex than a simple diagram of labeled lines connecting labeled points along the axis. Alternatives will be discussed, drawing on coding logic used in other sensory systems and neural pathways.

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FCOI Declarations: None.

#O57

Temporal Coding of Taste in the Brainstem: What Is It Good For?

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The nucleus of the solitary tract (NTS) and the parabrachial nucleus of the pons (PbN) are respectively the first and second "relays" in the central pathway for taste in the brainstem of non-primate mammals. While several decades of research in anaesthetized animals have described the contributions of both rate and temporal coding of taste, more recent recordings from awake freely licking rats paint a richer, more detailed picture of the nature of how taste stimuli are represented in the brainstem. While there is abundant evidence for temporal coding of taste quality in cells recorded from awake rats, the relatively lower values of information conveyed in awake vs. anesthetized rats suggest that ensemble coding may be necessary for perfect identification of a tastant. More importantly, the presence of a variety of response types (e.g. lick-related responses, anti-lick responses, taste-olfactory responses, etc.), in addition to purely taste responses, in both brainstem nuclei underscore the idea that the sensory and motor components of taste are inextricably linked. Thus, the streams of activity arising from afferent and efferent limbs are blended in the responses of brainstem neurons. Furthermore, feedback from more central structures in the gustatory pathway can refine the temporal code for taste in the brainstem, an effect that may enhance performance in taste-related behavioral tasks. In all, we suggest that responses to taste stimuli in NTS and PbN neurons reflect the sensory-motor integration of information that is ultimately part of the neural circuit regulating ingestion.

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FCOI Declarations: None.

#O58

SYMPOSIUM: A MODERN TAKE ON CNS GUSTATORY PROCESSING:
FROM PRIMARY NERVE INPUT TO THE DRIVING OF CONSUMPTION DECISIONS

SATURDAY, 6 – 8 PM

Spatiotemporal Coding of Individual Chemicals by the Gustatory System

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While the olfactory system makes use of combinatorial coding to efficiently represent vast numbers of odors, the gustatory systems of animals as diverse as mice and flies are thought to only represent a small number of basic tastes. Studying the simple and tractable gustatory system of the moth *Manduca Sexta*, we observed that chemical-specific information is found in the spatiotemporal pattern of activity of the gustatory receptor neuron (GRN) population. This information is preserved by their post-synaptic target neurons, which integrate the activity of multiple types of GRNs to respond broadly to many tastants with chemical-specific patterns of spiking activity. In addition, moths exhibited chemical- specific gustatory behavior, indicating that chemical-specific information is preserved throughout a sensory-motor pathway. Our results suggest that like the olfactory system and the other major sensory systems (vision, somatosenstaion and audition), gustation uses a combinatorial code to represent large numbers of stimuli rather than small numbers of basic categories. Applying this interpretation to other animal systems may help explain several phenomena not readily accounted for in the basic taste framework, including: 1) the expression of large numbers of gustatory receptors sensitive to specific chemicals, 2) gustatory responses to chemicals not readily associated with any basic taste, 3) single cells responsive to chemicals associated with multiple basic tastes, but not every chemical from those basic tastes, and 4) the presence of chemical specific gustatory behavior in many species.

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#O59

SYMPOSIUM: A MODERN TAKE ON CNS GUSTATORY PROCESSING:
FROM PRIMARY NERVE INPUT TO THE DRIVING OF CONSUMPTION DECISIONS

SATURDAY, 6 – 8 PM

State Transitions in Gustatory Cortex during Taste Processing: Electophysiological Evidence and Computational Models

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Neural activity in gustatory cortex is dynamic over the timescale of one to two seconds during which a tastant covers an animal's tongue. We have analyzed multiunit data via Hidden Markov modeling (HMM), which combines spike trains from an ensemble of different units to reveal the single-trial dynamics of these neural responses. We found that the ensemble data was best explained as a sequence of noise-induced transitions between distinct ensemble states of activity that could be reproduced in a model circuit comprised of simulated spiking neurons. Intriguingly we found that such a transition-based model could provide a framework for decision making—such as the decision to swallow or expel a tastant from the mouth—that under reasonable biophysical assumptions would yield more optimal choices than the standard model of decision making by perfect integration of evidence. To connect our taste processing data with decision making, we focused on the transition to the particular state of neural activity that was most correlated with the palatability of the tastant, since the palatability is the attribute of a tastant measured by the ensuing behavioral choice. Indeed we found that the apparent ramping of the neural correlate of palatability previously found by the standard analysis of averaging across trials was artifactual. When each trial was aligned to the timing of the appropriate HMM transition, rather than to stimulus onset, a much sharper step in palatability correlation was revealed upon across-trial averaging. Indeed, the data was more consistent with an instantaneous transition that the dynamics produced by the standard integrator model. Moreover, these transition times—acquired from neural spikes in gustatory cortex—correlated significantly with the animal's behavior, confirming our hypothesis that the state transition was choice-related. Taken together, our data support a model of taste-dependent decisions made abruptly via a transition between distinct states of neural activity.

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FCOI Declarations: None.

#101

Functions of the GFL/Ret Signaling Pathway in the Development and Physiology of the Peripheral Taste System

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In the peripheral taste system, neurotrophin-4 and brain-derived neurotrophic factor, signaling through the TrkB receptor, are important mediators of axon guidance and survival of chemosensory geniculate neurons projecting to the anterior tongue. While the functions of the neurotrophins in the development of the peripheral taste system have been extensively explored, it is unknown whether additional families of neurotrophic factors are involved in this process. Furthermore, it is unknown whether distinct subpopulations of geniculate chemosensory neurons exist that can be delineated based on their dependence on different neurotrophic factors, as in other sensory populations. In this study, we provide evidence that the glial cell line-derived neurotrophic factor (GDNF) family ligands (GFLs), signaling through the receptor tyrosine kinase, Ret, have an important role in the development and physiology of the peripheral taste system. Utilizing Ret reporter lines, we observed that Ret is widely expressed in geniculate neurons at E12.5, but then becomes restricted postnatally to a subset of Phox2B+ geniculate ganglion neurons innervating fungiform papillae. Analysis of Ret germline knockout mice revealed that Ret deletion results in a significant loss of Phox2B expression in the geniculate ganglion as well as a loss of chemosensory innervation of fungiform taste buds. To determine whether the adult subpopulation of Ret+ neurons innervating the anterior tongue represents a specific sensory modality, we utilized a diphtheria toxin A system to eliminate Ret+ geniculate neurons specifically in adult mice. This resulted in the loss of tactile responses, but not taste reponses, of the anterior tongue using electrophysiologic recordings of the chorda tympani. Collectively, these data indicate that Ret is required for the early development of geniculate chemosensory neurons, and that Ret+ neurons in the adult represent a novel subpopulation of Phox2B+ neurons with unique physiologic properties.

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FCOI Declarations: None.

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#102

A Reduction in Sweet Taste Sensitivity in Pregnant Mice Correlates with Decreased Sweet Receptor Expression

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An alteration in maternal intake during pregnancy can permanently affect the metabolism, growth, and feeding behavior of the progeny, in both mice and humans. While much is known about how maternal diet affects offspring fitness, less is known about how gustation is involved in guiding and promoting food intake during this crucial period. Humans display intense food cravings and exhibit altered taste preferences during pregnancy; however, the mechanistic details underlying these changes during pregnancy are presently unclear. We investigated taste changes in pregnant mice using brief-access taste testing and found decreased sensitivity to sucrose during the mid/late stages of pregnancy. We hypothesize that altered taste preferences in parturition result from changes in the expression profile of taste buds of pregnant mice. Following this, we examined taste receptor mRNA expression as a potential pathway for the modulation of taste preference. Of the sweet receptor subunits, T1R2 expression was decreased during late pregnancy, while sweet and umami receptor subunits T1R1 and T1R3 were unchanged. Interestingly, the bitter receptors T2R5 and T2R8 appeared unchanged during mid/late pregnancy despite previous reports of increased bitter sensitivity during pregnancy. Our results imply that the various physiological changes induced by pregnancy may influence the taste transcriptome, and resulting feeding behavior during parturition.

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FCOI Declarations: None.

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#103

POSTER SESSION I
THURSDAY, 8 – 10:30 AM

Expression of the TrkB Receptor is Downregulated before Birth Dividing Taste Neurons into Three Subpopulations

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Taste neurons of the geniculate ganglion could be a single subpopulation of similar neurons or they could differentiate into subtypes during development, defined by differences in expression. Embryonically, brain-derived neurotrophic factor (BDNF) regulates the development of most taste neurons, but postnatally, BDNF becomes restricted to subpopulations of taste receptors cells with specific functions. We speculated that the receptor for BDNF, TrkB, may also become developmentally restricted to a specific neuronal subtype. To test this possibility, we used a combination of genetic labels, one that identifies taste neurons (*Phox2b*-Cre::tdtomato) and one for TrkB ($TrkB^{GFP}$), to quantify TrkB receptor expression in taste neurons. We found that only half of the Phox2b+ neurons (354±33) were TrkB+ (185±21) in whole-mount adult geniculate ganglia. To determine when TrkB expression starts to decline, we quantified the TrkB+ taste neurons at E13.5, E15.5, E17.5, P0, and P60 (adult). Most Phox2b+ neurons expressed TrkB during early embryonic ages (E13.5=96%, E15.5=91%), TrkB expression starts to decline by E17.5 (63%). By P0, only 58%±7 of taste neurons expressed TrkB, similar to adults (52%). However, a subset of non-taste TrkB-expressing neurons (P2X3+, Phox2b-) in the geniculate ganglion did not down-regulate TrkB (E13.5=93%±3, P0=95% and Adult=86%±1), indicating that the reduction in TrkB is specific to taste neurons. To determine if all the taste neurons that express TrkB embryonically are TrkB dependent, we quantified Phox2b+ taste neurons in conditional TrkB knockouts. By P20, only 30±5 taste neurons remained in geniculate ganglia of Phox2b-Cre:$TrkB^{GFP/lox}$ mice. Together, these findings indicate that TrkB expression/dependence divides gustatory neurons into 3 subpopulations, 1) neurons that always express TrkB and are TrkB dependent (52%), 2) neurons dependent on TrkB during development but no longer express TrkB in adulthood (40%), 3) neurons that never express or depend on TrkB (8%). Future studies will determine how differences in taste neuron differentiation are reflected in taste function.

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FCOI Declarations: None.

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#104

POSTER SESSION I
THURSDAY, 8 – 10:30 AM

Blocking TrkB-signaling Decreases TrkB+ Innervation to the Taste Bud Within Two Weeks

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The neurotrophin, BDNF, maintains innervation to the taste bud in adulthood. However, the receptor mechanisms, cell types, and timing of BDNF-TrkB signaling are unclear. The full length TrkB receptor for BDNF is only expressed in the ganglion neurons innervating taste buds, not in the taste bud cells. Here, we wanted to determine whether BDNF functions through full length TrkB receptor and whether disrupting TrkB-signaling is effective within two weeks, a timing consistent with a role of TrkB-signaling regulating innervation to new taste cells. Using transgenic mice with a chemically blockable (1NMPP1) TrkB signaling domain ($TrkB^{F616A/F616A}$), we blocked trkB-signaling with precise timing. To refine analysis of TrkB+ innervation, we used $TrkB^{CreER/+}$:tdtomato:YFP mice injected with tamoxifen such that TrkB+ neurons were labeled with red, green, or both (yellow) fluorescent proteins allowing analysis of TrkB+ innervation with single fiber resolution. All taste buds were innervated by TrkB+ fibers in our 3 control genotypes/treatment groups (TrkB:tdtomato:YFP receiving 1NMPP1 or vehicle, and $TrkB^{CreER/F616A}$:tdtomato:YFP receiving vehicle) but only 70% of taste buds were innervated by TrkB+ fibers in mice when TrkB-signaling was blocked for two weeks ($p<0.001$;$TrkB^{CreER/F616A}$: tdtomato:YFP receiving 1NMPP1). Also, 70% of taste buds are innervated by fibers of multiple different colors in the control groups indicating that these taste buds were innervated by more than one TrkB+ fiber. However, after two weeks of blocking TrkB-signaling only 35% of taste buds were innervated with TrkB+ fibers of more than one color ($p<0.001$). We are currently measuring number of terminal branches, total branching length, and average length of each branch in whole taste buds from each animal. Our data suggests that blocking TrkB-signaling reduced TrkB+ fiber innervation to the taste bud within two weeks. This means BDNF regulates innervation through the full length TrkB receptor and the timing of TrkB+ fiber loss is consistent with TrkB regulation of innervation to new BDNF+ taste receptor cells.

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FCOI Declarations: None.

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#105

Children and Nonnutritive Sweeteners: From Hedonics to Genes

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Children live in different sensory worlds than adults, preferring the sweet taste of sucrose at higher levels. In recent years, use of nonnutritive sweeteners (NNS) has increased, including in foods, beverages, and medicines consumed by children. Given the paucity of research on hedonic response to the taste of NNS, particularly among children, our objectives were two-fold: 1) to examine individual differences in liking of several NNS compared to sucrose among children and adults, and 2) to determine whether such differences were related to common variants of the bitter taste receptor gene, *TAS2R31*. A racially diverse sample of children (6-14 years) and their mothers (N=78) tasted, in randomized and counterbalanced order, 5mL of 1.05 M sucrose, 0.002 M sucralose, 0.010 M aspartame, and 0.012 M ace K, and rated their liking/disliking for the taste of each on facial hedonic scales. All participants were genotyped for several *TAS2R31* variant sites, including those previously shown to be related to perceived bitterness intensity of ace K among adults (R35W, L162M, A227V and V240I). Haplotypes were constructed by tracing the parental origins of the alleles, and subjects with the two common diplotypes were compared (RLAV vs WMVI). Children were more likely to like the taste of sucrose, sucralose, and aspartame (all $p<0.05$), but not ace K, than were adults. Overall, a greater percentage of children and adults liked the taste of sucrose (73%), sucralose (69%), and aspartame (79%) than the taste of ace K (37%). There was an additive effect of the *TAS2R31* gene on ace K hedonics: more subjects with two copies of the WMVI haplotype (78%) disliked the taste of ace K than those with one (34%) or no copies (6%; $p=0.01$). This genotype-phenotype effect did not generalize to sucrose or other NNS. Taken together, these data a) pinpoint a particular *TAS2R31* haplotype (WMVI) responsible in part for the hedonic response to ace K, and b) are the first to identify the relationship between this haplotype and ace K liking among children, highlighting the potential role of individual differences in vulnerability to overconsumption of NNS-containing products.

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FCOI Declarations: None.

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#106

Accelerated Olfactory Aging in Transgenic Mice Recapitulates Aging Human Olfactory Epithelium

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As a consequence of aging, patches lacking olfactory sensory neurons (OSNs) crop up across the olfactory epithelium (OE) in human. These regions termed areas of exhaustion have other common characteristics such as a loss of the globose basal cells (GBCs), the active stem cells. Metaplastic replacement by swaths of respiratory epithelium (RE) often occurs in areas formerly OE. The horizontal basal cells (HBCs), the reserve stem cells, appear unchanged with aging even in these regions of exhaustion. To investigate this age related damage to OE we have created a mouse model of accelerated aging in the OE by crossing an OMP-tTA knock-in mouse (Tet-Off) with a TetO-DTA mouse line. This bigenic mouse in the absence of Doxycycline will express Diphtheria Toxin subunit A (DTA) in OSNs expressing OMP, which will lead to the death of mature OSNs. With time, the damage to the OE progresses through a series of stages: 1) GBC proliferation increases (marked by BrdU, Sox2, NeuroD1); 2) subsequently GBCs decline; 3) GBCs and neurons disappear (exhaust); and 4) eventually OE undergoes metaplastic replacement with RE. The latter two recapitulate the phenotype commonly seen in the aging human OE. Areas characterized by exhaustion of GBCs and neurons are seen as early as 2 months of age consistently. There is no observable change in p63 levels in HBCs at any time examined, which suggests that they are remaining dormant. In the olfactory bulb (OB) glomerular size decreases, which is consistent with denervation as a consequence of neuronal death and inadequate replacement. All of these findings mirror what has previously been seen in humans making this a valuable model of aging. The reversible nature of the lesion also enables us to observe recovery of both the OE and OB after switching to Doxycycline feed. This mouse model, by providing a constant insult to the tissue that eventually "wears it down", is a better mimic of human pathology, as compared to methods using a harsh lesion (often one-time) that permits a degree of repair that is not characteristic of the aged OE. This model also gives us the opportunity to assay each component of the OE as it changes with aging.

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FCOI Declarations: None.

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#107

Age-related Decline Between Functional Connectivity of the Hippocampus and the Olfactory Network in Resting State fMRI

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Intranasal olfactory and trigeminal processing are closely linked but have different relationships to memory and cognition. Both memory and olfaction are known to decline with increasing age, and impaired memory and olfaction are early onset symptoms of age-related neurodegenerative diseases, while the trigeminal system is unaffected. This research investigated the effect of age on resting state functional connectivity of the olfactory and trigeminal networks. Two freely available resting state fMRI data sets were downloaded from the NITRC.org (Atlanta, New York), and combined with data collected in our lab to generate a large sample (N=103; 51 females) spanning the adult age range (20-61 years). Each data set was collected with similar acquisition parameters (i.e., T2* fMRI, TR = 2 sec, 180-200 volumes). Preprocessing included motion correction, detrending and global signal regression, smoothing with 8 mm^3 FWHM, and transformation into MNI space. Seed time courses were extracted in MNI space, defined from coordinates that purportedly anchor olfactory and trigeminal systems in activation studies. Seeds comprising the olfactory network (ON) included piriform cortex and oribtofrontal cortex. Seeds comprising the trigeminal network (TN) included anterior insula and cingulate cortex. Scanner site, sex, and age were used as covariates in group level analyses. Results are reported at $p < .001$, minimum cluster of 120 contiguous voxels. The ON showed stronger functional connectivity with the medial prefrontal cortex and posterior cingulate cortex, caudate, thalamus, and medial temporal lobes. The TN showed stronger functional connectivity with the precuneus, thalamus, brainstem, cerebellum, pre- and postcentral gyrus, and cingulate gyrus. Connectivity between the ON and parahippocampal gyrus was negatively correlated with age. These results are consistent with previously reported functional relations between olfactory and memory systems, and suggests that the intrinsic relation between olfactory and episodic memory systems may decline with increasing age, while the trigeminal system may be unaffected.

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FCOI Declarations: None.

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#108

POSTER SESSION I
THURSDAY, 8 – 10:30 AM

Measured Olfactory Dysfunction for U.S. Adults Aged 40+ Years: Prevalence and Risk Factors Derived from the 2012 National Health and Nutrition Examination Survey (NHANES)

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The NHANES 2012 chemosensory (taste and smell) exams are the first conducted on a representative sample of the U.S. population. In 2012, 70.5% (n=1,281) of NHANES participants aged 40+ years (N=1,818) completed the exam; non-participation was mostly due to time constraints, only 2% refused. Trained health technicians administered an 8-item forced choice odor identification task, scored by number correct: normosmic, 6-8, and measured olfactory dysfunction (MOD) as hyposmic, 4- 5, and anosmic/microsmic, 0-3. Interviewers recorded self-reported smell problems or alterations in "the past year," "since age 25," and "phantom odors." Questions were asked about colds/flu, dry mouth, nasal congestion, extracted tonsils or wisdom teeth, head injury, broken nose, sinus infections, if smell problems have been discussed with healthcare providers, and if quality of life (QoL) has diminished due to chemosensory problems. Multivariable logistic regression was used to adjust for the complex sample design and calculate odds ratios (OR) and 95% confidence intervals (CI). MOD was found in 12.4% (13.3 million; males, 55%, females, 45%) of adults aged 40+; anosmia/microsmia in 3.2% (3.4 million; males, 74%, females, 26%). Age-specific MOD prevalences were 4.2%, 12.7%, and 39.4% versus 10.2%, 18.2%, and 13.3% for reported smell problems during past year for ages 40-49, 60-69, and 80+, respectively. In multivariable analysis, MOD risk factors were: tonsillectomy, OR=2.1, CI: 1.7-2.5; heavy drinking, OR=1.9, CI: 1.7-2.1; multiple ear infections, OR=1.3, CI: 1.14-1.4; fair/poor health, OR=1.3, CI: 1.05-1.5. Self-reported poorer ability to taste food flavors, persistent tastes in mouth during past year, and diminished QoL also were associated with MOD. Higher income/education and physical activity were protective, reducing the risk of MOD.Prevalence of MOD in this representative national sample parallels findings from prior epidemiological studies. Risk factors for MOD suggest potential interventions that may delay or prevent some of the age-related decline in olfactory function.

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FCOI Declarations: None.

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#109

Mechanisms of Flavor Loss in Foods and Beverages

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Sense of smell that affects the quality of foods and beverages is started by activation of the receptor cell. In the cilia of this cell, signal transduction involves two ion channels, cyclic nucleotide-gated (CNG) and $Cl_{(Ca)}$ channels. It has been believed that the flavor-loss of foods/beverages is caused by a reduction of endogenous odorants. However, we have previously reported that the odorant responses of the receptor cells are suppressed by certain types of odorants though the suppression of CNG channel, and suggested that part of flavor loss is caused by such channel suppression. To investigate the molecular mechanism of this phenomenon, we examined the effect of 2,4,6-trichloroanisole (TCA) which is known to be a strong suppressor of the CNG channel (Takeuchi et al., 2013). In this work, we show that the suppression ratio (SR) observed in CNG channels is decreased depending on the parameters when membrane lipid components were changed in the native ORCs. In addition, we saw a positive correlation between LogD and SR when investigated with varieties of TCA derivatives. It is thus likely that a mechanism of channel suppression is the conformation change of channel proteins caused by a structural change of surrounding lipid bilayer. The least effective concentration of approx. 1aM (10^{-18}M) can be explained by this idea. We will also show that, besides TCA, foods/beverages contain chemicals when their evaluation is reduced in the market. This work is supported by JSPS 26115710, 26430016 and Daiwa Can Company.

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#110

Neural Activity Required at Adulthood for Maintenance of Terminal Fields of Nerves That Transmit Taste Information

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Neural activity plays a critical role in the development of many sensory systems (Fox et. al, 1996; Katz and Crowley, 2002). In most cases, this activity-induced plasticity is not maintained throughout an animal's life (Webster, 1983; Hook and Chen, 2007; Urzurumla and Gaspar, 2012). Work in our lab found that eliminating NaCl taste responses in mice by conditionally removing the functional epithelial sodium channel (ENaC) from taste bud cells throughout development led to a dramatic reorganization of the terminal fields of nerves that carry taste information to the nucleus of the solitary tract (NST). At adulthood, the chorda tympani (CT), glossopharyngeal (IX), and greater superficial petrosal (GSP) nerve terminal fields were 1.5X - 2X larger in knockout mice compared to their littermate controls. This shows that ENaC-mediated neural activity is necessary for proper development of gustatory circuitry (Sun and Hill, 2015 AChemS Abstract).

The present study was designed to explore the effects of eliminating NaCl-driven activity on terminal field organization when ENaC was removed from taste bud cells, only at adulthood. To do this, we crossed mice that expressed CreER under control of the Keratin 8 promoter with mice in which the *αENaC* gene was floxed. With application of tamoxifen at adulthood, these animals lost functional NaCl- driven taste responses in the CT and sensitivity to amiloride. All other taste responses were similar to that of littermate controls. We found that removal of ENaC mediated neural activity in adulthood resulted in significant reorganization of mature gustatory afferent terminal fields in the NST. Specifically, CT and GSP terminal fields were 1.4x and 1.6x larger than age matched controls, respectively. Unlike when ENaC was deleted from the tongue throughout development, there were no group-related changes in the IX terminal field. Therefore, the expansion in terminal fields follows the sensitivity of the respective taste receptor cells to NaCl and amiloride. These results suggest that the gustatory system remains plastic even following maturation of gustatory circuitry in adulthood.

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#111

Chorda Tympani Terminal Fields in Adult Rats are Greatly Diminished Following Neonatal Chorda Tympani Transection

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Taste afferents project to the nucleus of the solitary tract (NTS) where their terminal fields overlap substantially, creating the potential for competition between nerves. The organization of these terminal fields show a great deal of plasticity - changing as the result of early dietary manipulations or nerve transection. We have previously found that after rats receive transection of the chorda tympani nerve (CTX) at postnatal day 5 (P5), the glossopharyngeal (GL) and greater superficial petrosal (GSP) nerves show increased terminal field volumes in the dorsal zone of the NTS. However, it is not clear how CT projections are affected by P5 transection, and whether changes in CT terminal field organization complement GL and GSP reorganization. Further, since the CT does not regenerate after neonatal transection, it is unclear whether any of its brainstem projections remain intact. To determine the extent of CT reorganization after early denervation, the CT was labeled in adult rats that underwent unilateral CTX at P5 or sham surgery at P5 or P10. In neonatal rats, the CT was accessed anterior to the tympanic bulla and either transected or left intact. Fifty days later, the ipsilateral CT was labeled in the tympanic bulla with biotinylated dextran amine. Terminal fields were then traced in horizontal brain sections. Results indicate a significant and substantial loss of terminal field volume after early CTX ($p < .01$). Terminal field volumes for the dorsal, intermediate, and ventral zones after early CTX were 6%, 9%, and 13% of control animal volumes, respectively. Thus, the highest percentage of terminal field loss occurred in the same region where the GL and GSP terminal fields expand. These results suggest that GL and GSP terminal fields may expand as a result of decreased competition from CT fibers. CT terminal field volumes following transection at P5 are much smaller than those observed after CTX in adults, highlighting the greater sensitivity of the taste system to injury during development.

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#112

Microglia Response to Chorda Tympani Transection in Adult and Juvenile Rats

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Chorda tympani transection (CTX) is a commonly used model of injury-induced neuroplasticity in the taste system. Structural and functional effects of CTX on the gustatory system, in terms of both severity and duration, appear to be inversely correlated with age (e.g., Sollars, 2005). Following CTX, neonatal and juvenile rats (< 25 days of age, P25) demonstrate severe and permanent loss of fungiform taste buds, corresponding changes in fungiform papillae structure, and the CT fails to fully regenerate. In contrast, adult (P40 or older) rats exhibit a nearly complete recovery of both structure and function of taste buds and associated structures. The mechanisms underlying the differing effects of CTX across development remain unclear. Microglia, a CNS immune cell, has been shown to be substantially activated following CT injury in adult mice (Bartel & Finger, 2013). The current study examined whether microglia response to CTX in juvenile and adult rats shows an age-dependent differential activation. To test this, unilateral CTX was performed in P25 (juvenile) and P50 (adult) rats. Four days after surgery, horizontal sections (40 μm) of brainstem were sectioned on a vibratome and immunohistochemistry for Iba1 positive microglia was conducted. Microglia counts of all Iba1 positive cells in the rostral nucleus of the solitary tract (NTS) were obtained from both the intact and surgical sides of the brain. A significant increase in Iba1 positive microglia was found after P50 CTX ($p < .05$), with approximately double the numbers of microglia on the surgical side as compared to the intact side. Preliminary data indicate that this increase in Iba1 positive microglia was not exhibited following P25 CTX ($p > .05$). These findings suggest that differences in microglia activation in gustatory areas of the NTS occur across development, such that a higher degree of activation accompanies a greater degree of post-surgical recovery.

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#113

Over-expression of BDNF in the Mouse Olfactory Bulb does not Increase Adult-born Granule Cell Survival in vivo

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Brain-derived neurotrophic factor (BDNF) mediates CNS neuron maturation, dendritic growth and spine formation/plasticity during development, as well as in adulthood. Prior studies have show that increasing BDNF levels in the adult rodent subventricular zone (SVZ) increases the generation and survival of cells that migrate as neuroblasts to the olfactory bulb and develop into granule cells (Zigova et al., 1998; Bath et al., 2008). However, other studies using knockout of the BDNF receptor, tropomyosin receptor kinase B (TrkB), in adult-born neuroblasts have shown that lack of TrkB/BDNF signaling has no significant effect on the survival of adult-born olfactory granule cells (Galvão et al., 2008; Bergami et al., 2013). The aim of the present study was to determine if increasing endogenous BDNF levels in the olfactory bulb, rather than the SVZ, promotes enhanced survival of adult-born granule cells as they mature and integrate into the granule cell layer (GCL). We employed adult transgenic mice that express a BDNF transgene under the alpha-CamKII promoter. Granule cells express alpha-CAMKII, resulting in increased BDNF expression throughout the GCL, where the new cells become incorporated. Increased bulbar BDNF expression was confirmed by ELISA, and mice were treated with bromodeoxyuridine (BrdU) to label dividing progenitor cells in the SVZ. Mice survived for 2, 4, or 9 weeks. Bulb sections were processed for immunofluorescence and confocal microscopy to quantify Brdu+/Neu+ neurons in the GCL at each time point. Potential changes in SVZ cell proliferation also were examined using Ki-67 immunostaining, and levels of apoptotic cell death in the granule cell layer were assessed with TUNEL labeling. No significant differences in the numbers of BrdU+/Neu+ granule cells, Ki-67+ SVZ cells, or TUNEL+ bulb cells were found between transgenic and wild-type control mice. Our findings support the view that while BDNF has significant morphological effects on granule cells in vivo (Bergami et al., 2013; McDole et al., 2015), it has little impact on the survival rate of new granule cells that mature within the adult bulb.

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#114

Cyclophosphamide-induced Loss in the Murine Olfactory Systems

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Chemotherapy patients often experience profound chemosensory changes during and after drug therapy. Cyclophosphamide (CYP) is one of the first chemotherapy drugs with known cytotoxic and destructive effects, specifically on the taste cell cycle. **Similar to taste, the sense of smell is dependent on olfactory neurons that undergo replacement. Therefore, we asked how much a single injection of CYP would affect olfactory neurons?** Due to a lack of knowledge on how CYP affects olfactory neurons, we examined the effects of CYP on neurons in the main olfactory epithelium (MOE) and the vomeronasal organ (VNO). We used an antibody to Ki67, a protein expressed solely in cells undergoing division. ~100 male mice were given a single, IP injection of CYP (75 mg/kg) and sacrificed from 1 day to 125 days' post-injection. Mice were perfused with 4% paraformaldehyde, decalcified with EDTA, cryo-protected, sectioned and incubated with a Ki67 antibody (Thermo scientific). There were clear differences between MOE and VNO across all the time points. At 1 day post injection, the MOE looked damaged, especially in the dendritic region while the VNO was structurally unaffected. Both tissues showed a decrease in Ki67 protein label compared to controls. By day 2, neither tissue showed any Ki67 labeling. Between days 4 and 6 post injection there was a surge in Ki67 labeling, but this dramatically decreased by day 14. Recovery appeared complete by 30 days. However, Ki67 labeling was decreased again at day 45 and was almost absent at days 60 and 90 compared to age-matched controls. Ki67 labeling began to rebound by 105 days and appeared normal at 125 post injection. Our data suggest that olfactory tissue in the MOE was more affected by CYP than the VNO. However both regions showed long term cyclic depression in Ki67 labeling.

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#115

Disruption of Glomerular Targeting after Epithelial Lesion is Receptor Dependent

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In mice, each set of olfactory receptor (OR)-defined olfactory sensory neurons (OSNs) converge their axons and synapse within an average of 2 to 3 glomeruli per olfactory bulb (OB). The maintenance of this map is crucial for normal olfactory function. Although the robust regenerative capacity of the olfactory epithelium (OE)(including spatially-appropriate restoration of OR-defined OSNs) is well-established following injury, evidence suggests that glomerular targeting may be inaccurate and aberrant during re-innervation, thereby causing long-lasting olfactory dysfunction. Thus, disorganization in the projection from the OE to the OB is suggested in humans by the common complaints of phantosmia and parosmia during recovery from head trauma and other epithelial insults. We have previously reported significant errors in glomerular targeting after recovery from methyl bromide (MeBr) gas induced epithelial injury using the P2-ITL mouse line, which genetically labels P2 receptor-expressing OSNs. These abnormalities include both incomplete and multiple re-innervation of glomeruli. In order to assess the degree of targeting error after recovery from OE lesion, we utilized the receptor-tagged mouse lines M72-ITG and I7-ITG as well as P2-ITL. Left sided naris occlusion was performed in mice (n = 3) from each of the three lines prior to MeBr exposure to create unilateral lesions with an internal control. Nasal plugs were removed and the mice recovered for 8 weeks. OB sections were analyzed for glomerular targeting including number and completeness of glomerular re-innervation by comparison with olfactory marker protein (OMP)- staining. We found the degree of disruption in targeting was dependent on receptor type with P2 innervating the largest number of glomeruli (average = 7.5, 4.5, and 1.5 for P2, I7, and M72 respectively). The results are important in extending our understanding of the regenerative capacity of the olfactory system beyond OE recovery. In addition, our findings indicate the need to analyze multiple OSN types to provide a more complete assessment of recovery after lesion.

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#116

Subfunctionalization and Neofunctionalization of Drosophila Odorant Binding Proteins

Don Tucker Award Finalist

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The functions of most Drosophila odorant binding proteins (Obps) remain unexplored, and many exist in tandem arrays throughout the genome. As these genes most likely arose through recent duplication, genes within a cluster likely have partially redundant or pleiotropic functions. Here, we used the CRISPR -Cas9 system to generate two knock-out lines, the first lacking the four paralogs of the *Obp56a-d* cluster, and the second lacking the single *Obp56h* gene, another possible paralog of the *Obp56* cluster. Various phenotypic tests on these knockout lines demonstrate significant functional overlap, as well as novel pleiotropic functions. Both lines shared increased activity level, variability in development time and incidence of morphological abnormalities, as well as decreased viability in early development, development time, and attraction to food following starvation. The *Obp56a-d* KO line uniquely showed decreased height of pupation while the *Obp56h* KO line showed increased copulation duration and decreased aversion to 2-heptanone. Reinserting the *Obp56a-d* genes one-by-one and in various combinations in a PhiC31 integration site engineered in their original location during CRISP-Cas9 excision will enable reconstruction of their functional evolutionary history. Supported by NIH grant GM059469.

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#117

Sensory Physiology and Olfactory Behavior in Drosophila mojavensis

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Many organisms gain information from their external environment through chemical cues. Variation in responses to the chemosensory environment, such as the olfactory preference of insect herbivores for host plants, may result in differential reproductive fitness, and shifts in olfactory preference may contribute to reproductive isolation. Such disparities in olfactory behavior can reflect alterations in the genetic and neuronal underpinnings of the animals' sensory system. The fly *Drosophila mojavensis* feeds and breeds on cacti in arid regions of NW Mexico and the SW US. Four geographically distinct populations currently exploit four different cactus species that emit specific combinations of volatiles, and these serve as primary cues for host plant identification. Preliminary data show divergence among the populations in olfactory electrophysiology, behavioral preferences to cactus volatiles, odorant receptor gene expression, and in olfactory sensory neuron number. These results demonstrate that the peripheral nervous system has changed in response to different ecological environments and that these alterations contribute to population divergence.

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#118

Functional Characterization of Differential Alleles Associated with Variation in Drosophila Olfaction

Don Tucker Award Finalist

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Drosophila chemoreceptors have been studied extensively but little is known about genes regulating variation in higher-order processing of olfactory behavior. Previously, genome-wide association analysis in the *Drosophila melanogaster* Genetic Reference Panel identified candidate genes associated with variation in olfactory response to benzaldehyde which might be involved in higher-order processing of olfactory behavior. We identified polymorphisms in promoter regions of nine of these genes that show differential expression of a luciferase reporter gene *in vitro*. We then made transgenic flies in which these promoters (reference and alternative allele versions) drive the expression of Gal4. This enables cell-specific expression of fluorescent reporter genes and transgenes that activate or inhibit neuronal activity under UAS enhancers using the binary GAL4-UAS expression system. The promoter upstream of *Patj* drives GFP expression in distinct neurons in the adult central nervous system, emphasizing its putative role in higher-order processing of olfactory behavior. Results from these experiments will delineate the neural circuitries in which expression of these genes is associated with variation in olfactory information processing and can assess to what extent differential promoter activity of each of these constructs in defined genetic backgrounds may affect differences in olfactory behavior.

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#119

Non-classical Amine Recognition Evolved in a Large Clade of Olfactory Receptors

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Biogenic amines are important signaling molecules, and the structural basis for their recognition by G Protein-Coupled Receptors (GPCRs) is well understood. Amines are also potent odors, with some activating olfactory trace amine-associated receptors (TAARs). Here, we report that teleost TAARs evolved a new way to recognize amines in a non-classical orientation. Chemical screens de-orphaned eleven zebrafish TAARs, with agonists including serotonin, histamine, tryptamine, 2-phenylethylamine, putrescine, and agmatine. Receptors from different clades contact ligands through aspartates on transmembrane a-helices III (canonical Asp$^{3.32}$) or V (non-canonical Asp $^{5.42}$), and diamine receptors contain both aspartates. Non-classical monoamine recognition evolved in two steps: an ancestral TAAR acquired Asp$^{5.42}$, gaining diamine sensitivity, and subsequently lost Asp$^{3.32}$. Through this transformation, the fish olfactory system dramatically expanded its capacity to detect amines, ecologically significant aquatic odors. The evolution of a second, alternative solution for amine detection by olfactory receptors highlights the tremendous structural versatility intrinsic to GPCRs.

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FCOI Declarations: None.

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#120

Expression of Candidate Fat Taste Receptors in Human Fungiform Papillae

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An increasing body of evidence from humans and animal models suggests the existence of a taste modality associated with fat via their breakdown products, fatty acids. The peripheral mechanism associated with fat taste has not been established. There have been several candidate taste receptors and ion channels associated with fat taste, including CD36, GPR120, GPR40, GPR41, GPR43, GPR84, and several types of delayed rectifying potassium (DRK) channels, with most of them identified due to their high affinity for free fatty acids *in vitro*. The expression pattern of the fat taste receptors in human taste tissue is the basis for the understanding of fatty acid perception mechanism. Here we report the expression of the candidate fat taste receptors and the associated taste cell type in human fungiform papillae. RNA and protein samples were extracted from the fungiform papillae biopsies collected from eight participants (5 females and 3 males, age 25-50) using TRIzol isolation. Through the real time reverse transcription polymerase chain reaction (RT-PCR) and western blotting analysis, we examined the expression of mRNA and protein. The double-staining immunohistochemistry was then applied to locate the expressed receptors within specific taste tissue in human fungiform papillae. As a result, CD36, GPR120, GPR43, GPR84 and several types of DRK channels such as KV1.2, KV2.2 were identified in taste cells embedded in human fungiform papillae. The study shows a large difference of the expression pattern of the candidate fat taste receptors between humans and animal models and provides a solid ground for the further study exploring oral perception of fatty acids.

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FCOI Declarations: None.

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#121

Taste Cell Responses to Medium Chain Saturated Fatty acids are Mediated by GPR84

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Previous research has shown that GPR84 is a novel GPCR activated by medium-chain saturated fatty acids (MCFAs) of C_9 to C_{14} in length (Wang et al., J Biol Chem 281: 34457, 2006). We have used cellular, molecular and behavioral assays to investigate MCFA transduction in mouse taste cells and found that GPR84 plays an essential role in this process. RT-PCR and qPCR showed that the mRNA of GPR84 is highly expressed in taste cells. Using whole cell patch clamp recording, we found that capric acid (C_{10}) induced an inward current in wild type taste cells at resting potentials, and this current was significantly reduced in taste cells from GPR84 knockout mice. The calcium response to capric acid in GPR84 knock out taste cells is significantly smaller than the wild type taste cells. In order to determine the mechanisms underlying MCFAs transduction pathway, antagonists of several important players that might be involved in this pathway were used in electrophysiological experiments. Capric acid-induced currents were significantly inhibited by GDP-β-S and inhibitors of protein kinase A (PKA), which suggested that the currents were activated downstream of G protein activation and PKA. However, these currents were not affected by the PLC and PDE inhibitors, suggesting they are not involved in the MCFA taste transduction pathway. Our results are consistent with MCFAs being sapid compounds that work through a pathway that is independent of the well-documented pathway for long chain unsaturated fatty acids involving TrpM5 (Liu et al., J Neurosci 31: 8634, 2011) and suggest increasing complexity to mechanisms underlying the 'taste of fat'.

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#122

ß-Estradiol Modulates Responses to Fatty Acids and Saccharin in a Clonal Taste Cell Line

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Accumulating research suggests that there are sex differences in food preferences. It has been hypothesized that reproductive hormones play a role in these differences. Estradiol, a sex hormone found primarily in females, is thought to partake in helping shape taste preferences. It has been shown that 17β-estradiol (E2) plays an important role in intracellular calcium fluxes in many cell types. The aim of this study was to elucidate the effects of E2 on Taste Bud Derived C1 cells (TBD-C1), a clonal taste bud cell line. TBD cells have been shown to express various indicators of taste receptor cells (Sako et al., 2011) and respond to various tastants. Specifically, we have detected G protein-coupled taste receptors for sweet (*T1R2, T1R3)* and fat (*GPR120*) in TBD-C1 cells. It is widely accepted that gustatory signals mediated by these receptors lead to increase in intracellular calcium concentrations. We characterized the effects of a wide range of concentrations of linoleic acid, a polyunsaturated omega- 6 fatty acid (3, 10, 30, 100 μM) and saccharin (2, 5, 10, 25 mM) on fura-2 loaded TBD-C1 cells. To date, the data suggest E2 enhances fatty acid and sweet responses when presented acutely thus leading to a significant increase in $[Ca^{2+}]_i$ when compared to fatty acids and saccharin stimulation alone. However, longer treatments with E2 elicits the opposite response. Long-term incubation with E2 may be altering activity within the cellular signaling pathway contributing to the transduction of fatty acids and saccharin by directly modulating transcriptional events within TBD-C1 cells. We are currently exploring both acute and long-term treatments with estrogen in taste cells.

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#123

Of Mice and Light

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Type III taste cells in the mammalian taste bud are necessary to generate a nerve response to sour and some salty stimuli, whereas Type II cells respond selectively to sweet, bitter, or umami taste stimuli. Type II and Type III cells may also interact within the taste bud to modulate the output signal from the taste bud. Studies of Type III cell function in the context of the taste bud are difficult due to off-target effects of sour stimulation because applying acid (sour) stimuli to intact or semi-intact taste buds causes intracellular acidification in all cells, not just Type III cells. To avoid non-specific acidification and/or direct activation of other cell types, we have developed a mouse that expresses light-activated channel Channelrhodopsin (ChR2) under a Type III-specific genetic Cre driver. In this mouse, an IRES-Cre recombinase construct follows the Polycystic Kidney Disease 2-Like 1 (*Pkd2l1)* stop codon. PKD2L1 is expressed exclusively in Type III cells, and drives ChR2 expression successfully in taste buds across the tongue. Optogenetic activation of Type III cells in the anterior tongue of an anesthetized, *Pkd2l1*-Cre, ChR2 mouse generates a nerve response in the chorda tympani nerve, which innervates anterior tongue. This nerve response is both repeatable and consistent, and allows for specific activation of Type III cells without acid application. With this novel tool, we can examine whether activation of Type III cells in the absence of overall acidification of the epithelium modulates responses of the chorda tympani nerve to taste qualities mediated by Type II cells, which would support the hypothesis that taste buds function as integrators of taste information.

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#124

The Role of Alpha-gustducin and ITPR3 in Salty Taste in Mice

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The involvement of Type 2 taste cells in the transduction of salty taste is suggested by a recent finding that knockout of calcium homeostasis modulator 1 (CALHM1) influences NaCl acceptance and preference of mice. This is because CALHM1 is localized specifically in Type 2 cells, where it is responsible for releasing ATP, the final step of G protein-coupled receptor (GPCR)-mediated taste transduction. Here, we used mice with knockout of other proteins found in the GPCR-mediated taste transduction cascade in Type 2 cells to molecularly dissect this salty taste transduction pathway. In 48-hr two-bottle choice tests, mice with knockout of alpha-gustducin (*Gnat3*) had normal preferences for NaCl, whereas mice with knockout of *Itpr3* had diminished preferences, akin to the *Calhm1*KO mice. Multiunit recordings from the chorda tympani (CT) nerve were made in alpha-gustducin knockout mice. Taste-evoked CT response sizes showed a progressive increase as the concentration of NaCl flowed over the tongue increased from 56 to 1000 mM, which suggests that alpha- gustducin is not essential for normal transduction of high concentrations of NaCl. Overall, our data indicate that high concentrations of NaCl activate gustducin-independent components of the G- protein-mediated transduction pathway found in Type 2 taste cells.

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FCOI Declarations: None.

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#125

Spilanthol Modulates Sensitivity to Sodium in Mouse Taste Bud Cells

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Unsaturated alkylamides induce tingling and cooling sensations (Sugai et al, 2005; Albin and Simons, 2010) in the mouth. These compounds also inhibit 2 pore potassium channels, depolarizing cells (Bautista, et al., 2008). Whether these alkylamides can regulate sensitivity to sodium in mouse taste bud cells is an open question. Using fluorescence calcium imaging, we found that micromolar concentrations of spilanthol, an unsaturated alkylamide, significantly enhanced the responses of taste cells to 140 and 200mM in taste bud cells, but not trigeminal neurons. We used potassium depolarization, TrpM5-GFP, T1R3-GFP mice and immunohistochemical method to further identify taste cell types and found that the most common taste cells enhanced by spilanthol are type III cells and few type II cells. Pharmacological experiments suggest that one or several of the KNCK family of potassium channels is/are involved in the enhancement. The enhancement can be significantly reduced by the reduction of extracellular calcium. These results suggest that saltiness may be enhanced by unsaturated alkylamides.

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#126

Action Potentials in CALHM1-KO Type II Taste Bud Cells

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Taste bud type II cells fire action potentials (APs) in response to taste substances that trigger non-exocytotic neurotransmitter release. Recently, we showed that CALHM1 is specifically expressed in type II cells and acts as a voltage-gated ATP-release channel required for sweet, bitter and umami taste perception (Nature, 2013). CALHM1 also regulates cortical neuron excitability (PNAS, 2012), suggesting that it may modulate action potential firing in type II cells. Here, we investigated the properties of APs in TRPM5-GFP-expressing type II cells in the circumvallate papillae isolated from wild-type (WT) and CALHM1-KO mice. We first determined that small residual inward tail currents previously observed in CALHM1-KO type II cells (Nature, 2013) were absent in the present study, suggesting that they were probably due to non-specific leak and/or contamination by other currents. Thus, CALHM1 contributes to all of the non-selective voltage-gated current in type II cells. Surprisingly, deletion of CALHM1 does not significantly alter AP properties (thresholds, overshoots and half-widths), resting membrane potential (~ - 65 mV) or cell membrane input resistance (R_m). Carbenoxolone (CBX), a connexin and pannexin inhibitor, previously shown to inhibit tastant-evoked ATP release (Huang et al., PNAS, 2007; Murata, et al. J Neurophysiol., 2010), does not inhibit CALHM1-associated currents in type II cells. Instead, we have found that 50 uM CBX reduces outward K^+ currents by >50% in CALHM1-KO type II cells, reducing the amplitude of the action potential after-hyperpolarization, causing Na^+ channel inactivation and reducing the number of APs fired.

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#127

In vivo Juxtacellular Labeling of Rat Geniculate Ganglion Neurons Demonstrates Both Peripheral and Central Processes

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Primary afferent neurons of the chorda tympani nerve (CT) convey information to the central nervous system about sensory properties of food. CT fibers have cell bodies in the geniculate ganglion (GG) that respond to chemical, thermal and mechanical properties of oral stimuli. Despite the obvious importance of their neurobiological and sensory roles, there is no comprehensive understanding of the basic biology of GG neurons. We have recorded *in vivo* from GG neurons during natural stimulation of the tongue with a wide battery of stimuli, and measured receptive field size, sensory response properties, and fiber latency and conduction velocity. Our results add to demonstrated heterogeneous GG properties while detailing differences in receptive field characteristics, conduction velocity of the afferent fibers and additional response properties that include sensitivity to cooling stimuli. Details of termination of individual GG neurons in the rostral nucleus of the solitary tract (rNST) remain unknown. We hypothesize that single GG neurons with known neurobiological characteristics have a specific termination pattern. To identify the brainstem terminal projections of single neurons, we applied the technique of juxtacellular labeling to the GG neurons. The GG was exposed using a ventral approach and a glass extracellular recording electrode containing Neurobiotin was used to isolate a single neuron. Once isolated, current was passed through the electrode to iontophorese the label into the neuron. After a post-ejection survival period the rats were perfused with phosphate-buffered paraformaldehyde, the GG and brainstem removed and horizontal sections prepared. To date we have successfully labeled 23 GG neurons. Of these the label filled either the central or peripheral process, but recently we have filled both the central and peripheral processes and are currently tracing the central termination patterns in rNST. With juxtacellular labeling of GG neurons we have the ability to define central termination details of taste ganglion cells.

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FCOI Declarations: None.

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#128

The Interglomerular Circuit Directly Inhibits Mitral/tufted Cells

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Sensory processing shapes our perception of the external world. In the mammalian olfactory system, odor information is encoded by combinatorial activity patterns of olfactory bulb (OB) glomeruli, the earliest stage of synaptic processing. Glomeruli are richly interconnected by short axon cells (SACs), which form the interglomerular circuit (IGC). It is still not clear how the IGC impacts MTC output to downstream neural circuits. We addressed this question by combining *in vivo* and *in vitro* electrophysiology with optogenetics and found that: (1) direct, monosynaptic IGC-MTC inhibition is mediated by GABA release from SAC-MTC synapses, (2) gap junction-mediated electrical coupling is strong for the SAC-MTC synapse but negligible for the SAC-ETC synapse; (3) brief IGC-mediated inhibition is amplified and temporally prolonged by the intrinsic properties of MTCs, lasting hundreds of milliseconds to seconds; (4) IGC mediated inhibition evokes negligible rebound excitation in MTCs but strong rebound firing in ETCs; (5) sniff frequency IGC activation *in vivo* generates persistent MTC inhibition. Thus, brief sensory input activates the IGC, which suppresses MTCs in neighboring glomeruli for hundreds of milliseconds, reducing their response to subsequent input. This may ensure that when it comes to glomerular activation and impact on downstream olfactory networks, *"the early bird gets the worm"*.

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#129

Receptors for ATP, 5HT and GABA are Differentially Expressed and Functional in Geniculate Ganglion Neurons

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Taste cells synthesize and release neurotransmitters, including ATP, 5HT and GABA, which act in paracrine fashion on neighboring taste bud cells. ATP, secreted from Type II taste cells following taste stimulation, acts on afferent terminals via P2X2 and P2X3 receptors. However, the roles of 5HT and GABA in signaling to afferent neurons are not as well understood. Previous studies have shown that geniculate ganglion neurons that innervate taste buds express receptors for 5HT and for GABA. Thus, we asked if ATP, 5HT and GABA act on separate classes of afferent neurons. We addressed this question using RNAseq, single-cell RT-qPCR and Ca^{2+} imaging on acutely isolated geniculate ganglion neurons. Using RNAseq and RT-qPCR, we confirmed that the most prominently expressed receptors are *P2rx2*, *P2rx3*, *P2ry1* for ATP; *Htr3a*, *Htr1d* for 5HT; and *Gabara1*, *Gabarb2*, *Gabarb3*, *Gabarg1*, *Gabarg2* for GABA. To assess if individual neurons express receptors for all 3 transmitters, we used single-cell RT-qPCR. 20% of the cDNA from each of 29 neurons was used for each PCR for receptors and for Snap25 as a normalization control. 27 of 29 neurons (93%) neurons expressed Gabra1, an essential subunit of most ionotropic GABA-A receptors. Regarding receptors for 5HT and ATP, 12 of 29 (41%) of neurons expressed both Htr3a and P2rx2/3; 9 of 29 (31%) neurons expressed only P2rx2/3; 6 of 29 (21%) neurons expressed only Htr3a. To assess functional patterns, we used Ca^{2+} imaging to examine transmitter-evoked responses in geniculate ganglion neurons isolated from mice in which sensory neurons express the Ca^{2+} reporter, GCaMP3. In aggregate, of 102 neurons, 39 (38%) responded to both 10 μM ATP and 10 μM 5HT, 57 (56%) responded only to ATP and 6 (6%) responded only to 5HT. A smaller subset of neurons was also tested for GABA; the majority displayed GABA (100 μM)-mediated inhibition of KCl-depolarization. Thus, both RT-qPCR and Ca^{2+} imaging demonstrate that geniculate ganglion neurons display functional groupings by responding to ATP, 5HT or both, and that the large majority are also responsive to GABA.

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#130

Palatability of Three Basic Tastes in a Cross-species Approach

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Taste sensitivities across mammalian species are thought to depend on feeding strategies shaped by evolution. A growing body of evidence obtained from wild species occupying different environmental and behavioral niches supports this notion. Yet surprisingly little has been documented on the taste sensitivities of domestic cats and dogs, companion animals of substantial social and commercial significance. Consequently we have conducted a series of palatability tests focusing on 3 basic taste categories (salty, sour, and bitter) in cats and dogs, and compared the results to those obtained in similar studies of an omnivorous mammal animal model—*Rattus norvegicus*. Twelve tastants, 4 from each basic taste category, each were examined at 3 concentrations. Palatability in cats and dogs was evaluated by monadic consumption of tastant solutions relative to water, and in rats by a high throughput operant taste system. In the salty category, NaCl was appetitive, whereas K_2SO_4 was aversive, to all 3 species, but at lower concentration for rats (30 mM) compared to cats and dogs (100 mM). Other ionic compounds tested in this category (KCl, and NH_4Cl) were aversive to rats but had no effect on cats and dogs. Therefore, rats seem to be more sensitive to salty tastants than cats and dogs. Among the sour stimuli, organic acids (citric, lactic, and ascorbic) were aversive at 100 mM to all species. In contrast the inorganic phosphoric acid was neutral to rats at all concentrations, appetitive to cats at 1 mM, and aversive to both cats and dogs at 10 mM. Species-specific differences were most evident in the bitter category—although quinine was commonly rejected at all concentration tested, responses (whether aversive, neutral, or appetitive) to denatonium benzoate, naringin, and L-phenylalanine were observed to be both species- and concentration-dependent. Together, our data reveal for the first time some commonalities in the taste sensitivities across cats, dogs and rats, especially regarding sour tastants, that could be relevant to the development of food for companion animals.

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#131

Oromotor Taste Reactivity Measured during a LiCl-induced State Reveals the Rapid Formation and Retention of a Conditioned Taste Aversion in Rats with Lesions in a Subregion of Insular Cortex Implicated in Taste Aversion Learning

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Using a new lesion mapping system, we showed that insular cortex lesions (ICX) encompassing the posterior half of the traditionally defined gustatory cortex (GC) and adjacent portions of visceral cortex (VC) attenuated the expression of a conditioned taste aversion (CTA) as assessed by 1- and 2-bottle retention tests administered days after the last conditioning trial. To begin to discern whether the effect was due to an acquisition (taste-visceral processing) and/or memory deficit, we used a serial taste reactivity (TR) paradigm to assess the aversion as it was acquired, followed by a TR retention test 3 days later. Ibotenic acid (ICX) or PBS (SHAM) was injected in the IC, and intraoral (IO) cannulae were implanted. During conditioning, rats received 30-s IO infusions of 0.3 M sucrose every 5 min (for 45 min) starting directly after injection of 2.0 mEq/kg lithium chloride (Li) or sodium chloride (Na). TR during the IO infusions was video-recorded and later scored. Whereas Na-injected SHAM (N=6) and histologically confirmed ICX (N=6) rats displayed comparably high levels of ingestive TR and almost no aversive TR throughout conditioning, Li-injected SHAM (N=9) and ICX (N=10) rats systematically reduced ingestive TR while increasing aversive TR across the conditioning session. For the retention test, all groups were first injected with Li and then TR to IO sucrose was assessed 20, 25, and 30 min later. SHAM and ICX rats that were originally injected with Na continued to display high levels of ingestive TR, with virtually no aversive TR. By comparison, SHAM and ICX rats originally injected with Li exhibited significantly less ingestive and more aversive TR, providing evidence that a CTA was retained, even in ICX rats. These data suggest that this region of IC is not necessary for the formation of, or memory for, a CTA, as measured in this TR paradigm. The nature of the disparity between traditional intake versus TR tests on CTA expression following these cortical lesions may relate to the class of response measured and/or presence of Li during the conditioned response assessment and may provide clues as to the functional significance of IC.

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#132

Olfactory Preferences are Just a Matter of Taste: Retronasal Learning Requires taste Cortex

Don Tucker Award Finalist

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The mammalian olfactory system operates dually: odors in the environment are sampled through nasal inhalation (called orthonasal), and odors present in the mouth are sampled through exhalation and mastication (retronasal). Understanding the different kinds of olfactory information collected through each olfactory mode is important for understanding flavor perception and food seeking behavior. In this work, we ask whether the mode of odorant experience affects the acquisition of olfactory preferences in rats. Animals are trained to associate one of two ortho- or retronasally delivered odors with a sweet-water reward using a highly controlled olfactometer system. We then optogenetically inactivate cortical taste area gustatory cortex (GC) during testing to assess the involvement of the taste system in the expression of olfactory preference. We found that retronasal presentation of odors rapidly induces odor preference, and that this preference is specific to the modal context in which it occurs; retronasally learned preferences are *not* expressed if animals are tested in the orthonasal olfactory mode. However, orthonasal preferences are "rescued," and even expressed *more strongly* than retronasally learned preferences, if learning includes both a retro- and orthonasal component. This retro- and retro-facilitated preference expression is eliminated during GC optogenetic inactivation. Taken together, these findings suggest that retronasal-taste associations occur directly and rapidly, while orthonasal-taste associations occur more slowly or indirectly through retro-facilitated *second order learning*. GC is required for this expression of retro- and retro-facilitated odor preferences *even in the absence of taste stimulation*, highlighting the intrinsic connection between taste and retronasal smell as orally-sourced chemosensory streams.

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#133

The Experienced Palette is a Smart One: Investigating the Role that the Gustatory Cortex Plays in the Impact of Experience on Taste Learning

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In conditioned taste aversion (CTA), an animal learns to avoid a particular taste that has been paired with malaise. Familiarity with future conditioned taste stimuli (CS) is known to influence the strength of aversion learning; the effect of familiarization with innocuous tastes—tastes other than the CS—has received little investigation, however. Our previous work demonstrated that pre-exposure to salty and sour tastes strengthened a later learned aversion towards novel sucrose, and showed that the phenomenon scaled with both the number of tastes in the pre-exposure array and with the number of pre-exposure sessions. The present studies begin an inquiry into the neural underpinnings of this phenomenon, using c-FOS, optogenetics, and (in future work) electrophysiology. The use of c-FOS enabled us to survey many possible relevant brain sites, but inquiry focused on gustatory cortex (GC), which is known to be involved in the integration of experience with taste behavior. Rats that had been pre-exposed to innocuous tastes demonstrated higher levels of c-FOS in GC after exposure to novel sucrose, compared to animals that had been pre-exposed to water alone. Furthermore, optogenetic inhibition of GC during pre-exposure sessions reduced the magnitude of a later CTA to novel sucrose, rendering taste pre-exposed rats identical to rats exposed to water alone. These results suggest that GC plays a controlling role in determining the impact of innocuous experience on learning-related behavior.

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#134

Elucidating Mechanisms Underlying Odor-Mediated Memory Consolidation in the Human Brain

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Odors have been shown to be key players in targeted memory reactivation (TMR), a technique used to manipulate memory consolidation. During olfactory TMR, subjects learn a memory task in the presence of an odor, and then the same odor is presented during a subsequent period of sleep or wakefulness. Reactivating memories during non-REM sleep often *improves* memory performance for the associated task upon waking, while reactivating memories during wake often has no effect or even *worsens* memory performance in paradigms involving memory interference. Despite intriguing behavioral outcomes, the neurobiological mechanisms through which odors influence memory consolidation remain unclear. Here, we developed a novel olfactory TMR paradigm to probe the neural mechanisms underlying olfactory TMR. In our paradigm, subjects learn spatial locations of pictures from different categories during functional magnetic resonance imaging (fMRI). During learning, pictures are paired with distinct category -specific odors, which serve as cues for subsequent reactivation. Then, subjects undergo reactivation during fMRI scanning. Thus far, results have been collected from 5 subjects that underwent reactivation during wakefulness. Our preliminary findings convincingly show that each picture category is associated with a unique fMRI pattern "signature" of neural activity across voxels in visual cortex during learning. Behaviorally, memory performance for reactivated categories declined compared to non- reactivated categories following memory interference. These preliminary findings suggest that reactivating memories during wakefulness influences memory consolidation in a category-specific way, rendering reactivated picture categories vulnerable to memory interference. Further data will be collected for subjects undergoing reactivation during both sleep and wakefulness, and ongoing fMRI analyses will investigate neural activity during reactivation across both conditions.

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#135

The Impact of Chemosensory Cues of Anxiety on Fear Perception – An fMRI Investigation in Healthy Individuals and Autism Spectrum Disorder Patients

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Recent evidence suggests that the experience of stress can be communicated between individuals via chemosensory cues. Nonetheless, the neural networks engaged in the interaction of processing of socio- emotional stimuli and chemosensory cues remain largely unknown. Moreover, no study to date has looked at the effects of anxiety chemosensory cues on social cognition in Autism Spectrum Disorders (ASDs). Axiliary sweat gathered on cotton pads collected from 14 male students awaiting an oral examination (anxiety stimuli) and participating in the ergometer training (control stimuli) served as the chemosensory cues. In study 1, 24 healthy, normosmic participants were presented with chemosensory cues of anxiety (versus control cues) and completed a parametrically morphed (neutral to fearful) emotion recognition task during fMRI. In study 2, 17 ASD patients and 17 matched controls completed the emotion recognition task during fMRI. In Study 1, behaviourally, healthy participants rated more discernible facial expressions as more fearful. Crucially, the increased fearfulness of the face corresponded to increased modulation in the left insula and middle occipital gyrus extending to fusiform gyrus under exposure to the anxiety cue. In Study 2, ASD patients were biased towards higher fear perception in neutral faces under exposure to chemosensory anxiety cues, while HC showed higher fear ratings under anxiety cues for more fearful faces (smell x fearfulness level x group interaction). Ongoing fMRI analyses test how the chemosensory cues are processed in the patients at the cerebral level. The current results suggest that chemosensory anxiety cues facilitate processing of socially relevant stimuli in healthy individuals and provide preliminary evidence for differential effects of chemosensory anxiety cues on fear perception in ASD patients, pointing to a potentially important role of anxiety chemosensory signals in promoting fear processing in ASD.

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#136

Human Olfactory Fear Generalization Across Binary Odor Mixtures

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Stimulus generalization serves an adaptive function, allowing us to use prior experience to guide behavior toward newly encountered stimuli. This behavior has been studied in various sensory systems in many species. However, little is known about the nature of generalization in the human olfactory system. Here, we used a fear conditioning paradigm to study olfactory generalization across varying binary odor mixtures. Behavioral discrimination training and generalization test sessions were conducted with 16 subjects, using two sets of binary odor mixtures. In order to account for inter-subject perceptual variability, subjects made perceptual ratings for a set of equally spaced mixtures ranging from one pure odorant to a second pure odorant. Based on these perceptual ratings, we selected two conditioned stimuli, both of which were binary mixtures but one was paired with shock (CS+), and one was never paired with shock (CS-). Subsequently, subjects underwent a discrimination training session entailing the presentation of only the CS+ and CS- mixtures. On each trial, subjects indicated whether the delivered odor was associated with shock. Across both odor sets, learning accuracy reached significantly above chance ($p<0.02$). Subjects returned the next day to perform the main task, which included six generalization test runs. During the test runs, subjects were exposed to a set of perceptually-equally- spaced mixtures (derived from the baseline ratings on Day 1 and excluding the CS+ and CS-) and asked to indicate whether that particular mixture was associated with shock. These responses were used as an index of olfactory generalization. We found a behavioral generalization gradient which peaked near the CS+ in the direction away from the CS- (peak shift) alongside a significant main effect of mixture ($p<0.001$). Our results suggest that generalization occurs behaviorally in a manner similar to that observed in the human visual system (Kahnt, et al. 2012). Future functional imaging analyses will provide insight into the neural mechanisms mediating olfactory generalization.

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#137

Long-Lasting Enhancement of Odor-Evoked Periglomerular Activity after Olfactory Fear Conditioning

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Olfactory fear conditioning can selectively alter the neural representations of odor stimuli as early as the primary sensory input to the brain (Kass et al 2013). This plasticity could be related to learning-induced changes in olfactory bulb glomerular circuitry, which controls sensory input gain and is dynamically regulated by neuromodulatory and cortical networks involved in fear learning. Here, we performed *in vivo* cell type-specific optical neurophysiology to evaluate the effects of olfactory fear conditioning on periglomerular (PG) circuitry in adult mice. Fear conditioning consisted of a single day of training with 10 trials of a ~15 sec odor (the CS^+) paired with a footshock (or 10 shock or odor alone trials for control groups). We compared odor-evoked GCaMP signals in GAD65-expressing PG cells 1 day before, 1 day after, and 1 month after training in each individual mouse, and measured odor-evoked freezing in a novel context at similar post-training time points. During imaging and test sessions, subjects were presented with a panel of 4 odors including the CS^+ and 3 unexposed odors. Fear conditioned mice exhibited stimulus-evoked freezing that generalized across all 4 odors (after non-discriminative conditioning) and that was observed even 1 month after learning, whereas little freezing was observed in control animals. In parallel, we found that fear conditioning resulted in a robust, non-specific enhancement of odor- evoked GCaMP signals in GAD65-expressing PG cells. This generalized enhancement occurred just 1 day after learning and persisted up to 1 month. These data show that fear conditioning causes relatively rapid and long-lasting changes in olfactory coding. Such changes might decrease discrimination between threat-predictive and neutral stimuli and promote generalized anxiety.

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#138

Anxiety Body Odor Induces a Stress Response in the Recipient: Evidence from Recognition of Dynamic Facial Expressions and Heart Rate Variability

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Body odors play a key role in communicating social information. In particular, their effects are maximal when their presence is paired with a meaningful social context. Schematic faces as well as static faces have been widely used as social stimuli. However, they miss a key feature of our phenomenological experience, characterized by multisensory dynamic stimulations. Here we investigate how the body odor of people experiencing a transitory anxiety state vs. the body odor of relaxed people i) biases the recognition of dynamic facial expressions and ii) modulates parasympathetic activity in recipients. Participants hooked up to an electrocardiogram and to an olfactometer categorized the emotion of a face morphing from a neutral expression to either an angry or happy expression. Results obtained via a between-subjects design on 52 participants indicate that smelling the anxiety body odor impairs dynamic facial recognition accuracy, increases the time necessary to produce accurate categorizations, and it reduces the cardiac parasympathetic activity. Altogether, these results suggest that in social situations that simulate the multisensory and dynamic features of real-life social contexts, anxiety body odors will induce in recipients a stress responses impairing both arousal and cognitive-emotional skills.

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#139

Chemosensory Threat Signals: The Case of Strangers' Body Odor

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As for visual stimuli associated with threat, also chemosensory stimuli signaling threats may enjoy prioritized processing. Indeed, smelling the body odor of a stranger activates the cerebral network processing threat-related stimuli and renders the visual processing of a neutral face more similar to that of an angry face as well as it increases general detection performance. These results suggest that the body odor of a stranger acts as a threat signal. To directly test this hypothesis, we investigated whether the body odor from strangers produce a psychophysiological profile comparable to that of a perceptually non-discriminable neutral odor paired with irritating electrical shocks in a classical conditioning procedure. Results indicate that at baseline, participants show significantly increased sympathetic activity (arousal), as measured via skin conductance responses (SCR), in response to exposure to the masked odor of strangers as compared to when they smell the masker odor alone. After conditioning to both odors, there was no statistical differences in SCR between the masked body odor and the masker alone. This indicates that post conditioning, there was no difference in saliency (threat), as operationalized by arousal, between the two odor conditions. Further, a three-alternative forced choice test indicates that post-conditioning the two odor conditions were not consciously discriminable. We conclude that the body odor of strangers is perceived as an intrinsically threatening signal, which is able to alter physiological reactivity in a way compatible with classical conditioning effects, the most used experimental paradigm to elicit "fear-related" behaviors.

Funding Acknowledgements: Supported by the Louise Slade Fellowship to VP and the Knut and Alice Wallenberg foundation (KAW 2012.0141) to JNL.

FCOI Declarations: None.

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#140

POSTER SESSION I
THURSDAY, 8 – 10:30 AM

Blunted Olfactory Function in Combat Veterans with and without PTSD

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Compared to healthy combat veterans (CV-PTSD), we previously reported that combat veterans with PTSD (CV+PTSD) had increased "sensitivity" to burning-related odors but blunted odor-elicited physiological responses and reduced gray matter volume in olfactory cortex. We sought to examine this apparent paradox (e.g. increased odor "sensitivity" with underlying decreases in odor structure and function) by assessing the relationship between burned rubber (BR) odor-elicited brain activation and trauma-related factors. 15 CV+PTSD and 30 CV-PTSD with similar combat experiences underwent clinical olfactory testing (threshold and identification) and an fMRI odor challenge test during which BR, lavender (LAV), and an odorless control were delivered. Compared to published norms, odor threshold was impaired, but to a similar degree, in both groups (CV+PTSD= -4.42; CV-PTSD= -4.23, p>.1), with CV+PTSD showing a trend toward more impaired odor identification (CV+PTSD=33.9; CV-PTSD=36.2, p=.08). Both BR and LAV activated all 1° and 2° olfactory regions of interest (all ps<.05). However, CV+PTSD, but not CV- PTSD, showed blunted activation specific to BR in right anterior piriform (APir), left anterior insula, and left OFC (all Diagnosis X Odor interaction ps<.05). OFC activation was positively related to trauma exposure in CV+PTSD (r =.54, p=0.04), but negatively related in CV-PTSD (r = -.40, p=0.03). Additionally, APir and amygdala activation was negatively and positively related to avoidance/numbing symptoms in CV+PTSD (ps<.05) and CV-PTSD (ps=.08), respectively. While some olfactory blunting was demonstrated in combat veterans regardless of PTSD status, the 2 groups showed opposite relationships between region-specific odor activation and both trauma exposure and post-trauma levels of avoidance/numbing. A non-deployed control group as well as longitudinal assessments are necessary to determine whether differences in BR-elicited activation patterns signify a premorbid risk factor, a post-trauma positive-adaptive or PTSD-related change in brain function.

Funding Acknowledgements: NIMH grant K01MH090548.

FCOI Declarations: None.

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#141

POSTER SESSION I
THURSDAY, 8 – 10:30 AM

WITHDRAWN

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#142

Subjective and Objective Gustatory and Olfactory Functioning during Chemotherapy in Breast Cancer Patients and Women without Breast Cancer

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Changes in chemosensory perception are burdensome side effects of chemotherapy in cancer patients. Literature is inconsistent in reporting the nature and prevalence of these changes due to differences in study design, tumour type and treatment. We aimed to follow and characterize olfactory and gustatory changes subjectively and objectively in breast cancer patients treated with chemotherapy. 30 women with newly diagnosed breast cancer treated with chemotherapy and 30 age matched women without breast cancer were followed in the course of time. Patients were measured before start of chemotherapy (T1), halfway (T2) and after the end of chemotherapy (T3); a similar time frame was used for the control group. Subjective assessment of gustatory and olfactory function was measured using a visual analogue scale (0-100mm). Objective gustatory and olfactory function was assessed with Taste Strips and the Sniffin' Sticks (Burghart Wedel, Germany) respectively. Preferences for taste intensities were assessed using five concentrations of sucrose and NaCl in lemonade and tomato juice respectively. Preliminary analyses (21 patients, 14 controls) show that subjective gustatory and olfactory function decrease during and shortly after chemotherapy for breast cancer patients (taste: T1: 78±19; T2: 51±25; T3: 59±27; smell: T1: 69±23; T2: 58±23; T3: 61±28) compared to the control group (taste: 72±15; smell: 67±19). Total taste strip scores show a decreased gustatory function during chemotherapy in patients (T1: 11.3±3.2; T2: 9.4±4.3; T3: 10.3±3.1) compared to the control group (T1: 11.8±2.1; T2: 11.14±2.9; T3: 11.7±2.1), and seem to recover after chemotherapy, but there were no clear effects for specific tastes. Objective olfactory function show no clear change during chemotherapy compared to controls. Subjective and objective gustatory function seem to correspond in showing a decreased function during chemotherapy and a recovery after the end of chemotherapy, while results for olfactory perception are less clear. These results give more insight in on sensory perception during chemotherapy in breast cancer patients.

Funding Acknowledgements: This research was supported by Top Institute Food and Nutrition, Alpe d'Huzes (grant nr UW2011-4987) and the Dutch Cancer Society (grant nr: UW2011-5268).

FCOI Declarations: None.

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#143

Reduced Caudate Connectivity in Phantosmia Patients using Resting State fMRI

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Phantosmia is a disorder characterized by the perception of odors in the absence of any olfactory stimulus. While some phantosmias manifest after a brain injury, a tumor, or a neuropsychiatric disorder, many patients present with an idiopathic onset of phantosmia, often with no other neurological or psychiatric symptoms. While previous imaging studies with PET or fMRI have used olfactory stimuli to investigate phantosmia, we used resting state fMRI to measure the olfactory network connectivity in phantosmia patients without any exogenous stimuli. After identifying 18 regions of usual olfactory pathways and surrounding structures, we calculated a matrix for the strength of connectivity for all possible pairs and performed an independent t-test comparison for each of these values between the phantosmia and control groups. Post-hoc voxel-based analysis using right caudate as a seed region demonstrated remarkable spatial localization of differences between groups to well- established olfactory structures. Compared to the age and sex-matched control group, we found that phantosmia patients have lower connectivity between the right caudate and the amygdala (two-sample t-test, Left: p=0.022 / Right: p=0.005) as well as between the right caudate and the putamen (Left: p=0.022 / Right = 0.014). These data suggest that decreased connectivity between these olfactory structures and the caudate nucleus may play a role in phantosmia. Future studies examining these areas with fMRI comparisons between phantosmia and control groups will better elucidate the caudate's role in olfactory dysfunction.

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FCOI Declarations: None.

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#201

Myelination of the Developing Anterior Commissure

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The olfactory system employs two large myelinated tracts: the lateral olfactory tract (LOT) conveys afferent information from the olfactory bulb to the olfactory cortices, while the anterior commissure (AC) coordinates processing on the left and right sides of the brain. Recent studies from our lab have indicated that the two tracts have different developmental histories. For example, using immunostaining for myelin basic protein (which is involved in the compaction of myelin during development) as an assay, the LOT begins myelination about postnatal day (P) 8 and is substantially myelinated by P10 while the process occurs in the AC between P11-15. The present work employed electron microscopy (EM) and immunohistochemistry to better understand AC myelination in P10, 20, and 30 mice as well as subjects that had a single external naris surgically occluded on P1 and were reared to P30 (NOSX). Samples of the AC as it crossed the midline were obtained to allow for quantitative observation of the entire structure in a uniform location. Very few myelinated axons were observed at P10, and only thin myelination observed. By P30 about 18% of the axons were fully myelinated in both control and NOSX animals. No topographic organization of myelinated axons within the AC was observed. The development of oligodendrocytes was traced with several biomarkers. Subjects examined at P10 exhibited a high density of early markers (PDGF, NG2). The pattern of staining changed by P20, when a high density of markers for mature, premyelinating oligodendrocytes (Olig2, CC1), was observed. By P30 tissue was dominated by markers for full myelination (heavy staining of MBP, relatively low density of PDGF and NG2, and equal density of CC1 as seen in P20). The results indicate that substantial development occurs between P10 and P30 within the AC and that unilateral naris closure does not appear to affect the process.

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FCOI Declarations: None.

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#202

Postnatal Pruning of Primary Gustatory Afferent Terminal Fields in the Mouse NST May Be Driven by Microglia

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Age-related decreases in terminal field sizes occur in a variety of sensory systems and at various neural levels. In rat, we found that the terminal field sizes of the greater superficial petrosal (GSP), chorda tympani (CT), and glossopharyngeal (IX) nerves decrease postnatally within the nucleus of the solitary tract (NST). By contrast, rats born to mothers fed a Na-restricted diet only from embryonic day 3 (E3) to E12 resulted in a lack of postnatal pruning. Here we show that the CT and GSP in mice also prune postnatally by about 2X from postnatal day 20 (P20) to P30. The terminal field size of the IX does not change with age. Similar to rats, the CT and GSP terminal fields of mice fed the Na-restricted diet from E3-E12 increased about 2X from P15 to P30. In fact, the E3-12 dietary manipulation yielded terminal fields in adults that resembled those of young, control mice. No group-related differences were apparent for the IX terminal field. To begin an examination of potential mechanisms for the normal pruning of terminal fields and possible reasons for the lack of pruning in the Na-restricted mice, we examined microglia resident in the NST. The bases for this derives from identification of microglia in the NST (Bartel, 2012) and from work showing microglia to be an important player in pruning of retinal ganglion cell terminal fields in the rodent lateral geniculate nucleus (Stevens et al., 2007). We found that microglia decrease in number with age where the GSP and CT terminate in the control mouse NST. Unexpectedly, there were significantly fewer microglia in E3-E12 Na-restricted mice at P15 and at P60 compared to age-matched controls. To extend these findings, we examined the development of terminal fields of mice in which C3 of the classic complement cascade was deleted throughout development. The CT and GSP terminal fields failed to prune in C3 knockout mice during development. In fact, the CT terminal field sizes in mice >P25 resembled that of the E3-E12 Na-restricted mice. These are intriguing results that may ultimately provide insights into molecular/cellular mechanisms of taste circuit development.

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FCOI Declarations: None.

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#203

Olfactory Function and Structural Integrity of Entorhinal Cortex and Hippocampus in Non-demented Middle-aged and Older Adults at Risk for Alzheimer's Disease

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Neurodegeneration in Alzheimer's disease (AD) begins years, perhaps decades, before cognitive impairment. Early markers of disease are critical if intervention is to occur before irreversible damage. Because olfactory impairment precedes cognitive deficits in AD, it is a potential biomarker; however, the neural substrate is incompletely understood. Emerging risk factors for AD include lifestyle factors such as inactivity and obesity. The San Diego Odor Identification Test has been shown to reflect left hippocampal volume loss in patients with AD (Murphy et al., 2003). Its potential to reflect brain structural integrity in non-demented middle-aged and older adults at risk for AD because of metabolic syndrome (MetS), which has obesity at its core, is unknown. We investigated medial temporal lobe (MTL) thickness, hippocampal volume and the relationships between these measures of brain integrity and odor identification performance and metabolic risk factor burden. Participants were 65 non-demented, middle-aged and older adults, with and without MetS. We analyzed T1-weighted MRI images acquired at 3T and San Diego Odor Identification Test scores. Middle aged adults with MetS showed thinner MTL, particularly in entorhinal cortex, a site of early AD pathology. Greater MetS burden predicted thinner MTL in middle aged adults, particularly in entorhinal cortex. Older adults showed hippocampal volume loss. Poorer odor identification was associated with thinner entorhinal cortex and smaller hippocampal volume. Given the progression of AD neuropathology from entorhinal cortex to hippocampus, these associations in non-demented adults at risk for AD because of MetS are of particular significance because they support the potential for olfactory impairment to serve as an early indicator of neurodegenerative processes.

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FCOI Declarations: None.

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#204

Odor-Shape Cross-Modal Associations: A Developmental Trajectory

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Adults show consistent cross-modal associations between trigeminal odors and pointed shapes, and between olfactory odors and rounded shapes (McCall, Goubet, Engelman, & Willis, 2011). In the current study we explored the developmental trajectory of odor-shape cross-modal correspondences and the role of trigeminality, identification, and pleasantness in these processes. Children (6, 10, 13, n = 50, 41, 39) and adults (n = 25) were asked to match trigeminal (sage, menthol, ginger, lavender) and olfactory odorants (French vanilla, banana, baby oil, jasmine) with either a round or a sharp shape, to identify and rate the hedonic value of odorants. Trends analyses showed a significant linear trend indicating that the trigeminal odors became more strongly associated with sharp shapes with age while the olfactory odors became more significantly associated with round shapes with age ($p <$ .01). All age groups, including the 6 year olds, matched olfactory odorants with round shapes significantly more often than chance. However, only middle-schoolers and adults matched trigeminal odors with angular shapes above chance. Six year olds were significantly less accurate in identifying odorants compared to all other grades ($p < .01$) but identification did not improve significantly between 10 and adulthood. Identification scores for olfactory odorants were significantly higher than trigeminal odorants ($p < .01$), although identification remained very low overall. Correlations between hedonic judgments and sharpness or roundness were not significant suggesting that odor-shape associations were unrelated to odor liking. The low identification scores suggest that the acquisition of odor-shape associations may not rely on semantic identification. Instead we propose that consistent and reliable odor-shape associations are based on meta-cognition about the affordances of odors and olfactory objects. Increased knowledge and experiences with odors in conjunction with meta-cognitive skills allow for the abstraction of odor characteristics, such as their association with various shapes.

Funding Acknowledgements: Gettysburg College.

FCOI Declarations: None.

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#205

Olfactory Sensory Neurons Transiently Express Multiple Olfactory Receptors during Development

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In mammals, each olfactory sensory neuron randomly expresses one, and only one, olfactory receptor (OR)—a phenomenon called the "one-neuron-one-receptor" rule. Although extensively studied, this rule was never proven for all ~1,000 OR genes in one cell at once, and little is known about its dynamics. Here, we directly tested this rule by single-cell transcriptomic sequencing of 178 cells from the main olfactory epithelium of adult and newborn mice. To our surprise, a subset of cells expressed multiple ORs. Most of these cells were developmentally immature. Our results illustrated how the "one-neuron-one-receptor" rule may have been established: At first, a single neuron temporarily expressed multiple ORs—seemingly violating the rule—and then all but one OR were eliminated. This work provided experimental evidence that epigenetic regulation in the olfactory system selects a single OR by suppressing a few transiently expressed ORs in a single cell during development.

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FCOI Declarations: None.

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#206

BMP Receptor Alk2 Mediates Mesenchymal-Epithelial Interactions for the Development of Mouse Tongue and Taste Papillae

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The formation of taste organs, including the tongue, taste papillae and taste buds, is a complex process that requires epithelial-mesenchymal interactions governed by multiple signaling pathways. Among the many molecules and pathways, regulatory roles of bone morphogenetic proteins (BMPs) are important. However, which of the multiple BMP receptors are involved, and the role of their tissue/cell specific distribution is unclear. To understand how BMP signaling in tongue mesenchyme regulates mesenchymal interactions with lingual epithelium, we examined the expression of the type I BMP receptor ACVR1 (ALK2) in the developing tongue and genetically altered the functions of ALK2 in a tongue mesenchyme-specific manner using *Wnt1-Cre* driven system. The expression of ALK2 emerged in the nerve branches at the base of tongue swellings at E11.5. At E12.5, ALK2 expression was seen in the tongue mesenchyme in addition to the nerve fibers. At E13.5-E15.5, ALK2 was also detected in the tongue epithelium. *Wnt1-Cre* driven constitutive activation (ca) of ALK2 (Wnt1-Cre/caAlk2) in the tongue mesenchyme led to a dramatically smaller and misshapen tongue with the formation of multiple fungiform papillae at E12.5 and E15.5. Additionally, the circumvallate papilla and pharyngeal tongue region were missing. Conditional knockout (cKO) of Alk2 driven by *Wnt1-Cre* resulted in tongues with ankyloglossia at E12.5. Moreover, fungiform papillae on the mutant tongue had an altered distribution and formed in the median furrow, a normally papilla-free region. Our data indicate a distinct, level- specific role of BMP signaling in lingual mesenchymal-epithelial interactions for the proper formation of taste organs.

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FCOI Declarations: None.

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#207

K14-Cre and Dermo1-Cre Each Labels a Subpopulation of Taste Bud Cells in Mice

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In a study to demonstrate that taste bud cells are derived from the local surrounding epithelium, an inducible *Cre* driven by the promoter of the epithelial marker K14 (*K14-CreER*) was used and *K14-CreER* partially labeled taste bud cells upon tamoxifen treatments, suggesting other source(s) of progenitor/stem cells contribute to taste bud formation and renewal. Our recent findings using *P0-Cre*, *Dermo1-Cre* and *Vimentin-CreER* mouse models demonstrated the underlying connective tissue cells contribute to a population of taste bud cells. In the present study, we used (1) *K14-Cre* which specifically labels epithelial cells and their derivatives, and (2) double Cre with both *K14-Cre* and *Dermo1-Cre* that labels epithelial and mesenchymal cells, respectively, as well as their derivatives. The distribution of reporter RFP^+ cells was analyzed in the lingual taste buds that were labeled with pan-taste cell marker Keratin 8 (K8). In 2- and 8-week old mice, *K14-Cre* labeled the entire layer of tongue epithelial cells. The labels were especially intense in the spines of all the filiform papillae. RFP^+ cells were abundant among $K8^+$ cells, but a population of $K8^+$ RFP^- cells was found in each type of taste papillae, i.e., fungiform, foliate and circumvallate. However, in the *K14-Cre* and *Dermo1-Cre* double Cre positive mice, lingual taste buds were almost fully labeled with RFP signals. Our data suggest a dual origin of taste bud cells from both the surrounding epithelium and the underlying connective tissues.

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FCOI Declarations: None.

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#208

BMI, Sleep Duration Associated with Sweet Taste Preference in Children

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Risks of weight gain increase as sleep duration decreases, with evidence strongest in children. The effects of sleep on the chemical senses have been understudied. We assessed sweet taste preference in 4-6 year-olds to identify relationships between preferred concentration, acute sleep duration, and BMI z-scores. Children were recruited from daycares and WIC clinics throughout Ohio and Illinois. Sweet taste preference was assessed using the Monell Forced-Choice, Paired-Comparison Tracking Procedure. Height, weight, and parental reports of habitual sleep duration and amount of sleep obtained the night prior to testing were recorded. The Human Subjects Review Board approved the study. Intraclass correlation (ICC) (two-way mixed model, absolute agreement) between the two preferred concentrations was evaluated. Pearson's or Spearman's correlation coefficient and Mann-Whitney tests examined correlations and differences between groups. 93 children have completed testing so far. ICC for the 4 year-olds (0.063; N=49) suggested the preference data were unreliable. ICC for the 5 year-olds (0.165; N=27) and 6 year-olds (0.172; N=17) were small, suggesting reliability, so data from 5 and 6 year-olds were combined. No differences in preference existed when comparing children who achieved the recommended sleep duration prior to testing (N=21) to those who did not (N=23). A trend was observed for a negative association between preferred concentration and sleep duration the night prior to testing among overweight/obese (OVOB) participants (N=11) ($r=-0.533$; $p=0.09$). BMI z-score was significantly and negatively associated with sleep duration in the OVOB ($r=-0.746$, $p = 0.008$) but not with preferred concentration. Significant differences in preferences were noted between lean and OVOB participants ($14.1 \pm 8.3\%$ vs $19.3 \pm 7.8\%$; $p=0.039$), with OVOB participants preferring more concentrated solutions. Our results suggest that in OVOB children, decreased sleep duration is likely associated with an increased sweet preference. Testing is continuing in order to obtain a larger sample size to confirm.

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FCOI Declarations: None.

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#209

Plasma Concentrations of Sucralose after Ingestion of Sucralose Containing Beverages in Children and Adults

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Sucralose is partially absorbed after oral ingestion, with the majority excreted in the feces. Interestingly, high doses of oral sucralose are associated with relatively lower plasma concentrations than low doses, which may be due to activation of a gut efflux transporter. Few data are available on circulating concentrations of sucralose, especially after ingestion of doses found in commercially available products. We therefore conducted a same-subject cross-over study in healthy adults and children. Ten healthy adult volunteers (mean age 29.7 yr, BMI 25.8 kg/m^2) consumed 355mL water containing 0, 68, 170, or 250mg sucralose (equivalent to 1-4 diet sodas). A second group of adult volunteers (n=11, mean age 27.4 yr, BMI 26.3 kg/m^2) consumed 355mL Diet Rite Cola™ (68mg sucralose and 41mg acesulfame-potassium (ace-K)) or 68mg sucralose and 41mg ace-K mixed in seltzer in randomized order prior to a glucose load. Normal-weight children (n=10, mean age 9.1 yr) were randomized to consume 24mL water containing 0 or 68mg sucralose in an identical study design. Blood was collected before beverage ingestion and serially for 120 min. At the relatively low sucralose doses provided, sucralose absorption was directly proportional to the amount ingested, and was similar when administered alone (in water) or combined with other ingredients (ace-K or diet soda). Average plasma sucralose concentrations were 40% higher after the 250mg compared to 170mg (AUC 38,035 ± 9,007 vs. 27,423 ± 5,987 ng/mL/120 min). Similarly, sucralose AUC increased 3 -fold after 170mg compared to 68mg (AUC 27,423 ± 5,987 vs. 9,206 ± 949 ng/mL/120 min). Sucralose AUCs were comparable whether administered in water, combined with ace-K, or in Diet Rite Cola™ (AUC 9,206 ± 949 vs. 9,320 ± 1,168 vs. 8,281 ± 1,168 ng/mL/120 min). Plasma concentrations were similar in children and adults when doses were adjusted for weight. We conclude that intestinal sucralose absorption is similar regardless of the vehicle of administration, and increases in a linear manner when provided within the range of reasonable human sucralose consumption.

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FCOI Declarations: None.

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#210

Fatty Acid Signaling Pathways Play a Role in Weight Gain and Weight Loss on High Fat Diets

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It is well known that fatty acids provide the chemosensory cues found in dietary fat. Fatty acids are transduced using a pathway similar to that used for sweet, umami, and bitter stimuli. Fatty acids are transduced via specific GPCRs, PLCß2, second messengers IP_3 and DAG, calcium release, and the cation channel TrpM5. Our current research aims to elucidate the role fatty acid signaling pathways have on the control of dietary fat intake. Through use of genetic knockout mice ($TrpM5^{-/-}$), compared to their wildtype counterparts ($TrpM5^{+/+}$, WT), we have shown that $TrpM5^{-/-}$ mice eat significantly less and gain significantly less weight and body fat than WT mice when put on a high fat diet. However, this difference is only seen in males. Female WT and $TrpM5^{-/-}$ mice exhibit the same weight gain and caloric intake on high fat diets indicative of a previously unknown sex difference in this pathway. Subsequently, we have begun to explore the role of fatty acid signaling pathways in weight loss following administration of high fat diets. Again, male and female $TrpM5^{-/-}$ and WT mice were placed on either a control or high fat (HF) diet *ad libitum* for four weeks. Following this feeding regimen, mice were placed on food restriction where all mice received 60% of their normal daily caloric intake on the control (low fat) diet until they reached 80% of their pre-restriction weight. Interestingly, there are no significant differences between the male and female $TrpM5^{-/-}$ mice in terms of the rate of weight loss, in either the HF or control diet groups. Additionally, there were significant differences in rates of weight loss for the female $TrpM5^{-/-}$ mice compared to the female WT mice, with the $TrpM5^{-/-}$ mice being more resistant to weight loss. Male $TrpM5^{-/-}$ and WT weight loss studies are currently taking place in our lab. This is consistent with our previous claims that TrpM5 plays a major role not only in fatty acid taste transduction, but also apparently contributes to the control of dietary fat intake.

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FCOI Declarations: None.

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#211

Sustained Weight Loss and Altered Lipid Taste Sensitivity Induced by Modulation of Salivary PYY and Excendin-4 in Mice

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Peptide tyrosine-tyrosine (PYY), known to induce satiation when present in plasma, has recently been described to modulate body weight (BW) and food intake (FI) of diet induced obese (DIO) mice when included in saliva. Similarly, glucagon-like peptide 1 (GLP-1), a GI peptide present in both plasma and saliva, also modulates BW and FI. The purpose of this project was to study the anorexigenic effect of sustained elevation of PYY and Excendin-4 (Ex-4, a GLP-1R agonist) in saliva of mice. Using recombinant Adeno-associated virus serotype 8 (rAAV8) we performed gene transfer of GFP (control), PYY, Ex-4, and PYY-Ex-4 dual vectors to the submandibular salivary glands of C57BL/6 mice fed a high fat diet. We observed a significant ($p<0.05$) decrease in BW of mice treated with either Ex-4, or PYY-Ex-4 dual vectors when compared to controls. Notably, PYY-Ex-4 dual mice displayed a significant decrease in BW as early as 1-week post vector administration while Ex-4 mice alone did not demonstrate a significant loss until 8 weeks post-injection. PYY mice, while demonstrating a decreasing trend in BW gain with respect to GFP mice, did not show a significant difference in BW during the 12-week experiment. To determine whether the anorexigenic effect of salivary PYY and Ex-4 is modulated through taste perception, we utilized a Davis Rig gustometer to generate brief access taste response curves for a panel of tastants for all groups of treated mice. Notably, the Ex-4 as well as the PYY-Ex-4 dual group displayed a significant increase in sensitivity to intralipid stimulus, suggesting that this taste modality plays a role in BW modulation. Unexpectedly, PYY mice also demonstrated a significant decrease in bitter taste responsiveness although the influence of this change on BW accumulation remains to be determined. In conclusion, a sustained elevation in salivary PYY and Ex-4 enhances weight loss and is correlated with changes in lipid responsiveness. Moreover, engaging both PYY and GLP-1 downstream signaling has a higher effect than PYY or Ex-4 alone, suggesting a synergistic effect of dual treatment and potential therapeutic application.

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FCOI Declarations: None.

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#212

Are Low Bitter Tasters More Successful at Weight Loss One Year Post-Bariatric Surgery?

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We examined if chemosensory function was associated with one-year weight loss success after bariatric surgery in morbidly obese females, and, if chemosensory function varied between the morbidly obese and non-obese controls. Method: In a case-control design and convenience sampling, we evaluated 46 morbidly obese females pre-surgery (Pre-Op) and 36 female bariatric patients 1-year post-surgery (Post- Op), who underwent either gastric bypass (n=12) or sleeve gastrectomy (n=24). The Post-Op group was derived from the highest (n=22) and lowest tertiles (n=14) of 1-year excess weight loss. The bariatric patients and 215 non-obese controls rated intensities of quinine, NaCl and propylthiouracil (PROP); ratios of bitter to NaCl were analyzed. The patients self-reported their smell and taste function as well as rated the sweetness, flavor (nose plugged/unplugged) and liking of jellybeans. Results: Either smell or taste alterations were self-reported by 20% of Pre-Ops, similar to the 2011-2012 NHANES frequencies (Rawal et al, 2015). Among all Post-Ops, 25% self-reported smell and 33% taste alterations since surgery, which did not vary by weight loss success. Interestingly, 33% reported improved ability to taste salt, sugar, sour and bitter since surgery. From measured taste, Post-Op patients in the most successful weight loss group averaged lower bitter ratios than non-obese controls, less successful Post-Ops or Pre- Op patients. The patient groups did not vary significantly in jellybean sweetness, flavor or liking. Summary: Bariatric surgery was associated with self-reported chemosensory improvements and alterations, but these perceived changes were not related with weight loss success at one year. However, those more successful in weight loss had lower measured bitter taste function. Our findings are consistent with those from the Tepper laboratory, which show that PROP nontasters consume more energy than do super-tasters (Shafaie et al, 2013), and, thus low bitter tasters may have more benefit from surgery that limits intake.

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#213

Diet Induced Salivary Protein Expression is Stimulus Specific

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A subset of salivary proteins (SPs) are upregulated after 5-day exposure to tannic acid (TA, 3% of diet, n=8) and quinine (0.3% of diet, n=8). We have previously demonstrated that the presence of these proteins alters diet acceptance. Both TA and quinine are bitter, and both cause substantial decreases in total food intake during the first 4 days of exposure (p's<0.05). We sought to clarify that the induced changes were specific to the stimuli and not a broad response to either aversive taste stimuli or food restriction. We explored three lines of evidence to demonstrate that the protein expression is stimulus specific. First, protein analysis demonstrates that exposure to the two stimuli cause differential expression of proteins. Bands containing cystatin S glutamate- rich protein B (GRP-B) and an uncharacterized protein are upregulated by exposure to both diets. Quinine exposure alone upregulates a band containing submaxilary gland androgen regulated protein 2 (SMR2), while TA exposure upregulates bands containing the related, SMR1. TA also upregulates a band containing prolactin inducible protein. Second, we tested the hypothesis that SPs were altered by diet restriction. Rats (n=5) were pair-fed control diet (g/kg) matching the intake of rats offered the quinine diet. No SPs were altered in response (p's<0.9). Lastly, we tested the hypothesis that changes could be driven by any aversive taste stimulus. We fed rats (n=8) a diet containing 8% SOA, a man-made bitter stimulus. Although individual intakes were highly variable on the SOA diet, none of the SPs upregulated by TA or quinine were altered by SOA exposure (p's>0.1).

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FCOI Declarations: None.

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#214

Haplotype-specific TAS2R38 mRNA Expression in Human Fungiform Papillae and Nasal Epithelium

Don Tucker Award Finalist

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Taste cells in human fungiform papillae express the*TAS2R38* gene, which encodes the T2R38 bitter taste receptor. One of its two common receptor haplotypes (T2R38-PAV) responds to the bitter chemical phenylthiocarbamide (PTC) whereas the other haplotype (AVI) does not. However, people with one or more *TAS2R38* PAV haplotypes vary considerably in their bitterness ratings for PTC; these perceptual differences among people of the same genotype are due in part to PAV mRNA abundance in fungiform taste papillae. Upper airway cells also express T2R38 where it has a different function: innate immunity. These differences among individuals are similarly based on *TAS2R38* genotype as evidenced by the observation that nasal cells of PTC-tasters (PAV haplotypes) respond to acyl-homoserine lactones (signaling molecules from bacteria that trigger a defensive reaction) while those from non-tasters do not. Prior studies have also established that the non-taster allele (AVI) is an independent risk factor for chronic rhinosinusitis requiring surgical intervention. However, as is the case for bitter taste intensity, people with one or more *TAS2R38* PAV haplotypes may vary in their immune response based on *TAS2R38* PAV mRNA abundance. Thus, it would be useful if a taste test could not only establish *TAS2R38* genotype but also the degree of nasal *TAS2R38* mRNA expression. To that end and as part of a larger and ongoing clinical study, two human subjects provided fungiform and nasal epithelium biopsy samples and we quantified haplotype-specific *TAS2R38* mRNA expression. The results to date suggest that the relative abundance of the *TAS2R38* PAV haplotype in nasal tissue is higher (2.6 fold; qPCR) than fungiform papillae expression and that high expression in one tissue predicts higher expression in the other tissue (in fold-values, qPCR relative to GAPDH): Subject A, fungiform=0.09 nasal=0.23; Subject B, fungiform=1.64, nasal=4.32. Biopsy of human fungiform taste papillae and nasal epithelium from the same subject yield mRNA suitable for quantification of haplotype-specific mRNA expression.

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#215

The Effects of Polymorphisms in the Tas1r3 Taste Receptor Gene on the Temporal Firing Pattern of Mouse Brainstem Gustatory Neurons

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Some neurons in the rodent nucleus of the solitary tract (NST) fire with short intervals characteristic of bursting. Furthermore, an NST cell's tendency to burst is related to its responsiveness to basic taste qualities, and depends on genetic factors. For example, there is a positive correlation between short-interval firing and the size of the taste-evoked response to sucrose in mice from the sweet -hyposensitive inbred strain 129P3/J (129), but not in the sweet-sensitive C57BL/6ByJ (B6) inbred strain. We proposed that the relationship in 129 mice may serve to amplify central sweet responsiveness and compensate for their poor peripheral sensitivity to sweeteners. To test this hypothesis, we looked at the relationship between NST bursting and sweet responsiveness in littermates from a 129.B6-*Tas1r3* segregating congenic strain. The mice had a 129 genetic background, and either did or did not have one copy of the *Tas1r3* allele from the B6 strain (B6/129 and 129/129 mice, respectively). The size of sucrose responses was significantly larger in B6/129 relative to 129/129 congenic mice, and there was a significant positive correlation between short-interval firing and sucrose response size in only the 129/129 group. Our data confirm that NST cells from 129/129 mice show the greatest tendency to burst in those neurons with the largest sucrose responsiveness. Moreover, this relationship is not present when sensitivity to sweeteners is increased by the presence of the B6 allele for *Tas1r3*. Our results are consistent with a mechanism to generate burst firing in particular NST neurons, based in part on which incoming gustatory signals are most in need of being amplified.

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FCOI Declarations: None.

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POSTER SESSION II
THURSDAY, 8 – 10:30 PM

WITHDRAWN

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#217

Functional Relationship between Oral and Intestinal Sweetener Receptors

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Recent studies have shown that sucralose enhances insulinemic and glycemic responses to an oral glucose tolerance test. We sought to determine whether metabolic responses to sucralose were associated with individual differences in sweet taste perception. To assess metabolic responses to sucralose, 12 human subjects underwent two oral glucose tolerance tests (1.39 M glucose neat) and two oral glucose tolerance tests with added sucralose (1.39 M glucose + 5 mM sucralose). Blood samples were collected periodically for 90 minutes and analyzed for plasma glucose, insulin, and glucagon. To assess psychophysical responses, concentration-intensity functions were determined for each subject using glucose, sucralose, and sodium chloride. Participants rated the sweetness of the glucose + sucralose mixture used in the oral glucose tolerance tests. Differences in metabolic outcomes were determined using repeated measures ANOVA with stimulus and time as within subject factors. The relationship between metabolic and psychophysical response to sucralose was assessed using Pearson's correlations. We also stratified subjects by tertile in terms of insulin AUC response from consuming glucose neat and compared psychophysical responses across tertiles using repeated measures ANOVA and t-tests. We found significant associations between taste sensitivity and insulin responses. Increased insulin AUC from glucose neat correlated with greater perceived sweetness from the glucose + sucralose drink ($R=0.683$, $p<0.05$). Individuals in the highest tertile of insulin AUC from glucose neat found 1.58 mM sucralose 60% sweeter ($p<0.05$) and 1.39 M glucose + 5 mM sucralose 46% sweeter than did individuals in the lowest tertile. There were no differences in the perception of glucose neat or sodium chloride between groups, which indicated that perceptual differences between groups were specific to sucralose. These findings provide evidence of a relationship between sweet taste perception and metabolic responses triggered by sweetener receptors in the gut.

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FCOI Declarations: None.

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#218

Differential Expression and Localization of GPCRs to Cilia of Neurons in the Olfactory Bulb

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Primary cilia are signaling centers for numerous extracellular cues. In the olfactory system, cilia are necessary for the detection of odors as well as regulating horizontal basal cell function in the regeneration of the damaged olfactory epithelium. Odorant receptors (ORs), 7-transmembrane GPCRs, localize to cilia on olfactory sensory neurons where they are able to interact with odors in the nasal cavity. In the brain, it is known that a subset of GPCRs also localize to the primary cilia of neurons. The role of these ciliary GPCRs in regulating neuronal function however remains unclear. Recent work has demonstrated the neuronal cilia are important for regulating dendritic growth and branching patterns. Additionally, several of these GPCRs are receptors for neuromodulatory cues, and may be involved in regulating neuronal response to stimulation. To address their role in neuronal function, analysis of cilia, and ciliary GPCRs, across neurons in the olfactory bulb (OB) was performed. Antibody staining for AC3 and ARL13b revealed cilia present on interneurons surrounding glomeruli, mitral and tufted cells and granule interneurons. Interestingly AC3 staining appears strongest in cilia from glomerular interneurons. The localization of several GPCRs, including SSTR3, MCHR1, DRD1, HTR6, CHRM2, CHRM3, and CHRM5 to OB neuronal cilia was also tested. Of these receptors, only SSTR3 and MCHR1 were detected in cilia on neurons in the OB, however neither was present in cilia of glomerular interneurons. SSTR3 was present in cilia on mitral cells and granule cells, while MCHR1 localized primarily to granule cells with minimal co-localization with SSTR3. Ciliary localization of SSRT3 and MCHR1 was lost in BBS4 knockout mice, suggesting both specificity in staining and that ciliopathies could also affect olfactory function through alterations in the OB. Furthermore, the localization of these receptors to granule interneurons suggests a mechanism by which the function of these cells can be modulated through hormonal regulation. Further study will help to elucidate the functional roles of primary cilia on OB neurons.

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#219

Identification of Mammalian Olfactory Receptors Responsive to an Opponent Odorant Pair

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An intriguing question is whether mammalian olfactory receptors (ORs), as known for other G protein- coupled receptors (GPCRs), induce a network of intracellular signaling through both G protein- dependent and -indepedent mechanisms in a ligand-dependent manner, often referred to as ligand-induced selective signaling (LiSS). Towards this end, indirect evidence suggests that odorants can inhibit mammalian olfactory receptor neurons (ORNs) through activation of phosphoinositide 3-kinase (PI3K) signaling using the same OR that excites the cell via activation of adenylate cyclase III (ACIII). To explore this possibility further, we isolated native rat ORNs responsive to an opponent odorant pair and identified the OR expressed by the cells using single cell degenerate RT-PCR, ascertaining that the ORNs expressed OMP to indicate that they were mature ORNs. We then tested their ligand specificity using heterologous HEK293T-based assays for both the ACIII and PI3K pathways. Using this approach we identified a single OR that activated ACIII signaling in response to octanol and PI3K signaling in response to citral, and show that citral inhibited the ACIII response evoked by octanol in a manner consistent with the properties of the original native ORN. This finding strengthens the idea that at least some mammalian ORs participate in LiSS and argues that at least one function of LiSS is to mediate opponent input into ORNs.

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#220

Strain Dependent Responses of Olfactory Sensory Neurons to 14 Diverse Odors

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We used the electroolfactogram (EOG) to survey the responsiveness of olfactory sensory neurons to 14 diverse odors in mice from 6 inbred strains (C57BL/6J, CAST/EiJ, NOD/ShiLTJ, PWK/PhJ, and WSB/EiJ). These strains, which are founders of the Collaborative Cross, capture much of the genetic variation in laboratory mice; they thus provide a useful screen to assess genetic variation in EOG responses to odors. To characterize their odor responsiveness, we recorded from olfactory turbinates 2b and 3 from 4-6 mice from each strain. Using a humidified air delivery tube we presented the odor headspace of a 0.1 M solution (in DMSO) of the following odors for 100 ms each: 2-ethylfenchol, diacetyl, guaiacol, citronella, caproic acid, 2-butanone, octyl acetate, isobutylaldehyde, beta ionone, isovaleric acid, cis-3-hexenol, ±carvone, citronellol, and androstenone. These odors were selected not only because they represent diverse functional groups, but also because many of their olfactory receptors (ORs) have been identified using cell culture, or have been found to be associated with specific ORs in a genotype/phenotype association study. In addition, several of the odors do not have known ORs. Using one-way ANOVAs (6 strains x 1 odor), we found significant strain differences in peak current, latency to peak, and rate of rise (peak current/latency to peak), but not for every odor. The peak response to citronella was blunted in CAST compared with NZO mice. In addition, the CAST latency to peak was shorter in response to 8 of the 14 tested odors compared with the NOD strain. Furthermore, the NOD and NZO strains showed different rates of rise for 2 of the odors without known ORs (2-butanone and isobutyraldehyde) in addition to citronella, and caproic acid. The C57 and NOD strains, and NZO and WSB strains also showed different rates of rise in response to 2-butanone. Most of the strain differences observed involved the NOD and NZO strains. Therefore, the NOD strain, a model of Type I diabetes, and the NZO strain, a model of Type II diabetes may be useful for elucidating diabetes-related changes in olfaction and/or identifying candidate ORs for an odor.

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#221

Probing the Olfactory Code using Antagonists

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The olfactory system recognizes each odor using a unique subset of odorant receptors. Although the specific pattern of odorant receptors activated by an odorant code for the odorant's identity, there are few, if any, explicit predictions relating odorant receptor activity patterns to olfactory perception. Given that the receptor activation patterns encode the odorant identity and intensity, antagonizing these receptors should alter perception of the odorant. Here we targeted a trace-amine associated receptor (TAAR) due to previous work indicating that genetic knockout of a single TAAR eliminated innate aversion to a predator odor (Dewan et al., 2013). Human TAAR5, activated by trimethylamine (TMA), is the only human TAAR with a published ligand (Wallrabenstein et al., 2013). Using a cell-based luciferase assay, we first identified an *in vitro* antagonist for hTAAR5 ($p<0.0001$), and then tested the antagonist in a psychophysical paradigm. Using an olfactometer, we presented subjects with one of the following odors: TMA alone, the antagonist alone, or TMA with the antagonist. Subjects were asked to rate the intensities of the odor qualities in each mixture. TMA-alone and antagonist-alone were not different in intensity ($p = 0.217$). The antagonist decreased the perceived intensity of TMA relative to both TMA-alone ($p<0.01$) and TMA mixed with an intensity-matched control odor ($p<0.05$). Antagonist odors promise to be a powerful tool for examining the contribution of individual receptors to odor perception.

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Comparison of Olfactory Sensitivity in Sensory Neurons and Behaving Animals

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The ability to detect and discriminate a broad range of environmental chemicals and to translate that information into meaningful cellular responses is essential for the survival of all individuals. This challenge is met by different chemosensory systems. Among those, the main olfactory system is indispensable for the perception of volatile odorants. While the peripheral odor detection mechanisms have been extensively investigated, little is known about how sensitivity of individual olfactory sensory neurons (OSNs) relates to the detection threshold in behaving animals. Furthermore, there has been a long-standing disparity between the sensitivity of physiological responses of olfactory neurons and behavioral thresholds measured in intact animals. Using a combined approach of gene targeting, electrophysiology, in vivo imaging, and behavior, we examined odor detection thresholds of single OSNs in the olfactory epithelium, glomeruli in the olfactory bulb, and behaving mice. We focused on a class of main olfactory receptors - the Trace Amine-Associated Receptors (TAARs). Our results indicate that TAARs contribute significantly to setting behavioral thresholds to specific odors (amines). Perforated patch-clamp recordings from knobs of TAAR-expressing OSNs exhibited very low detection thresholds. *In vivo* imaging of the corresponding glomeruli in awake mice showed slightly higher detection thresholds, with responses from the same glomeruli in anesthetized mice being less sensitive. Behavioral detection thresholds reported by a go—no go assay were similar to those observed in glomerular responses of awake animals. Together, our results show that behavioral thresholds can closely follow the sensitivity of one or a few populations of genetically defined OSNs. In addition, active processes in awake animals, such as sniffing, make an important contribution to odor sensitivity. More generally, our combinatorial approach allows us to characterize for the first time how chemical detection at the level of OSNs relates to olfactory perception in the behaving animal.

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FCOI Declarations: None.

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#223

Simple Measurement of Automated Analysis of Olfactory Behaviors: Results and Concerns

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Olfactory sensory neurons of the nasal cavity project environmental chemical information to the olfactory bulb (OB) for processing. The OB consistently alters neuronal physiology in response to changing environmental stimuli. This alteration is known as plasticity, and is commonly identified by a drop in dopamine following a loss of sensory activity. The cellular mechanisms of plasticity, and the effects on behavior, are unknown. In the present studies, we sought to measure the behavioral changes driven by neuronal plasticity. We first established a behavior standard with a manual q-tip task, where animals investigated scented cotton swabs. The first four trials of the q-tip task are blank (10ul mineral oil) followed by a scented test trial (typically 1:1000 acetophenone in mineral oil). Investigation time during each trial is recorded. Animals were considered able to perceive the test odor if investigation was greater in the 5^{th} trial over the 4^{th} trial. For automation, we utilized a behavioral olfactometer to produce equivalent habituation-dishabituation responses. Increased poking behavior and latency to first poke were measured. Animals remain habituated when the 4^{th} and 5^{th} trial response are similar, and dishabituate when the 5^{th} trial odor is novel to the 4^{th} (n = 12, p-value = 0.1, n = 14, p-value = 0.009), validating animals respond exclusively to the odor. We then treated animals with PBS and Triton to alter animal physiology. This established the ability to manipulate animal olfactory behavior (n = 6, p = 0.019, n = 6, p = 0.41). We then sought to measure odor detection of Amyl-acetate (AA), a typical odorant used, with a reported threshold concentration of 1.6E-6 AA:Mineral oil. In our task, animals respond to concentrations of 1E-8 (n = 8, p = 0.0498). We are unclear on why we received these results. Unlike other published tasks, latency was measured, and our results show quick response (~5s) to odorants reported undetectable. Perhaps animals unconstrained by motivational state can respond to cues that do not necessarily encode odor identity. Further investigation into latency as a response metric may help answer this.

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FCOI Declarations: None.

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#224

When All Noses Agree: Cross-Cultural Universalities in the Sensory Rejection of Malodor

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Cultural norms may govern the visible expression of emotion; yet, humans are biologically prepared to show the same expressions across cultures. The expression of disgust, for instance, has an important self-serving function, by adaptively reducing sensory intake of potentially noxious stimulation through the eyes and nose. However, some cultures value emotional constraint with regard to the visible outings of emotions, such as facial expressions. Can cultural differences in emotion expressiveness impact the degree of sensory rejection? More specifically, does the cultural norm of 'emotion moderation' present in East Asians (vs. Westerners) blunt sensory rejection responses, when these individuals are exposed to a "universal" bathroom malodor that signals microbial threat? To test this, 59 participants (29 East Asian) were exposed to "universal" bathroom malodor (vs. control odor) and images of dirty toilets, clean toilets, and neutral items. Sniffing behavior, facial muscle activity, and explicit judgments of the odors and images served as our (in)visible indicators of sensory rejection. The results showed that participants from *both* cultures reacted with a negative affective, $F(1,57) = 5.01$, $p = .029$, (disgusted: $F(1,57) = 3.22$, $p = .078$) facial expression to malodor (and dirty toilet images). Negative information, regardless of whether it came from smell or from the image, was shown to 'spill over' to the respective judgment of images, $F(2,114) = 4.66$, $p = .011$, and the odor, $F(1,57) = 14.13$, $p < .001$. Moreover, exposure to malodor *generally* resulted in the sensory rejection of malodor through shallower, $F(1,54) = 17.06$, $p < .001$, and less voluminous sniffs, $F(1,54) = 7.08$, $p = .010$. In sum, the present coherent set of results showed that sensory avoidance of universal malodor was not impacted by cultural expression norms.

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Nostril Differences in Olfactory Performance

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In analogy to other sensory systems, both nostrils sometimes exhibit an asymmetry in olfactory sensitivity. Recent studies suggest that several medical conditions affect olfactory capacity differently on both nostrils. However, nostril differences remain relatively understudied as most tests are conducted in a birhinal way, making it impossible to detect these impairments.

The first objective of this study was to examine differences in olfactory sensitivity between both nostrils. For this, we measured olfactory performance in 278 healthy participants from 4 different age groups (group A 6-15 years, n=43; group B 16-35 years, n=146; group C 36-55 years, n=52; group D >55 years, n=37) using a monorhinal adaptation of the Sniffin' Sticks test, assessing odor threshold, discrimination and identification. We observed a significant effect of age group in all three tests, with best results in groups B and C. Further, we observed an effect of gender in threshold and identification tests, with women outperforming men. We further observed an effect of age group on the difference between both nostrils: in the threshold test, the difference between the nostrils is significantly lesser in group A than in all other groups. This part of the study allowed us to establish normative data with cutoff values. The second objective was to compare a group of 180 patients with hyposmia from the 4 different age groups with those healthy controls. As expected, patients scored significantly lower than healthy participants, compared to controls of their age group. Further, differences between nostrils were significantly more pronounced in patients than in controls. In conclusion, this study enables us to evaluate differences in nostril sensitivity in both healthy participants and patients with hyposmia.

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FCOI Declarations: None.

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#226

Sip and Spit Versus Sip and Swallow: Differences in Perceived Intensity

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While the myth of the tongue map has been consistently and repeatedly debunked in controlled studies, evidence for regional differences in suprathreshold intensity has also been noted by multiple research groups. Given differences in physiology between the anterior and posterior tongue (fungiform versus foliate and circumvallate papillae) and differences in total area stimulated (anterior only versus whole tongue, pharynx, and epiglottis), small methodological changes (sip and spit versus sip and swallow) may substantially influence resulting data. We hypothesized instructing participants to swallow solutions would result in greater intensity ratings for taste than expectorating the solutions, particularly for umami and bitter, as these qualities were previously found to elicit regional differences in perceived intensity. Solutions of sucrose (sweet), a monosodium glutamate / inosine monophosphate (MSG/IMP) mixture (umami), isolone (a bitter hop extract), and quinine HCL (bitter) were prepared with reverse osmosis water; 70 participants rated all stimuli for sweetness, bitterness, and umami under two conditions (spit or swallow) in a counterbalanced crossover design. In repeated measures ANOVA, main effects of condition (spit versus swallow, $p=0.03$) were observed; we also found interactions of condition by taste quality ($p<0.001$) and tastant nested within taste quality ($p<0.001$). Specifically, rated intensity was greater for: swallowing vis-a-vis spitting, swallowing bitter tastants compared to spitting bitter tastants, and swallowing isolone versus spitting out isolone. Thus, the explicit instructions given to participants may be crucial in investigating differences in taste intensities, as spiting versus swallowing liquid stimuli may lead to different estimates of perceived intensity.

Funding Acknowledgements: Funds from the Pennsylvania State University.

FCOI Declarations: None.

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#227

Perceptual Maps of Hop Varieties Generated by Two Groups of Beer Drinkers

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Hop varieties may be evaluated via formal or informal methods. In formal evaluation, descriptive analysis is used, which takes weeks or months. This entails training a small group of judges on a common lexicon before samples are blindly rated for intensity on attribute scales. Alternatively, a common informal method is the hop-rub: whole cones are rubbed in the hands and judged by a content area expert (a brewmaster or hop farmer) against what would be expected for that variety. Critically, both methods use some type of expert, not actual beer consumers. To explore the use of a rapid profiling method on hop varieties, free sorting was conducted on 9 samples (7 varieties; 2 blind duplicates) in 2 sets of naïve participants segmented by their typical beer consumption habits. The first group (n=30) exclusively consumed light-style, American lager beer styles, while the second group (n=32) exclusively consumed what they considered 'craft' or 'micro-brew' beers. Participants first sorted the hop samples into groups based solely on aroma similarity in a semantic free task. After groups were formed, they were asked to endorse a list of 24 common hop aroma descriptors selected from prior literature in a check all that apply (CATA) task. Inspection of the perceptual maps generated via multidimensional scaling (MDS) indicate participants successfully differentiated between the hops, but the two sets of participants did not differ in how they perceived the hops, or in the number of groupings they formed in sorting. Conversely, there was evidence the light-style beer drinkers checked significantly fewer attributes in the CATA task to describe their groupings. These data suggest sorting can be used to profile hops, and any differences between types of beer drinkers are primarily linguistic, and not perceptual.

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FCOI Declarations: None.

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#228

Finding the Lower Limits for Olfactory Detection of Vanilla Using the Wheeler-UTC (WUTC) Odor Threshold Test

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Previous attempts to establish a lower limit for olfactory detection of vanilla using the Wheeler-UTC odor threshold test (WUTC) have been difficult for healthy participants; thresholds for this odorant appear to be much lower and more variable than initially anticipated (Jones, J., Tumlin, H., Mckinney, J., Tewalt, W., Ozbek, I.N., 2013). To identify a significant threshold for vanilla, 19 participants were given an extended version of the WUTC, which consists of a range of step-wise concentrations for vanilla among other odorants. Initially this test included 9 steps of vanilla, but had to be extended to 17 steps, and finally 30 steps to find a reliable threshold for this odorant. The concentrations were administered to each participant twice in a randomized order by a trained research assistant. A threshold was established for vanilla in 17 participants on the 30-step test. Median vanilla threshold was 13.52 ppm with a standard deviation of 53.10 ppm (skewness = 1.94). The thresholds ranged from 0.75 ppm to 182.56 ppm. A more accurate range was established for vanilla detection thresholds. Significantly more steps, therefore lower concentrations, were needed to find a lower limit for vanilla than other common odorants used in olfaction sensitivity tests. Funding was provided by the William H. Wheeler Center for Odor Research.

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FCOI Declarations: None.

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#229

Is Methyl Anthranilate Indeed a Contributor to Fruity Aroma in Strawberry?

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Current strawberry commercial cultivars, *Fragaria* x *ananassa* Duch., are the result of crosses that have improved production yield, disease resistance, fruit size, handling and storage, but have narrowed flavor diversity. Recently breeders are trying to introduce "old" flavors (i.e. flavor from wild type species) into new cultivars. Such is the case with methyl anthranilate (MA), which was found to be unique in some wild strawberry species such as *Fragaria vesca*. However, high amounts of MA can impart an unpleasant perfumey and soapy flavor. The objective of this study is to determine which concentration range of MA increases fruitiness flavor in strawberry. Strawberries were pureed and volatiles were stripped or not using a rotary evaporator. Methyl anthranilate was added to the puree at concentrations ranging 0.001 ppm to 10 ppm. Samples were presented as series of three-alternative-forced choice (3-AFC) tests with increasing MA concentration (ASTM E679-04). Untrained panelists (21) participated and were instructed to taste and identify the spiked samples among the three samples presented. Panelists could detect MA at 0.1, 1, or 10 ppm. Some found increased sweetness in some samples while others noted objectionable grape flavor. The highest level of MA found in 'Mara-de-Bois' strawberry was 6.05 ppm. The combination of MA with other volatiles will be studied further.

Funding Acknowledgements: ARS base funds.

FCOI Declarations: None.

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#230

Effects of Linguistic Labeling on Identification of Gustatory–Olfactory (Acid-Lemon) Flavor Mixtures

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An earlier study (Brewer et al, Chemical Senses 2013) showed that linguistic labels can systematically modify the identification of gustatory-olfactory flavor mixtures containing different proportions of sucrose and citral. Here, we ask how analogous labels affect the identification of gustatory-olfactory mixtures containing varying proportions of citric acid and lemon. Subjects often confuse citric acid and lemon (Marks et al, AChemS, 2015), probably because of their strong association in experience; consequently, labels might help disambiguate these flavorants and thereby exert an especially strong effect on identification. The experiment asked subjects to identify the stimulus on each trial as having either “mostly acid” or “mostly citrus.” In one condition, no label preceded each stimulus. In another condition, each stimulus followed a label, ACID or CITRUS, which, the subjects were informed, usually though not always named the stronger flavor component; that is, the label was probabilistically valid, based on the proportions of the two flavorants. The results showed that the label modified the identification responses, with subjects responding “acid” or “citrus” more often when the flavor mixture followed the corresponding (probabilistically valid) label, ACID or CITRUS. These results are compatible with a decision-theoretic model (Brewer et al. 2013), in which labels induce shifts in response criteria - although, conceivably, the labels might induce changes in the sensory representations of the flavor mixtures themselves. The magnitude of the effect of labeling with citric acid and lemon appears to be similar to the magnitude of the effect that Brewer et al. previously found with sucrose and citral.

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FCOI Declarations: None.

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#231

Olfaction Modulates Visual Self Perception

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People have the tendency to evaluate themselves more positively than they would others, a phenomenon known to psychologists as self-enhancement. This positivity bias is most often reported in social judgment. Little is known about whether it is also manifested in more intrinsic behavior. Here we use the binocular rivalry paradigm and ask whether self-enhancement prolongs visual awareness of the image of oneself relative to that of a stranger, and whether the enhancement is modulated by olfaction. Subjects viewed two competing visual images consisting of the subject him/herself and that of a stranger in the presence of a pleasant and an unpleasant smell, respectively, and reported when they noticed the alternation of the image from one to the other. We showed that the pleasant smell significantly prolonged the time in which the self was visible, whereas the unpleasant smell did so in the case of the stranger. To demonstrate that the association was due to self-enhancement and not to familiarity with the self, we introduced the images of two equally familiar individuals, one celebrated and one notorious, using an otherwise identical paradigm. We showed that the pleasant smell significantly prolonged the time to perceive the celebrated person, while the unpleasant smell did so for the case of the notorious person. These results demonstrate that olfaction modulates visual self-perception, and open a new avenue for the study of self-enhancement effects in general.

Funding Acknowledgements: University funds from Baylor College.

FCOI Declarations: None.

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#232

Effects of Ambient Light-color Cues on Olfactory Performance

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Previous studies have highlighted the fact that food and drink color cues can modulate chemosensory perception and acceptability. Recently, color cues related to other contextual factors such as cutlery and tableware items were found to influence chemosensory perception of food and drinks. However, even though people have increasingly experienced exposure to a variety of colored lights with the increased popularity of multicolored light-emitting diode (LED) bulbs, little attention has been paid to the impact of ambient light-color cues on smell and taste perception. Several studies have demonstrated that ambient light color could affect sour taste and wine flavor, but little is known about its impact on orthonasal odor perception, so this study was aimed at determining how ambient light color might affect orthonasal odor perception. Participants were asked to conduct odor sensitivity tasks and/or odor discrimination tasks via the Sniffin' Sticks test under three different light color conditions: white, blue, and red; blue and red colored lights were selected because they were found to alter emotional factors such as arousal and calmness. Based on previous findings that olfactory performance was affected by emotional status, it was hypothesized that red and blue light colors might increase participants' performance with respect to odor sensitivity and odor discrimination. Gender-related influence of light color on olfactory performance was also examined. Our findings demonstrate how light color cues modulate olfactory performance and provide new insight in the chemosensory field regarding the application of LED bulbs.

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FCOI Declarations: None.

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#233

Stability of Cross-Modal Associations between Odors, Colors, and Abstract Visual Forms

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Odors evoke associations with colors as well as abstract visual forms. Very few studies have explored the reliability of these cross-modal associations over time. We examined the consistency of odor-color and odor-shape associations in 60 human adults across two test sessions six weeks apart, and evaluated the relative influence of semantic or perceptual features in mediating these associations. Twelve odors were presented orthonasally at each session. At the first session, participants completed odor-color and odor-shape association tests, and evaluated each odor by rating intensity, familiarity, pleasantness, and identification. At the second session the color, shape and identification tasks were repeated. Shape-odor associations were more stable over the two sessions (70.42% identical responses from time 1 to time 2) than color-odor associations (39.58%), while odor identifications were stable only 49.36% of the time. Logistic regression was used to predict whether associations remained stable, using sensory/perceptual ratings (eg., intensity, pleasantness, familiarity) and semantic features (identification accuracy and consistency) as predictors. The strongest predictors of a stable color match were the strength of the odor-color association reported at time 1 (B=.271, $p < .05$) and consistency of identification (B=1.015, $p < .01$; $R^2 = .122$). By contrast, the stability of associations with abstract shapes was not predicted by odor identification. Only the reported strength of odor-shape associations at time 1 predicted whether those associations would remain stable (B = .237, $p < .05$; $R^2 = .38$). Furthermore, whether a particular odor was associated with rounded or pointed shapes was predicted by ratings of pleasantness (B = -.005, $p < .05$) and odor trigeminality (B = -2.00, $p < .01$; $R^2 = .212$), but unlike color associations was not related to identification accuracy. The results support the idea that odor-color associations are mediated by semantic identification while odor-shape associations are rooted in less variable structural or amodal mappings between stimulus features.

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FCOI Declarations: None.

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#234

Rapid Olfactory and Visual Association in Humans: An Olfactory fMRI Study

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Recent evidence shows that the visual system can modulate the way humans process olfactory information. A recent olfactory fMRI study provided strong evidence to support significant olfactory -visual association. However, whether or not this multisensory association depends on the semantic meaning of the visual cues remained unclear. Therefore, the purpose of this study was to further determine the functional networks involving this associative learning behavior using three novel olfactory-visual association fMRI paradigms that paired three categories of visual cues (neutral symbol, the semantically congruent word “smell” or the incongruent word “look”) with a specific odor stimulation. Similar to a classical conditioning paradigm, each fMRI paradigm included an acquisition phase, in which a specific visual cue was paired with the odor, followed by a test phase where only a visual cue was presented. It was hypothesized that the visual cues paired with the odor could produce activation in the primary olfactory system and the semantically congruent visual cue would produce stronger activation compared to the incongruent or neutral cues. The study cohort included only young healthy participants, each of whom completed the three olfactory fMRI paradigms on a Siemens 3T MRI system. The fMRI data were processed with SPM12. The results demonstrate significant activation in olfactory and visual regions during the testing phase of each paradigm, which involved presentation of a visual cue which was paired with an odor during the acquisition phase. Furthermore, significantly greater activation in olfactory-related brain regions was observed during the testing phase for a paired visual cue compared to an unpaired visual cue. Taken together, these findings strongly support our hypothesis of a process of rapid association occurring between the olfactory and visual systems.

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FCOI Declarations: None.

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#235

Localization Using Air Puff Stimulation: An fMRI Investigation

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The chemosensory and somatosensory portions of the trigeminal nerve are considered to be relatively independent. The close interaction between the trigeminal and olfactory systems, however, is thought to be mediated primarily through the chemosensory portion of the trigeminal nerve. The goal of this fMRI study, therefore, is to further characterize the trigeminal network in terms of somatosensory stimulation using fresh air puffs. Fourteen subjects (mean age 23 ± 2 years, 8 females) with normal smell function completed the "fresh air puff" fMRI paradigm at 3.0T. Three discrete fresh air puffs of duration 0.5 sec each were sequentially delivered to either the right, left, or bilateral nostrils, with an inter stimulus interval of 18 sec, and for a total duration of ~10 min. No visual or other cues were provided and a constant flow of fresh air was delivered bilaterally at 1 L/min. An MR compatible olfactometer was used to deliver continuous air as well as the three air puffs. An out of magnet experiment reported a success rate of more than 95% to localize air puffs. Our novel fresh air puff fMRI paradigm detected robust fMRI activation in the trigeminal network. In particular, air puffs stimulated the POC as well as the insular cortex which is considered to be a secondary olfactory structure. Other brain areas that were activated during left, right and both nostril stimulation included the primary olfactory cortex (POC), superior temporal gyrus, cingulate gyrus, cerebellum and sensorimotor cortex. The left sensorimotor cortex had a significantly greater number of activated voxels during right nostril stimulation, demonstrating contralateral functional organization. Our results highlight the importance of insula in integrating somatosensory information in the trigeminal network, and show that the somatosensory portion of the trigeminal nerve may also be linked to the olfactory system in humans.

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FCOI Declarations: None.

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#236

Taste Responses of Neurons Within an Anatomically Defined Region of Gustatory Cortex

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Gustatory cortex (GC) is an important integrative area, where gustatory information in single neurons may be merged with that of other modalities. Both physiological and imaging approaches have led to often-disparate conclusions about the organization of gustatory information in this area. In these studies, we used neuroanatomical and imaging approaches to delineate the likely area of insular cortex given to gustatory function, and to characterize taste responses within this delineated area. Anterograde tracers were injected into the taste thalamus (VPMpc) of mice, and the thalamic terminal field was characterized across the cortex. Cytoarchitecture, activity-evoked Fos expression, and cell type-specific expression were also investigated as markers of gustatory cortex. Working within the delineated area, we used two-photon imaging to measure taste responses in layer 2/3 neurons located just posterior to the middle cerebral artery, in a region previously shown in the rat to have an overlapping representation of taste quality (Accolla et al., 2008). Basic taste stimuli (NaCl, sucrose, citric acid, and quinine) were delivered to the oral cavity of mice and changes in calcium fluorescence were measured from multiple cells simultaneously. We recorded taste responses from 741 cells in 6 mice. We found that over 70% of cells responded to more than one tastant. Further, a non-biased, hierarchical cluster analysis revealed multiple clusters of cells responding best to either individual tastes or combination of tastes. Finally, comparisons of breadth of tuning and taste specificity with individual cell location suggest this region is primarily composed of more broadly tuned, taste-specific cells with no apparent spatial organization in terms of individual taste coding.

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FCOI Declarations: None.

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#237

Evidence for Central Regulation of Taste Sensitivity

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Sensitivity to taste fluctuates within individuals. Much of this variability has been attributed to peripheral mechanisms. Here we test a hypothesis about a central source of variation. In prior studies we have shown that a higher-order network of brain regions important for focusing attention can modulate insular taste cortex and influence taste perception. In the current study we used fMRI to test whether this "top-down" modulation of insular gustatory cortex and perception extends beyond the time point of directed attention to produce a more prolonged effect on taste sensitivity. 29 participants rated the intensity of tastants and retronasal olfactory (RO) stimuli and had brain response measured with fMRI before and after they performed a difficult flavor discrimination task, which required directing attention to the flavors. This procedure was then repeated using a visual, rather than flavor task. Supporting our hypothesis, response in the anterior insular gustatory cortex was greater and the rated intensity of taste, but not RO stimuli, significantly increased after the flavor, but not the visual task (p=.028). These results demonstrate that focusing attention on flavor results in a sustained increase in taste, but not RO sensitivity and suggests that the effect depends upon modulation of insular cortex. The findings cannot be attributed to a general effect of attention because taste sensitivity and insular response are not influenced by directed attention to visual stimuli. Moreover, the results suggest that the effect is specific to gustation since retronasal olfaction was unaffected. We conclude that central mechanisms contribute to intra-individual variation in taste sensitivity.

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FCOI Declarations: None.

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#238

Designer Receptors Exclusively Activated by Designer Drugs (DREADD) Inactivation of Forebrain Inputs to the Parabrachial Nucleus in Rats Reveals Dissociable Contributions to Benzodiazepine Hyperphagia in Rats

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The parabrachial nucleus (PBN) receives input from forebrain sites such as the lateral hypothalamus (LH), central amygdala (ceA), and gustatory cortex among others, as well as input from ascending afferent gut- and gustatory-related signals. Thus, the PBN is an ideal nexus for afferent gustatory signal modulation by ascending and descending signals related to post-ingestive, motivational, and learned cues. Previously we found that PBN application of the benzodiazepine (GABA-A agonist), chlordiazepoxide (CDP), increased licking to appetitive and aversive tastants but it did not affect licking to water or capsaicin, a trigeminal stimulus. We are now using the Designer Receptors E xclusively Activated by Designer Drugs (DREADD) technique to selectively and transiently inactivate specific pathways to the PBN. Using the Gi-coupled DREADD, AAV-hSyn-hM4D(Gi)-mCherry, we tested DREADD application within the: (1) LH; (2) ceA; and (3) combined LH and ceA, all coupled to intraPBN application of the DREADD ligand clozapine-N-oxide (CNO; 100μM; 0.6μL/3min infusion) to allow transient inactivation of the defined forebrain projections within the PBN during 1-h access tests of licking to either 0.2 M sucrose or 0.3 M NaCl, paired with systemic CDP (10mg/ml/kg b.w.) or saline vehicle injections. Under control conditions, we replicated the CDP effects on licking to both sucrose and NaCl. Inactivating the LH appeared to affect motivation equally under saline and CDP conditions, decreasing meal duration and pause duration. Inactivating the ceA appeared to affect hedonic value, producing decreases in first minute licks under saline but not under CDP, and further eliminating CDP-induced increases in licks per burst. Preliminary ongoing work with double LH and ceA inactivation suggests a synergistic effect on ingestive behavior. DREADD proves to be an effective technique to transiently manipulate projections to the PBN as a means to identify key sites contributing to benzodiazepine modulation of ingestive behavior within the PBN.

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FCOI Declarations: None.

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#239

Differential Signal Processing in Aubpopulations of Optogenetically Identified rNST Neurons

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Glutamatergic projection neurons and GABAergic interneurons comprise major neuron classes in the rostral (gustatory) nucleus of the solitary tract (rNST). Moreover, there is heterogeneity within each type based on morphology, projection status and ion channel composition. To begin to define how these distinct populations process excitatory afferent and inhibitory signals, we performed *in vitro* recordings from a transgenic mouse expressing ChR2 in GAD65-positive GABAergic neurons. GABA+ and non- GABAergic neurons (GABA-) were identified by their responses to pulses of blue light (455nm) delivered through an optical fiber (200um). Voltage and current-clamp protocols determined the presence of two hyperpolarization-gated currents, I_A and I_H. The solitary tract (ST) was electrically stimulated at different frequencies to define afferent responses in the presence and absence of light-driven activation of GABAergic circuitry. As a group, GABA- neurons (a population presumably including glutamatergic projection neurons) responded to ST stimulation with higher rates of discharge compared to GABA+ cells (P=.03). However, responses of GABA+ neurons were variable; those expressing I_H were more responsive (P=.03). In contrast to a transgenic strain that mainly identified GABAergic neurons in the ventral rNST (Wang and Bradley, 2010), I_A was expressed in both GABA+ (52%) and GABA- (38%) cells. Moreover, the presence of I_A was correlated with the susceptibility of GABA- neurons to inhibitory influences. ST-evoked responses in GABA- cells with I_A were more strongly suppressed by optical stimulation (p=.01) compared to GABA- neurons lacking I_A (NS). These data suggest that I_A could potentiate GABAergic feed forward inhibition in a subpopulation of projection neurons. In the presence of feedforward inhibition onto GABA+ neurons, afferent signals would be augmented in neurons with I_H, but suppressed in those with I_A. Finally, some GABA+ neurons were themselves subject to inhibition, suggesting an rNST substrate for disinhibition.

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FCOI Declarations: None.

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#240

POSTER SESSION II
THURSDAY, 8 – 10:30 PM

Optogenetic Stimulation of Gustatory Cortical Input onto the Nucleus of the Solitary Tract of the Rat Enhances Taste Information and Learning

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Spike timing of taste responses in the nucleus of the solitary tract (NTS) is known to convey information about taste quality. However, the inputs that shape and refine spike timing are unknown, and its behavioral relevance is unclear. Here, we used optogenetic tools to study how input from the gustatory cortex (GC) to the NTS shapes the temporal pattern of taste responses in awake, freely licking rats. We then used a Go-no-Go (GnG) paradigm to study the behavioral effects of selective enhancement of GC-NTS input. Initially, we infused viral constructs containing Channelrhodopsin 2 bilaterally into the GC. After 2-4 weeks, we implanted an optrode consisting of a fiber optic implant attached to a bundle of 8 tungsten microwires into the taste-responsive portion of the NTS. Following recovery, rats were water deprived and placed in an experimental chamber where they experienced a "taste-only" or GnG paradigm. In the taste-only paradigm, trials of 5 consecutive licks (12μL/lick) of a tastant (0.1 M NaCl, 0.1 M sucrose, 0.1/0.01 M MSG/IMP, 0.01 M citric acid, 0.001 M quinine, or artificial saliva) were interspersed with 5 rinse licks of artificial saliva presented on a VR5 schedule. After several days of taste-only recording, rats were switched to the GnG paradigm: a single cue stimulus lick (always 0.1 M NaCl) was presented followed by 5 dry licks and then 3 licks of a test stimulus (0.1 M MSG/IMP or 0.1 M KCl). After a 1 s timeout, the task was to continue licking for a 3-lick 0.5 M sucrose reward if the test and cue stimuli match and to withhold licking if they were different. Incorrect responses were punished by 3 licks of 1-2 mM quinine and a 5 s timeout. In a random half of each trial type, GC terminals in the NTS were stimulated optically (25 Hz of 473 nm laser light at 8-10 mW) for a maximum 1 s following the first test lick. Our data show that stimulation of centrifugal input from the GC can change the temporal characteristics of taste-evoked spike trains in the NTS and increase the fidelity of tastant information relayed by NTS neurons. In the setting of this optogenetically-induced increase in fidelity, there was improved performance in the GnG paradigm.

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FCOI Declarations: None.

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#241

Factors Associated With Inaccurate Self-Reporting of Olfactory Dysfunction in Older U.S. Adults

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Self-report of olfactory function has been shown to have poor sensitivity (i.e., people with true olfactory dysfunction are not likely to accurately report it). We aimed to identify factors associated with this lack of awareness of smell dysfunction. Objective odor identification was evaluated using a validated 5-item test in respondents from the National Social Life, Health, and Aging Project (NSHAP), a representative sample of home-dwelling, older U.S. adults ages 57-85 (n=1,468). Self-reported sense of smell was assessed using a 5-point Likert scale (poor to excellent). Using multivariate logistic regression, we tested factors that might influence poor sensitivity of self-reported olfactory function. Analyses included age, gender, race/ethnicity, education, marital status, cognition, comorbidity, smoking, depression, anxiety, self-rated mental and physical health, and measures of social activity/network.Although 22.0% of older adults in the U.S. had objective olfactory dysfunction (≤ 3 items correct out of 5), only 12.4% reported their sense of smell as poor or fair. Among those with olfactory dysfunction, 74.2% did not recognize it; these individuals were more likely to be older, African American, unmarried, and have worse cognitive function compared to individuals who recognized their dysfunction ($p<0.05$, all). Interestingly, these individuals who were unaware of their olfactory dysfunction had greater cognitive deficits ($p<0.001$) at five-year follow-up compared to normosmics, whereas those who were aware of their olfactory dysfunction did not ($p=0.06$). In summary, age, race, social context, and cognition are significantly associated with lack of awareness of olfactory dysfunction in older U.S. adults. We also found that individuals who are unaware of their dysfunction have poor neurologic outcomes. Given that self-reporting of olfactory dysfunction has low sensitivity, understanding the factors that are associated with inaccurate assessment of dysfunction may help clinicians decide which patients require objective smell testing.

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FCOI Declarations: None.

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#301

Modulation of Olfactory Bulb Odor Responses and Feeding-related Behavior by Nutritional Status Peptides

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Feeding behavior depends strongly on palatability that is determined, among other factors, by the smell of the food. Increasing evidence that nutritional status impacts the olfactory system suggests the presence of feedback from feeding control circuits modulates odor perception and impacts food intake. The mechanisms of this feedback are not fully understood, and we propose in this study to investigate the direct effects of two nutritional status signals (orexin and leptin) on olfactory bulb (OB) odor responses and how this impacts food seeking behaviors. Using wide field calcium imaging on transgenic mice expressing GCaMP3 in the OB neurons, we investigated the impact of nutritional status on OB odor responses. We found that glomerular odor responses to food odors were higher after a 24h fast compared to when they were fed. To understand the basis of this fasting-induced change, we tested the impact of anorexigenic and orexigenic signals on glomerular and mitral cell responses to odors using wide field and two photon imaging. We first tested leptin, an anorexinergic peptide reported to modulate olfactory sensitivity when injected intra-cerebro- ventricular or systemically. We found that systemic, icv and OB topic application of leptin equally reduced glomerular and mitral cell odor responses, suggesting a direct action of leptin on the OB. We are currently testing the effects of orexin, an orexinergic peptide also reported to modulate olfactory sensitivity, on OB odor responses. Our preliminary data show that orexin icv injection increases glomerular odor responses.To explore behavioral consequences of direct action of leptin and orexin on the OB, we used a food finding task in C57Bl6 mice implanted with chronic OB cannula. We found that the animals find their food faster when fasted for 24 hours than when *ad libitum* fed, and that this effect can be reversed by leptin. Future work will investigate whether orexin can shorten the time to food finding in *ad libitum* fed animals.

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FCOI Declarations: None.

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#302

POSTER SESSION III
FRIDAY, 8 – 10:30 AM

Evaluating Dietary Quality and Taste Preferences with a Simple Liking Survey: Application to Studying Individuals with Morbid Obesity

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Feasible ways are needed to screen for dietary behaviors and taste preferences in clinical settings. We examined the internal validity and reliability of a brief survey to assess food preferences and dietary behaviors among morbidly obese females considering weight loss surgery. Survey-reported liking is a proxy for dietary intake, correlating with reported food intake and biomarkers of nutritional status (Sharafi et al, 2015) and linking taste genetics with diet and health (Pallister et al, 2015). Methods— Enrolled were 109 morbidly obese females awaiting bariatric surgery, who completed a 100-item liking survey of foods/beverages, physical/ sedentary activities, and pleasurable/unpleasurable experiences. They were oriented to the survey with examples of activities that could be liked (winning the lottery, succeeding), neutral (doing a routine chore), and disliked (running out of money, paper cut) on a bi- directional, horizontal scale labeled at either end with strongest disliking/liking of any kind and mid-point of neither like/dislike. The survey took <10 minutes to complete. Survey items were averaged into nutritional, sensory (bitter, sour, spicy), and activity groups. The nutritional groups were formed into a dietary quality index (Sharafi et al, 2015) and, with activities, into a behavior index. The indexes had internal reliability (alpha>0.65) and were normally distributed. The most liked items were fruit, watching television, vegetables and sour items (listed from highest); least liked were alcoholic beverages, spicy foods, bitter beverages, and salty items (listed from disliked to barely liked). In exploratory principal component analyses, >50% of variability in either index was explained by 2 factors—less healthy (sweets, fats, salty, television) and more healthy (vegetables, fruits, fiber, physical activities) behaviors. Women who reported greater liking for bitter beverages and spicy items had significantly higher dietary quality. Summary: A simple liking survey is a feasible and relatively valid/reliable tool for assessing dietary and taste-related behaviors in a clinical setting.

Funding Acknowledgements: USDA Hatch and Connecticut Institute for Clinical and Translational Science.

FCOI Declarations: None.

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#303

POSTER SESSION III
FRIDAY, 8 – 10:30 AM

Is the Association between Sweet and Bitter Perception due to Genetics?

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Perceived intensities of sweetness and bitterness are associated and both are influenced by genetics. However, whether the association between sweet and bitter tastes is due to a shared genetic pathway has not been well studied in humans. Here, in a sample of 1901 adolescent and young adults (53.8% female; 243 MZ and 452 DZ twin pairs, 511 unpaired individuals; age 16.2±2.8 years), we used multivariate genetic modelling to estimate the covariance between the perceived intensities of four bitter compounds (6-n-propylthiouracil (PROP), sucrose octa-acetate (SOA), quinine HCl and caffeine) and a sweetness factor (weighted mean ratings of glucose, fructose, neohesperidine dihydrochalcone and aspartame). Sweetness was moderately associated with SOA, quinine, and caffeine (phenotypic correlations (r_p)=0.35-0.40), mainly due to a shared genetic factor (genetic correlations (r_g)=0.46-0.51), with 23% of the genetic variance in sweetness (heritability (h^2)=0.36) overlapping with 46-49% of the genetic variance in quinine (h^2=0.40) and caffeine (h^2=0.34), and 94% in SOA (h^2=0.40). In contrast, only 3% of the genetic variance in sweetness was associated with PROP, which increased to 15% after adjusting PROP for the *TAS2R38* diplotype (h^2 decreases from 0.73 to 0.40 after adjustment). Overlap in genetic variance between SOA, quinine and caffeine with PROP increased from 1-9% to 12-24% after adjusting for *TAS2R38*. In addition, we show that these associations were not inflated by scale use (i.e. a preference for the same rating) as the cross-trait (e.g. PROP with SOA, quinine, caffeine and sweetness) correlations for both MZ and DZ twins were weak (r_{MZ}=0 to 0.10, r_{DZ}=0.03 to 0.08) and there was little evidence for mediation of behavioural factors (general cognitive ability or personality). This study supports an overlap of genetic variance between the perception of sweet and bitter tastes. The finding facilitates further investigations for the common transduction and perceptual mechanisms in humans.

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FCOI Declarations: None.

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#304

Gustatory Contributions To Sodium Appetite: A Microstructural Analysis

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When injected with the diuretic furosemide, rats increase their intake of sodium and lithium salts. It is claimed that sodium-depleted rats seek out a salty taste. Regardless of physiological depletion, however, rats that are not thirsty prefer midrange NaCl concentrations (e.g., 0.15 M) to concentrations which are, presumably, both 'saltier' and 'less salty'. Meal pattern analysis (i.e., microstructural analysis), in which the onset time of each lick of a liquid 'meal' is recorded, has been used to make inferences about the relative contribution of appetitive and aversive tastes, and motivational state, to total intake. In this experiment, furosemide-injected (8 mg/kg, s.c.), sodium-restricted, rats were presented with one of 3 concentrations of NaCl (0.05, 0.15, and 0.32 M), 0.15 M NaCl mixed with 100 mM amiloride, 0.032 M Na carbonate, or 0.3 M sucrose in 24-minute sessions in an automated lickometer. In familiarization sessions, all rats were also previously tested with water while thirsty. Rats differed in total licks depending on test substance ($F(5,47) = 17.7$, $p < 0.001$). Total licks for 0.15 M NaCl ($M = 3074$) and 0.05 M NaCl ($M = 2743$) did not differ (Tukey-HSD); amiloride reduced licking for 0.15 M NaCl ($M = 2177$) to levels equivalent to 0.32 M NaCl ($M = 1919$) unadulterated. However, tastant did not affect either the number of bursts of licking (runs of 3 or more licks separated by pauses of less than 1 s) or the size of bursts ($F(5,47) = 1.78$, $p = 0.13$ and $F(5,47) = 2.22$, $p = 0.07$ respectively). Unexpectedly, we found that the median interlick interval for rats licking water varied as a function of group ($F(5,47) = 18.0$, $p < 0.001$), with the (future) amiloride and carbonate groups ($M = 153.3$ ms) similar to each other and significantly lower than each of the other 4 groups ($M = 174.7$) which did not differ from each other. As the amiloride and carbonate groups were formed from rats purchased from a different breeder (Charles River) than the other groups (Harlan), this suggests caution in comparing microstructure across the 6 groups. Subsidiary analysis focusing on the NaCl and sucrose groups were largely consistent with results reported above.

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FCOI Declarations: None.

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#305

Parabrachial Nucleus Melanocortin Receptor Agonist Injection Modifies Parabrachial Gustatory Neuron Responses

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Although known to influence gastrointestinal feedback and satiety, we discovered that the melanocortin receptor (MCR) agonist melanotan II (MTII) also affects hedonic taste evaluation (Baird et al., 2011). We also found that these effects are blocked when the MCR antagonist SHU-9119 is preinjected into the parabrachial nucleus (PBN). Here we used hybrid electrodes to microinject MTII into the vicinity of single PBN gustatory cells during *in vivo* extracellular recording in rats. After each PBN neuron was isolated, its gustatory response to 4 orally applied taste solutions (0.1M NaCl, 3mM quinine hydrochloride; QHCl, 0.5M sucrose, and 0.03M citric acid) and dh_2o was profiled twice. Then MTII (0.24nM/100nL) or vehicle was ejected from a pipette glued to the metal electrode and spontaneous activity was recorded for 10 minutes. Taste responses were recorded up 120 minutes or until isolation was lost. Six neurons were recorded after vehicle injection and 11 neurons after MTII injection. Overall, there was no significant effect of vehicle on taste responses or spontaneous activity. MTII significantly reduced spontaneous activity ($p<0.05$), but not in all cells. There was a significant reduction of net taste responses (controlling for spontaneous rate) overall ($p<0.05$). However, the reduction was specific mostly to NaCl, citric acid, and sucrose responses (not bitter QHCl), which resulted in a significant reduction of breadth of tuning (H; $p<0.05$). MTII had no effect on net water responses indicating that the drug effect was selective to gustatory responses. In 6/11 neurons the taste response was abolished within 5 minutes; however, in 4 of these neurons the response recovered within 75 min of the injection. Overall, taste responses were lowest in the first 30 minutes after injection with the cells showing significant recovery for citric acid and sucrose responses but not NaCl responses within the second 30-min phase of taste testing after MTII injection. These results provide the most direct evidence to date that MCRs in the PBN influence taste processing.

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FCOI Declarations: None.

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#306

Local Field Potential Oscillations Reflect Stimulus Dependent Activity in the Rat Solitary Nucleus

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The rodent taste system exhibits task-dependent and task-relevant synchrony between neural activity in several of its constituent parts. However, the role of synchronous neural activity in the brainstem (where early taste processing occurs) has not been investigated. We analyzed recordings from rats with chronically implanted 8 channel recording electrodes in the nucleus of the solitary tract (NTS). Both isolated single unit spiking activity and local field potentials (LFP) were recorded. During these sessions, freely moving rats accessed a computer controlled lickspout which dispensed small (~12 microliter) volumes of tastants. Tastant trials occurred in blocks of 5 deliveries of a particular stimulus. To assess the information content of the LFP signal and of the timing of spikes relative to the LFP, the raw LFP from each recording was filtered (and instantaneous phase & power values were obtained) via continuous wavelet transform into 55 frequency bands lying between 0.5 and 200 Hz. Information content of the LFP was examined by using phase & power information from individual trials to train a classification model and examining mutual information between the model's predictions and true class labels on a validation dataset. The information content of the relationship between spikes and LFP signals was assessed by representing each individual spike as a vector of instantaneous phase/power values from the time of the spike, and repeating the same analysis. Spikes were assigned class labels and used in the analysis if they occurred within 300ms of a tastant delivery. Across most subjects, amplitude changes in low-frequency LFP oscillations contained sufficient information to identify trial type with 3 to 5 times chance accuracy. Additionally, for a subset of cells some additional information was present in the timing of spikes relative to components of the LFP. These results suggest that LFP oscillations in the NTS reflect processing of sensory information, and maybe contribute to the construction of temporally precise patterns of spiking activity.

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#307

Ventral Tegmental Area Connectivity Suggests Processing of Taste-related Inputs with both Positive and Negative Hedonic Value

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Although the ventral tegmental area (VTA) is well known to be an important node in the neuronal network underlying reward, recent evidence indicates its activity also correlates with aversive stimuli (Lammel et al. 2013). We used neuroanatomical approaches to help elucidate the role of the VTA in reward-related taste and feeding. Anatomical tracing in C57BL/6J mice revealed inputs to the VTA from the parabrachial nucleus (PBN), central amygdala (CeA), and gustatory cortex (GC). Projections from the GC and PBN overlapped regionally in the anterior VTA, suggesting a role for descending modulation. The VTA is composed largely of dopamine (DA) neurons and, to a lesser extent, GABAergic neurons (Roeper 2013, Taylor et al. 2014). We also examined tyrosine hydroxylase (TH) immunocytochemistry with taste-evoked Fos expression in GAD-GFP transgenic mice. Surprisingly, immunohistochemical Fos expression was found in VTA DA (TH-positive) neurons when mice are stimulated intraorally with not only 1 M sucrose but also 0.03 M quinine, a bitter and aversive stimulus. These two stimuli, differing in hedonic valence, also activated similar numbers of PBN neurons projecting to the VTA. Quinine is not eliciting a pleasurable response, nor do mice avidly consume it, rather its activation of DA neurons suggests that the VTA is integrating taste input with other information relevant for reward functions, such as novelty or anticipation.

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FCOI Declarations: None.

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#308

POSTER SESSION III
FRIDAY, 8 – 10:30 AM

A Mathematical Model of the Impact of Convergence and Inhibition on Neurons in the Nucleus of the Solitary Tract

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The phenomena of broadly tuned neurons within the gustatory system remains a vexing problem, giving rise to alternative coding theories of labeled lines versus across neuron patterns. Several studies have observed that central neurons are more broadly tuned compared to peripheral afferents, suggesting afferent convergence. Other data suggest that tonic central inhibition narrows response profiles, at least partially offsetting the increase in breadth of tuning (BOT) which would otherwise occur. To systematically study the impact of convergence and inhibition on firing frequency and BOT in the rostral (gustatory) nucleus of the solitary tract (rNST), we constructed a mathematical model of its two major cell types: projection neurons and inhibitory neurons. First, we fit a conductance-based neuronal model to data derived from whole-cell patch clamp recordings of phenotypically identified neurons in a mouse expressing VENUS under the control of the VGAT promoter. Then we used (previously published) *in vivo* chorda tympani taste responses as afferent input to the modeled neurons and assessed how the degree and type of convergence of these afferents influenced the output frequency and BOT in our model cells. We compared this model cell output to *in vivo* (previously published) gustatory responses from the rNST. Finally, we assessed how pre-synaptic, postsynaptic, and feed-forward network inhibition impacted model cell output for comparison with the *in vivo* data. The results of our simulations demonstrate (1) increasing numbers of convergent afferents result in proportional increases in model output firing frequencies, but produce an increase in model output BOT only for the first two convergent afferents; additional afferents produce relatively little increase or even a decrease, (2) convergence of afferent input selected from the same best-stimulus class of CT afferents produces a better fit to real data from the rNST compared to convergence of randomly selected afferent input, (3) inhibition typically narrows the breadth of tuning and is necessary to more realistically model the *in vivo* rNST data.

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FCOI Declarations: None.

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#309

Characterization of Induced Pluripotent Stem Cell derived Olfactory Receptor Neurons

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With the groundbreaking work of Yamanaka and Takahashi in 2006 it became possible to generate induced pluripotent stem cells (iPSCs) from adult human somatic cells. We could prove that our generated iPSCs from human hair derived keratinocytes share the same characteristics like embryonic stem cells. They have a high proliferative potential and exhibit the capacity to differentiate into all three germ layers. With a self-established protocol iPSCs can be differentiated into olfactory receptor neurons (ORNs) together with a mixture of other neurons. The generated ORNs were characterized at the age of 160 days *in vitro* and could show positive staining for olfactory marker protein (OMP) and other olfactory markers. A non-invasive somatic cell source are keratinocytes from plucked human hair which grow out of the hair root. These cells can be reprogrammed subsequently via a lentivirus containing the four so called Yamanaka factors *OCT4, SOX 2, KLF4 and c-MYC* (OSKM) to undergo iPSC formation. IPSCs can be kept feeder free on Matrigel coated plates and can be directly used for further differentiation. The differentiation protocol involves several suspension and adherent steps and spans over at least 100 days. The result is a mixture of all kind of neurons with a higher amount of ORNs. To achieve a larger number of ORNs special cytokines and growth factors have to be added at different time points in the differentiation process. *In vitro* generated ORNs were fixed and stained against established olfactory marker, like OMP, ASCL1 or TUBB3 after 160 days. Here we could show, that our iPSC derived ORNs are positive against OMP, ASCL1 and partially against TUBB3. To prove that the antibodies work in our hands we stained human olfactory epithelium and could see the characteristic staining patterns. We could generate iPSCs from primary human hair derived keratinocytes and prove that they fulfill all requirements for pluripotent stem cells. With our self-designed differentiation protocol we could generate neurons which can be positive stained for several olfactory markers.

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#310

Differentiation and Characterization of Induced Pluripotent Stem Cell (iPSC) Derived Olfactory Receptor Neurons (ORNs)

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The inner cell mass of early embryos can be isolated as embryonic stem cells (ESCs) and have the potential to proliferate and to differentiate into all three germ layers. The overexpression of specific factors leads to a reprogramming of somatic cells to induced pluripotent stem cells (iPSCs) which are like the ESCs pluripotent and have a high proliferative potential. With the iPSC model it is not only possible to create patient specific cell lines to investigate diseases, it is also feasible to differentiate and characterize specific cell types which are not easily to be obtained like neurons or in our case olfactory receptor neurons (ORNs). For reprogramming we use keratinocytes from plucked human hair as somatic cell source. This method is a well-established non-invasive possibility to gain cell samples for reprogramming compared to the widely used method with human skin fibroblasts. After approximately four weeks after infection with a lentivirus containing the four reprogramming factors *OCT4, SOX2, KLF4* and *c-MYC,* iPSC colonies can be characterized for pluripotency via staining and germlayer differentiation. A lot of neuronal differentiation protocols are available, but until now there is none for ORNs. Our new established protocol includes several adherent and suspension steps with addition of various factors at different time points, mimicking the development of the ORNs, to achieve a higher amount of ORNs in our final neuronal culture. The ORNs in the neuronal culture were characterized via diverse methods like qRT-PCR and immunofluorescence for specific markers like OMP (olfactory marker protein) and functional assays like odorant depending calcium-imaging and electrophysiological experiments. We reprogrammed keratinocytes from plucked human hair to iPSCs and differentiated those to a neuronal culture with an increased amount of ORNs and proved their function with calcium responses after odorant stimulation.

Funding Acknowledgements: Institute of Neuroanatomy, Tuebingen University.

FCOI Declarations: None.

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#311

Network Formation and Regeneration in the Olfactory System of Xenopus laevis

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Comprehending the mechanisms that make lifelong neurogenesis possible has a clear interest for the better understanding of basic principles that govern cellular and molecular interactions in the nervous system, as well as a relevant clinical interest. The general inability of the nervous system to generate new neurons in order to replace those that have been lost is a formidable obstacle to recovery from neuronal damage caused by injury or neurodegenerative disease. The olfactory system is ideal to study the process of neuronal recovery, as it is known for its lifelong capacity to replenish olfactory receptor neurons lost during natural turnover, as well as its remarkable ability to regenerate after severe lesion. For this to be possible, neuronal stem cells must go through several stages of maturation, including proliferation, migration, differentiation, and integration, to become fully embedded in an existing neural circuitry. Here we investigated the timing of degeneration and subsequent regeneration of the receptor neuron population after transection of the olfactory nerve of larval *Xenopus laevis*. Results obtained using immunohistochemistry, as well as neuronal labeling and functional calcium imaging, indicate that neuronal cell death peaks 48 hours after nerve transection. Proliferating epithelial stem cells are quickly upregulated after lesion. Supporting cells maintain both morphological and functional integrity. The olfactory epithelium recovers its original morphology 1 week after transection, at which time the first axons reach the olfactory bulb. Only spontaneous activity of mitral/tufted cells is observed in the olfactory bulb during the first weeks after transection. After 3-4 weeks first glomerular responses were observed upon epithelial odor stimulus application, but the response and glomerular morphology were still clearly altered as compared to control. 3 weeks later olfactory bulb morphology and glomerular responses seem to have fully recovered, indicating that the olfactory system of larval *Xenopus* recovers morphologically and functionally 6-7 weeks after nerve transection.

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FCOI Declarations: None.

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#312

Isolated Progenitor Cells from Human Lingual Epithelium can be differentiated into functional Type II-like Bitter Taste Cells in vitro

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Taste cell progenitors give rise to different mature taste cell types *in vivo*, however little is known about molecular differentiation mechanisms. This study aimed to culture taste progenitor cells from human lingual biopsy tissue *in vitro* to gain new insights into human taste cell differentiation. We used biopsy samples from human lingual epithelium containing taste buds from fungiform and circumvallate papillae to obtain primary lingual cells. Different isolation protocols, media conditions and feeder cell based cultivation were tested systematically. Isolated cell populations were characterized using RT-PCR-based marker analysis as well as HTS-compatible cell-based functional assays, e.g. calcium imaging. We were able to isolate several proliferating cell populations from human lingual epithelium. Molecular analysis revealed that the isolated cells express several known markers of taste progenitor cells including KRT5, KRT14, and SOX2. To assess the differentiation potential of the isolated progenitor-like cells, differentiation was induced by culture media supplements such as fetal bovine serum. As a result of supplementation, we observed the differentiation of progenitor-like cells into cells which showed the spindle-like morphology of taste cells. Calcium imaging analyses of differentiated cells revealed functional responses to bitter taste stimuli including saccharin and denatonium benzoate whereas progenitor-like cells do not respond. Taken together, these experiments show that taste progenitor cells can be isolated from human lingual tissue. Moreover, these cells can be differentiated into type II-like taste cells which functionally respond to taste stimuli. Current research addresses differentiation capacity as well as the identification of factors driving the differentiation of lingual progenitor cells into mature taste cells.

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#313

Chronic Inflammation Brought on by Obesity Modulates Regenerative Capacity of Murine Taste Buds

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According to the National Institutes of Health, over two-thirds of adult Americans are overweight and over one-third are obese. Obesity is associated with an increased risk of diabetes, heart disease, and high blood pressure, among other serious health problems. Psychophysical research on obese humans, and behavioral recordings of animal models of obesity have shown that obesity promotes altered taste sensitivity and preference when compared to lean individuals, reversible with weight loss, suggesting that taste is modulated by ongoing obesity. Using a dual-cohort mouse model, we demonstrate the effects of obesity on the morphology and molecular expression patterns of the taste bud. Taste stimuli are recognized by receptors on the apical surfaces of taste cells. We hypothesize that obese mice experience a dysregulation of taste transduction machinery brought on by their obesity, representing a potential mechanism for the differences in sensitivity to taste stimuli from lean controls. Taste cell turnover is regulated by a complex network of signaling pathways, involving a unique balance between self-renewal, differentiation, and degeneration. We aim to show that chronic exposure to a high-fat diet may result in disruption of the balance between these pathways, leading to an overall reduction in the rate of taste cell renewal.

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FCOI Declarations: None.

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#314

Gene Therapeutic Rescue of Olfactory Function: Beyond the Periphery

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Ciliopathies are a group of genetic disorders that exhibit penetrance in multiple organ systems, including the olfactory system, resulting in olfactory dysfunction and anosmia through loss of cilia formation and function. Although progress has been made in identifying the genetic etiologies of anosmia, true curative therapies are currently unavailable. Previously, we demonstrated that mice expressing a hypomorphic mutation of *Ift88*, a gene necessary for cilia formation and function, had severely impaired olfactory function. Moreover, using adenoviral-mediated gene therapy, we rescued ciliation defects and peripheral odorant responses in these mice. However, the extent to which central olfactory circuits were rescued remained unclear. Here we examined olfactory dysfunction in mice lacking IFT88 in olfactory sensory neurons (OSN; OSN-*Ift88*$^{Fl/Fl}$) to elucidate the extent to which gene therapy can restore odor detection, axon guidance, glomerular patterning, and odor-guided behaviors. OSN-*Ift88*$^{Fl/Fl}$ mice exhibited substantial loss of OSN cilia, despite no change in the cellular composition of the olfactory epithelium. OSN-*Ift88*$^{Fl/Fl}$ mice also exhibited deficits in OSN function, which was supported by reduced electro-olfactogram responses, decreased tyrosine hydroxylase staining in the olfactory bulb, and aberrant odor-guided behaviors. Furthermore, by crossing OSN-*Ift88*$^{Fl/Fl}$ mice to M71-IRES- tauLacZ reporter mice (M71-LacZ-OSN-*Ift88*$^{Fl/Fl}$) we observed defects in axon targeting and glomerular patterning of OSNs expressing the M71 olfactory receptor. Importantly, adenoviral delivery of wild-type IFT88 to OSN-*Ift88*$^{Fl/Fl}$ mice restored both OSN functionality and behavioral impairment as well as axon targeting and glomerular patterning of the olfactory bulb. Together, these studies demonstrate that gene therapy can restore higher order olfactory function caused by loss of function mutations in cilia.

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FCOI Declarations: None.

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#315

POSTER SESSION III
FRIDAY, 8 – 10:30 AM

Evaluation of Irritation Potential of Chlorine Dioxide in Healthy Human Volunteers and Asthmatics

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Chlorine dioxide and a control chemical with irritation potential (ammonia) were evaluated for their ability to elicit an irritation response in both nasal and ocular mucosa among healthy and moderate asthmatic evaluators. To establish the irritancy of each of the chemicals, four types of thresholds were obtained: thresholds for the absolute detection level for perceiving intra-nasal sensory irritation, intra- ocular sensory irritation, combined ocular and nasal sensory irritation and a threshold for the absolute detection level of the odor. Additionally, participants rated perceived intensity of the odor, irritation and annoyance for each chemical. Evaluations of pulmonary function, using spirometry, were obtained following each exposure and were compared to that individuals' baseline for that day. The results shown are from 25 healthy and 15 asthmatic volunteers who completed the experiment (both ClO_2 and NH_3 exposures). For chlorine dioxide, the ocular and nasal irritation thresholds were higher for asthmatics than for healthy controls. The majority of healthy controls had ocular and nasal irritation thresholds above 0.1 ppm while all asthmatics' thresholds were above this concentration. Self-rated intensity, irritation and annoyance of chlorine dioxide vapor at and above sensory irritation thresholds fell between 'weak' and 'moderate'. Interestingly, while asthmatics were less sensitive to the irritation of both compounds, they were more sensitive to the perception of the ammonia odor. Importantly, no evidence of changes in pulmonary function following either chlorine dioxide or ammonia exposures were observed among either asthmatics or healthy controls.

Funding Acknowledgements: SCJohnson.

FCOI Declarations: None.

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#316

Trigeminal EEG Source Localization in the Pathophysiology of the Migraine

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In migraine, the trigeminal nerve is considered to be intricately involved in pathophysiology. However, it is not known, whether there are differences in the processing of trigeminal stimuli b In migraine, the trigeminal nerve is considered to be intricately involved in pathophysiology. However, it is not known, whether there are differences in the processing of trigeminal stimuli between migraine patients and healthy subjects. Aim of the present study was to investigate whether different ways of trigeminal activation would produce different patterns of brain activation in migraine patients compared with healthy subjects. The study was performed in two groups of patients (PM...migraine without aura, PMA...migraine with aura) compared with healthy controls (N). Each group involved 17 participants. After a screening session we recorded a 128channels EEG whilst the first branch of the trigeminal nerve was stimulated using 3 different conditions: 1) cutaneous mechanical stimulation with air puffs, 2) cutaneous electrical stimulation, 3) intranasal chemical stimulation with gaseous CO2. The grand average was computed for each condition. To look at the differences in topography distribution of the electrical field a series of T-test, randomization-test and dissimilarity have been performed between conditions and groups as well as the source analysis for each group and each condition. The PMAgroup performed significantly better in lateralizing the intranasal trigeminal stimulation in comparison to N and PM. In healthy controls, the electrical topographical distribution on the scalp was significantly different among the 3 conditions. Source analysis were within the first 100ms located in the bilateral frontal and prefrontal cortex, primary and secondary somatosensory cortex, motor and premotor cortex and cerebellum. In migraine patients, PMA patients had under CO2condition activities in more numerous brain areas. In the electrical and puff condition, there was more cortex stimulation in the PMA with a strong response inside amygdala, hippocampus and parahippocampal gyrus. Amygdala and hippocampus were already known to be associated with modulation of pain, our results suggest a primary involvement of those structures specifically in migraine.

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FCOI Declarations: None.

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#317

The Effect of Acupuncture on Olfactory Function in Patients with Post-infectious Olfactory Dysfunction

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The possibilities of therapy for patients with post-infectious olfactory dysfunction are restricted. Based on a study by Hauswald et al. 1998 we tested the effect of acupuncture on olfactory function in patients with post-infectious olfactory dysfunction in a randomized, single-blinded, placebo-controlled study. 60 patients with post-infectious olfactory dysfunction were included and were randomly assigned to two groups (verum- and placebo-acupuncture). Every patient received 12 acupuncture treatments. Before and after treatment a sniffin' sticks-test TDI-score (comprehensive score of odor threshold, discrimination, and identification) was performed. At baseline, patients did not differ in terms of olfactory function, age, and duration of the disease. After acupunctur, within the groups a significant improvement of the TDI-score was found in the verum-group, which was not seen in the placebo group. The duration of the disease and the TDI-difference correlated significant; no correlation was found between age, baseline TDI score and TDI score difference. Our data indicate, that acupuncture might be an alternative therapy for patients with post-infectious olfactory dysfunction. Best results were achieved with a short duration of disease. Older people as well as anosmics can benefit from this treatment.

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FCOI Declarations: None.

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#318

The Left Versus Right Nostril Odor Detection Test for Early Alzheimer's Disease: Increased Sample Size, Changes with Progression of Alzheimer's, and Performance with other Neurodegenerative Diseases

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To further assess a preliminary study of a quick and inexpensive odor detection test that showed evidence of high sensitivity and specificity for detecting early Alzheimer's disease (AD) (Stamps, Bartoshuk, Heilman, 2013) as well as determine the effects progression of AD, and changes associated with other dementing disorders. 107 new participants were added to the 94 of the prior study [51 AD, 52 mild cognitive impairment (MCI), 65 with other causes of dementia (OD), and 33 cognitively normal, healthy controls (C)]. After examining for nasal polyps, a container of 14g of peanut butter was raised from the bottom of a 30 cm ruler, 1cm per exhale. Upon detection, the distance from their nostril to the stimulus was measured. Each nostril was tested separately.Patients with AD were significantly more impaired at detecting an odor with their left nostril (mean distance = 5.9 cm) than with their right (mean distance = 13.6 cm) ($p<0.001$) and this asymmetry was significantly present only in the AD group ($F(3,197)=23.1$, $p<0.0001$). As AD progresses, the mean odor detection distance of the right nostril decreases significantly ($p<0.001$), until it is as equally impaired as the left nostril at 3.9 cm. While patients with other forms of dementia may have deficits of odor detection, this left versus right nostril odor detection test for early, but not late AD continues to show high diagnostic sensitivity and specificity.

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FCOI Declarations: None.

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#319

POSTER SESSION III
FRIDAY, 8 – 10:30 AM

Clinical Profiles Identify Individual Differences in the Impact of Oral Sensory Damage

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Oral sensory variation arises from anatomy, genetics, and nerve damage, but the role of damage is elusive because its sensory outcomes are hard to predict. Accounts of oral dysfunction show that whole- mouth sensation rises or falls in some cases and not at all in others, and these irregular effects have confounded a definitive model of oral sensory pathology for over a century. Four cranial nerves support whole-mouth sensation, suggesting that distinct patterns of nerve damage yield specific oral sensory outcomes. We have shown that regional oral sensory loss generates contralateral compensation that often sustains whole-mouth sensation, consistent with reduced inhibition among cranial nerve projection areas. However, oral disinhibition does not occur in everyone, and we believe that two sources of variation govern these outcomes: lingual anatomy (i.e., fungiform papilla expression) and sensory loss elsewhere in the mouth. Thus, we use lingual anatomy as an index of native oral sensory function and view sensory deviations from it as evidence of nerve damage, as this strategy reveals dysfunction against a background of normal oral sensory variation. As proof of principle, here we present spatial taste profiles across the range of oral anatomy to show that different combinations of anatomy and loss can produce whole-mouth outcomes that are indistinguishable from healthy sensation. For example, midrange whole-mouth sensation may reflect midrange anatomy + healthy function, lower papilla expression + mild regional loss and disinhibition, or high papilla expression + extensive damage. These profiles serve as a guide for clinical assessments of oral sensory function, demonstrating that spatial and anatomical data are essential for the proper interpretation of whole-mouth testing. Overall, we have identified factors modulating the impact of regional oral sensory nerve loss on whole-mouth sensation. These individual differences may explain the diverse clinical manifestations of flavor-related nerve damage, enabling a long-sought comprehensive model of oral sensory dysfunction that aids diagnostic efforts.

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FCOI Declarations: None.

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#320

Evaluation of the Olfactory System after a Mild Traumatic Brain Injury

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Olfactory dysfunctions are frequently reported by patients after a traumatic brain injury (TBI). In fact, a recent study reported that more than the two thirds of the participants present impairment (hyposmia) or a total loss of smell (anosmia) within few days after a TBI. To this day, no study aimed at evaluating olfactory function of the patients in the acute phase of a *mild* TBI, the most prevalent form. Here, we compared quantitative (Sniffin' Sticks) and qualitative (parosmia questionnaire) aspects of olfaction, cognitive abilities (Rivermead, Trail A-B, RCF, Digit Span, HVLT, EXACT, Codes and the Bells test) and emotional state (BDI, BAI, SF-36 and the Hamilton Anxiety questionnaire) of 9 patients with mild TBI with 9 matched controls within 1 month after the trauma. We found significantly higher parosmia scores (qualitative olfactory impairment) in the patients than in controls, but no difference in terms of quantitative olfactory measures. Moreover, patients with mild TBI presented abnormal scores in the scales of depression, anxiety, life quality, and several cognitive tests. Finally, we observed a strong and significant positive correlation between the subjective olfactory test (parosmia scores) and the depression and anxiety evaluations.These preliminary findings suggest the presence of olfactory impairment in the first month following a mild TBI, which however goes undetected with classical tests. Furthermore, our results suggest a connection between subjective alterations and emotional and cognitive impairment. We are currently analysing MRI data (diffusion and structural imaging) in order to identify the neuroanatomical underpinning.

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FCOI Declarations: None.

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#321

POSTER SESSION III
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Increased Endogenous Brain-derived Neurotrophic Factor Does Not Rescue Impaired Olfactory Neurogenesis in a Huntington's Disease Mouse Model

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Mutant huntingtin protein (mhtt) inhibits BDNF signaling, and is thought to exacerbate striatal neuron loss in Huntington's disease (HD). In HD mouse models, increasing BDNF in the striatum or ventricular ependyma slows disease progression, and diverts neuroblasts born in the adult subventricular zone (SVZ) from their normal destination, the olfactory bulb, to the striatum. Here, some cells survive and develop features of striatal neurons, raising interest in using SVZ-derived cells and BDNF therapeutically. In the HD R6/2 mouse strain, CNS pathology is accompanied by impaired olfactory neurogenesis. Migrating neuroblasts reach the bulb, however the number of surviving adult-born granule cells (GCs) is reduced, while apoptosis in the granule cell layer (GCL) is increased. Neuroblasts lack mhtt aggregates, while all bulb neurons, including GCs, contain aggregated mhtt protein, suggesting that accumulation occurs as new GCs develop. Maturing GCs also express the BDNF receptor TrkB, and mhtt's ability to disrupt BDNF signaling may contribute to GC loss in R6/2 mice. As increased BDNF promotes survival of SVZ-derived cells in the HD striatum, we tested its ability to rescue new GCs in R6/2 mice. Mice were crossed with a transgenic strain carrying a BDNF transgene under control of the alpha-CAMKII promoter. Olfactory GCs express alpha-CAMKII, and levels of bulbar BDNF were ~4-fold higher in the double transgenic mice compared to R6/2 mice. At 7.5 wks of age, mice were treated with BrdU and survival of new GCs was assessed 4 wks later. Relative to normal mice, loss of new GCs was increased by ~24% in R6/2 mice, with R6/2-BDNF mice showing a similar loss. Apoptotic cells were increased ~2.5-fold in the R6/2 mouse GCL, with R6/2-BDNF mice showing a small, but significant reduction in this. SVZ cell proliferation did not differ between R6/2 and R6/2-BDNF mice, but by 11.5 wks of age, GCL volume was reduced by ~14% in both genotypes. Both genotypes also showed a decline in tyrosine hydroxylase expression in the glomerular layer. Our results show that increased endogenous BDNF does not counteract the effects of mhtt on adult GC neurogenesis

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FCOI Declarations: None.

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#322

Localization and Ablation of p75 in the Mouse Olfactory Bulb

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The olfactory bulb (OB) receives synaptic input from olfactory sensory neurons and readily displays neuroplasticity following a loss in synaptic input. A loss of synaptic input has been shown to reduce tyrosine hydroxylase (TH) levels, the rate-limiting enzyme for dopamine synthesis, in the OB. The loss of TH, and therefore dopamine, is thought to increase OB sensitivity when sensory input is low. However, the cellular mechanisms responsible for these changes are unknown. Previous experiments have shown that activating the p75 neurotrophin receptor lowers TH levels. In the first experiment, we sought to determine if p75 is expressed by TH interglomerular neurons. ImageJ tissue analysis revealed that P75 and TH failed to colocalize on interglomerular neurons across three animals. In a second experiment, we sought to cause OB plasticity by ablating p75 expressing cells. Mice were immunolesioned with a p75 targeted saporin toxin on the dorsal surface of the left OB. A control toxin was used on the dorsal surface of the right OB. After six days of recovery, immunohistochemistry was performed to determine the expression and location of p75 within the OBs. ImageJ was used to quantify p75 fluorescent signal. We observed a 6-fold decrease in p75 signal for the lateral region of the immunolesioned OB compared to the control OB (n=2). We also aimed to identify if GFAP signaling would be affected by immunolesion. Present data displays no significant change in GFAP labeling after immunolesioning. Ongoing studies are being performed to determine if p75 labeling is colocalized with glia cells and if the decreased p75 labeling correlates with decreased TH expression. Additionally, studies are being performed to see if a measurable difference in the location and amount of c-Fos expression can be observed in mice from p75 immunolesion, as well as intranasal triton delivery. With these and future data, we can begin to provide a better explanation of p75's location and role in neural plasticity, specifically TH expression, within OB circuits.

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FCOI Declarations: None.

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#323

Differential Expression of ZEB2 in Olfactory Bulb Interneurons and Glia

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Zinc finger E-box binding protein 2 (ZEB2; zinc finger homeodomain 1b (ZFHX1b); SMAD interacting protein 1 (SIP1)) is a transcription factor with several established roles in the developing nervous system, including cell fate specification. Previous studies have reported that the ZEB2 is also expressed in the adult olfactory bulb (OB), but the role of ZEB2 in the adult OB and neurogenic niches that regenerate OB interneurons is not well characterized. In this study, we have used in situ hybridization and immunohistology to identify the OB cell types expressing ZEB2. In situ hybridization assays revealed wide-spread ZEB2 expression throughout the OB. Immunofluorescence studies, however, showed only low level expression of ZEB2 in nearly all glomerular and granule cell layer OB interneurons and no detectable expression in mitral/tufted cells. By contrast, astrocytes displayed the strongest expression levels in the OB. Examination of the rostral migratory and subventricular zone also showed strong ZEB2 expression in astrocytes, but little or no expression in migrating progenitors. Together, these studies suggest that neuronal and glial cell fate specification for interneurons and astrocytes, respectively, in the OB is maintained, in part, by differential expression levels and/or alternative splicing of ZEB2.

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FCOI Declarations: None.

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#324

POSTER SESSION III
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Defining a Function of Olfactory Bulb Processing via Comparison of Input and Output

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The goal of the present study was to identify the function(s) of the olfactory bulb by comparing its output with its input. The input olfactory receptor neurons were anatomically targeted via nasal infusion with an organic calcium sensitive dye. In the same preparation, genetically encoded voltage or calcium indicators were targeted to the output mitral and tufted cells using Cre-dependent AAV transduction in a transgenic mouse (Pcdh21) that expresses Cre recombinase in mitral and tufted cells. Wide-field epifluorescence imaging was used to measure odor-evoked activity in glomeruli across ~2 log units of odorant concentration in freely breathing anesthetized mice. The olfactory bulb output was more concentration invariant than the input, indicating that the olfactory bulb participates in generating the perception of concentration invariance of odor quality. Our approach should be useful in addressing other possible perceptual functions of the olfactory bulb. Knowing the olfactory bulb function(s) can help guide investigations of the bulb's synaptic network.

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#325

Temporal Response Profiles of Olfactory Bulb Cell Types during Prolonged Odor Stimulation

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Olfactory sensory input is shaped by olfactory bulb (OB) interneuronal circuits and transmitted by mitral and tufted cells to further brain areas. We are interested in how odor information at timescales relevant to behavioral adaptation/habituation is encoded at each of these levels of the OB. We used *in vivo* two-photon calcium imaging and k-means cluster analysis to record and analyze the temporal response profiles of olfactory sensory neurons (OSNs), juxtaglomerular (JG), superficial tufted (ST), and mitral cells (MC) before, during, and after a prolonged odor stimulation. We recorded GCamp3 activity from the terminals of OMP+ OSNs and from the soma of Thy1+ JG, ST, and MCs. We found that JG responses tend to follow a pattern of strong initial activity at odor onset followed by a decaying response of varying speed and almost no activity at odor offset. However, MC responses to the same odors exhibit a broad range of temporal patterns throughout and after the odor presentation. Using cluster analysis, we classified these response patterns into statistically significant response profiles. These profiles indicate that there are MCs that reflect the decaying juxtaglomerular response pattern; that come on and go off rapidly; that maintain their response through the entire odor stimulation; and even some that initially do not respond very strongly but ramp up their activity as the odor presentation progresses. In an animal, even neighboring MCs can have diverse response patterns to the same odor and individual MCs can exhibit distinct response patterns to different odors. We are currently analyzing the temporal patterns of ST cells to see if they are more similar to JG or MC responses. Further, we are collecting data from OSNs to get a sense of how these other OB cell types reflect or transform incoming odor information.

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POSTER SESSION III
FRIDAY, 8 – 10:30 AM

Intensity Invariant Readout of Olfactory Bulb Output is Facilitated by a Specific Inter-glomerular Circuit

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Perception is often invariant for stimulus identity (eg: face recognition) even when other stimulus features (eg: light levels) vary widely. In the mammalian olfactory system, little is known about the neural mechanisms that extract odor identity, while tolerating fluctuations in concentration. We hypothesized that a specific gain-control circuit in the olfactory bulb (OB) mediated by DAT+ cells, reformats the OB output to facilitate efficient read-out of concentration-invariant odor identity by target cortical areas. To test this, we monitored the activity of numerous OB output (Mitral/Tufted) cells to odors across varying concentrations (3 orders of magnitude) using 2-photon microscopy. Individual mitral cells exhibited a diverse range of concentration response profiles (CRFs) - some monotonically increased, or decreased with increasing concentration, while others peaked at intermediate concentrations. Importantly, the mean population response increased only modestly across this large concentration range. A simple model assuming divisive normalization implemented by the DAT+ cells was able to generate the diversity of the experimentally observed CRFs. We visualized the neural population trajectories using PCA and found that for a given odor, all sampled concentrations spanned low- dimensional manifolds. Additionally, across a large odor panel, mitral cells exhibited significantly larger dimensionality compared to that of tufted cells, highlighting the differences between these two OB output channels. To assess the ability of cortical targets to correctly identify odors, we trained a linear decoder with sparse and non-negative weights to classify odor identity irrespective of concentration. The cross- validated performance was >80%, which significantly dropped when DAT+ cells were specifically ablated. We conclude that a specific interneuron circuit in the glomerular layer formats the OB population output so as to facilitate concentration invariant odor identification by the cortex. This may be the first of many such transformations that ultimately lead to perceptually stable behavior.

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FCOI Declarations: None.

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#327

The Roles of Glomerular and Granule Cell Layers in Spatial and Temporal Odor Processing

Don Tucker Award Finalist

Francesco Cavarretta[1], Addolorata Marasco[2], Michael L. Hines[1], Gordon M. Shepherd[1], Michele Migliore[1]
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The olfactory bulb (OB) is organized in many glomerular units (GUs), which interact with each other at two levels, the glomerular and the granule cell layer. This interaction sculpts the representation of odors that occurs in the mitral cells, whose axons project to the pyriform cortex (PC). These actions subserve several crucial functions for odor recognition. However, neither the relative roles of the layers, nor how learning effects their computation, is yet understood. To address these problems, we used a realistic large scale 3D model of the OB which includes both the glomerular and the granule cell microcircuits. It was constrained to the experimental findings and used to unravel several essential features of OB organization, such as the spatially sparse and segregated organization of granule cells located below a glomerulus, which result from GU interactions during learning. Here, we show that the glomerular circuit transforms a dense and disorganized spatial representation (such as that experimentally observed for natural odors) into a sparse, normalized and contrast-enhanced one. This is essential for odor learning. Subsequently, over time, the granule cell circuit decreases the spatial representation overlaps of different odors, increasing the relative differences and improving the discrimination. This effect improves with the number of learned odors but decreases if they are too many. In the last case, the columnar organization of the granule cells is destroyed, which might be restored or avoided perhaps by neurogenesis or neuromodulation. Taken together, the results provide new insight into the fundamental role of GUs and their layered organization. The model predicts the input received at the PC, building the basis for further investigations in models that include it.

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FCOI Declarations: None.

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#328

POSTER SESSION III
FRIDAY, 8 – 10:30 AM

Sniffing Strategy as a Clue for Olfactory Discrimination Improvement: Neural Correlates

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Gathering of sensory information largely depends on sampling dynamics. In olfaction, the active sampling process is referred as sniffing (2-10Hz). At the behavioral level, it is well established that sniffing is highly dynamic, especially in frequency and flow rate (Wesson et al., 2008). The importance of sniffing frequency in animal's performance has been extensively evidenced (Abraham et al., 2004; Rinberg et al., 2006; Uchida and Mainen, 2003). Moreover, we showed that sniffing is partly modulated as a function of odorant molecules but is also adjusted synthetically depending on the odorant context in a discrimination task (Courtiol et al., 2014). At this stage, we wondered to what extent i) sniffing strategy in a discrimination task was acquired by experience and ii) such sniffing acquisition could be accompanied by neural correlates in olfactory, motor and limbic structures. For that purpose, rats were trained to discriminate odors in a double choice discrimination task while we simultaneously recorded both sniffing activity (using a whole-body plethysmograph) and neuronal signals (Local Field Potentials, LFP) in olfactory (olfactory bulb, olfactory tubercle, piriform cortex) and non-olfactory structures (hippocampus, striatum, cerebellum). We showed that most of sniff modulations were related to the procedure learning and were necessary for the acquisition of the discrimination task. At the neuronal level, we were looking for correlations between respiratory rhythm and brain oscillatory rhythms in the theta (2-10Hz), beta (15-30Hz) and gamma (30- 60Hz) bands. Notably, we investigated the evolution of LFP coherence and information flow between cortical areas as a function of learning stage. Analyses are in progress.

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#329

A Neuroinformatic Pipeline for Investigating Intra-laminar Heterogeneity in the Olfactory Bulb

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The modular, topographic organization of inputs to the olfactory bulb has long been appreciated. It is far less clear, however, whether this modularity is preserved, maintained, or otherwise altered in the intrinsic circuits of the bulb. Is the bulb comprised of a single invariant columnar unit tiled across space, or is it a system of functionally heterogeneous modules, organized in parallel? To begin investigating this question, we have employed a neuroinformatic strategy utilizing the in-situ hybridzation (ISH) maps of the Allen Brain Atlas -- a highly standardized compendium of gene expression for the C57 mouse brain, collected at cellular resolution. ISH images from the Atlas cataloging the expression of 49 Potassium Channel genes were nonlinearly registered to a common coordinate frame aligning mitral cell layers (MCLs) across images. Line profiles charting expression energy along the MCL were extracted, and PCA -based K-means clustering was used to identify candidate 'spatial modes' of potassium channel gene expression across the bulb. The first three such modes captured ~ 70% of expression variance across the MCL, and exhibited strong dorsoventral patterning and periodicity. These preliminary results indicate that there is substantial "zonal" patterning of potassium channel gene expression in the mitral cell layer. We are presently scaling our pipeline to encompass all genes (~20K) of the mouse genome.

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#330

Effects of Odors on Line Bisection in Teenagers

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One way of assessing nonverbal cognition is line bisection. It is predicted that a pleasant odor would impact upon accuracy of line bisection.On a convenience basis, 30 normosmic (3/3 Pocket Smell Identification Test), high school students**,** in an alternating order, were presented with either a vanilla odorized or unodorized surgical mask, worn for 1-minute and then bisected a line. After a 3-minute washout period, the task was repeated with the alternate mask. Subjects rated the hedonic perception of the odor and the distance from their marked bisection to the midpoint was measured.For the entire data set, seven showed greater accuracy in line bisection with the odor and 14 showed greater accuracy with the blank mask. Nine showed the same accuracy with a blank mask as with the odorized mask. Using a sign test with two tailed P-values, no statistical significance was found. Thus, using the odor neither improved, nor reduced the accuracy of line bisection. Looking at only those with positive hedonics towards the odor (n=20), four showed improvement, eight showed worsening, and 8 were identical. The sign test looking at two sided P-values, again demonstrated no significance (p=0.39). In an analysis of those with negative hedonics towards the odor (n=10), three showed greater accuracy with the odor, six showed greater accuracy with the blank, and one had no change. The sign test, looking at two tailed p-values, demonstrated no significant effect (p=0.51).The odor did not impact line bisection. Of those who viewed vanilla aroma in a hedonically favorable or negative manner also demonstrated no significant effect on line bisection. Further investigation using other paradigms to test the effects of odor and visual perception are warranted.

Funding Acknowledgements: Smell & Taste Treatment and Research Foundation, Chicago, IL USA.

FCOI Declarations: Alan R. Hirsch MD has ownership in the Smell & Taste Treatment and Research Foundation and consults with multiple companies and health care facilities.

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Taste-enhancing Effects of Egg Peptides Increased in High-temperature Stored Mayonnaise

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Peptides, which are produced by enzymatic proteolysis, heating, or fermentation, change the taste of foods. One of the changes is a taste-enhancing effect called “koku” in Japan, which typically enhances the food flavor including persistency. This effect has widely been utilized in the traditional and industrial food processing industries in recent years. Although egg is one of high-quality protein sources in the food industry, the “koku” effect of egg-derived peptides has not been studied in detail. The objective of this study was to investigate the koku effect of egg-derived peptides using thermal modified mayonnaise. Water-soluble extract from high-temperature stored mayonnaise (55 °C, 3 weeks) was freeze-dried and a resulting slight yellow powder (HTS-mayo) was evaluated in a Chicken bouillon by 12 trained panelists using the quantitative descriptive analysis method with a 7 point scale. Low-temperature stored mayonnaise (LTS-mayo, 5 °C, 3 weeks) was used as a control sample for each test. HTS-mayo (0.7%) showed a significantly stronger effect on enhancing the persistency of the bouillon taste compared with LTS-mayo. High-temperature storage caused develops of browning which measured as an absorbance at OD 450 nm. The increments of free amino acid as well as bound-type amino acid were determined by high-temperature storage. There were marked changes in some free amino acids (e.g. Leu, Phe, and Ala), which increased to 130-190%. These results indicate that increased hydrolysis of egg proteins and the occurrence of the Maillard-type reaction in mayonnaise during high-temperature storage create a taste-enhancing effect including persistency.

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#332

Suppression of Bitter Taste by Stearic Acid Microspheres that are Embedded in Edible Taste Strips

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The bitter taste of many drugs complicates the administration of medications to children. A related concern is that young children may have problems ingesting drugs in capsule or tablet form. The development of novel methods for dispensing medications to children is an important objective. One approach to alleviate these problems is to minimize the bitter taste of a drug, and to develop delivery methods that do not use capsules or tablets. The bitter taste stimulus sucrose octaacetate (SOA) was used as a model compound for bitter tasting drugs. In order to minimize the bitter taste of SOA, this carbohydrate was encapsulated in stearic acid-coated microspheres, which were then incorporated into edible strips that contained the sweet taste stimulus sucralose. Stearic acid microspheres were prepared by the hot-melt encapsulation method at a weight ratio of 5.5:1 stearic acid to SOA. Stearic acid and SOA were melted at a temperature of 90 °C, mixed, and poured into a rapidly stirred solution of HEPES buffer at pH 8.0 that was warmed to 50 °C. After cooling the solution, microspheres were collected and dried. The resulting microspheres varied in size from 100-500 µmeters in diameter. Colorimetric analysis of SOA content by the anthrone assay showed that weight ratios of stearic acid to SOA in dried microspheres ranged from 12:1 to >15:1. These loaded microspheres were then incorporated into thin films that were composed of pullulan, hydroxypropyl-methylcellulose, xanthan gum, and sucralose. Light microscopy studies indicated that microspheres were evenly distributed in dried films. Psychophysical studies then demonstrated that encapsulating SOA, and suppressing the bitter taste of SOA with sucralose, decreased gLMS bitter taste intensity values by approx. 40% when compared to control strips that contained only unencapsulated SOA. Also, microsphere-containing taste strips delayed the onset of bitter taste by approx. 15 seconds in our subject population. This novel approach successfully diminishes the bitter taste of SOA, and is a promising method for delivering bitter tasting drugs to young children and elderly individuals.

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FCOI Declarations: None.

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POSTER SESSION III
FRIDAY, 8 – 10:30 AM

Liminal Sensitivity for the Components of Mixtures in Rats: NaCl and Sucrose

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Psychophysical and neurophysiological assessments of the sensitivity of the gustatory system to controlled presentations of single chemical compounds have revealed some fundamental properties of stimulus efficacy and have helped define the parameters of neuronal response properties. However, taste stimuli are rarely ingested in isolation. Arguably, the gustatory system was adapted to identify and report the presence of numerous chemicals ingested concurrently. When mixtures have been studied, interactions such as suppression or synergy have been sometimes revealed. To begin systematically exploring the effect of stimulus background on detection of a target stimulus in a rodent model, we trained rats in a two-response operant task using a gustometer to respond to the presence of either NaCl (n=7) or sucrose (n=7) dissolved in water. The EC50s of the respective psychometric functions were measured and operationally defined as threshold. Subsequently, the same rats were either trained to respond to the presence of NaCl dissolved in a sucrose masker (0.3, 0.6, or 1.0 M, tested sequentially), or sucrose dissolved in a NaCl masker (0.04, 0.2, or 0.4 M). Detection thresholds were determined for the target stimulus dissolved in each concentration of the masker. All masker concentrations tested except 0.04 M NaCl increased EC50s for the target stimulus. However, the degree of shift in liminal sensitivity for either target was similar when plotted against the change in masker concentration relative to threshold for the masker. That is, sensitivity to the masker appeared related to the shift in detectability of the target stimulus when dissolved in the masker. Additional chemical combinations must be tested before these findings can be generalized, but it appears that the attenuating impact of a masker on the detection of a target stimulus is dependent on liminal sensitivity to the masking stimulus. It will be important to understand how such relations change as a function of the complexity of the mask and to begin to discern the neural circuits involved in the detection of specific taste signals in the context of noisy backgrounds.

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FCOI Declarations: None.

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POSTER SESSION III
FRIDAY, 8 – 10:30 AM

Microstructural Analysis of Water and Sucrose Consumption in Several Inbred Mouse Strains

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Our previous analysis of C57BL/6J (B6), DBA/2J, and 64 BXD progeny strains identified a significant quantitative trait locus underlying differences in lick rate. Mice vary not only in rate of licking, but in several other behavioral measures of consumption that can be assessed using "microstructural analysis". We compared 20-min consumption of water and 0.1 M sucrose in thirsty mice from a behaviorally-diverse panel of 18 inbred or congenic mouse strains. In these strains, median interlick intervals (ILI) generally averaged between 99.8 ms (SWR) to 134.4 ms (FVB). However, we identified 5 strains with slow lick rates (median ILI 149.3 - 163.4 ms): WSB, A, NZW, CAST, and PWK. Closer examination revealed that spout contacts for these strains were not as tightly rhythmic as in the other 13 strains, which may be indicative of physical (e.g., tongue length) or behavioral (e.g., hyperactivity) distinctions. In 20 min, some strains averaged 3 times as many licks to water (BTBR: 1471.8) as others (A: 495.2). Licking for sucrose was greater in every strain (range: D2, 940.2 to CAST, 2150.8), and was highly correlated with licks to water ($r = 0.929$, $p < 0.001$). Mice were not food restricted but were water-restricted during sucrose sessions. Nonetheless, as a general rule, sucrose-water differences were related to Sac taster phenotype. We also examined the patterning of "bursts" of licking (runs of 3 or more licks each separated by less than 1 s). The number of bursts across strains ranged from 9.1 (SWR) to 23.5 (BALB) for water and 14.0 (129) to 47.3 (B6) for sucrose. Interestingly, the number of bursts does not correlate with total licks in contrast to a strong correlation between total licks and the size of bursts, both for water ($r = 0.70$, $p = 0.001$) and sucrose ($r = 0.70$, $p = 0.001$). This analysis also identified strains differing strongly in ingestive style, which may prove interesting for further study. For example, of the 18 strains, SWR mice were among the lowest 2 for both water and sucrose in number of bursts, but were among the highest 2 for burst size. In contrast, B6 mice had the largest number but smallest size of sucrose bursts.

Funding Acknowledgements: University of Tennessee and Rollins College funds.

FCOI Declarations: None.

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Intermittent Home-Cage Access to Alcohol Shifts its Palatability and this Shift is Blocked by Sub- Chronic Nicotine Administration

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Previous work has shown that fetal exposure to alcohol (EtOH) shifts its palatability such that EtOH tastes less bitter to subsequently tested adolescent rats. Here, we determined whether a similar shift occurs when rats are exposed to EtOH in adulthood, and whether nicotine co-administration alters this shift. First, we tested rats in a brief-access licking paradigm in order to measure the palatability-driven, unconditioned licking to multiple concentrations of EtOH (1.25-40% v/v) and quinine (QHCl; 0.01-3.0 mM). Each stimulus was presented for three days in a counter balanced design, and curves were fit to the data of each rat using a 3-parameter logistic function. The $\log_{10}$ concentration at one-half asymptote (EC_{50}) for EtOH and QHCl were correlated (0.48, $P < 0.01$), suggesting an overlap in the processes underlying the aversion to the two stimuli. Following brief-access testing, rats were presented with an intermittent (24 h, 3x/week) two-bottle choice tests for four weeks, with 20% EtOH and water. Rats received either nicotine (0.4 mg/kg S.C.) or saline during the choice tests. Nicotine suppressed EtOH intake during the first two weeks of access ($F(1,15) = 6.26$, $P < 0.05$) compared to saline-treated rats, but intakes recovered by the end of the test such that total consumption of EtOH did not significantly differ between the two groups. Following this choice period, rats were again tested in the brief-access licking paradigm. Interestingly, the ethanol concentration-response curve in saline-treated rats was shifted rightward as a result of ethanol exposure, while the curve in nicotine-treated rats was not ($F(1,7) = 6.3$, $P < 0.05$). Because nicotine did not significantly alter total EtOH consumption, this blockade by nicotine is likely not due to differences in the amount of exposure to EtOH. These findings suggest that exposure to EtOH alters the taste-driven intake of EtOH in future exposures, and that nicotine blocks this shift by changing the representation of EtOH as a stimulus. Thus, these results may provide a basis for exploring the prevalent comorbidity existing between these two drugs of abuse.

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FCOI Declarations: None.

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POSTER SESSION III
FRIDAY, 8 – 10:30 AM

Recalled Taste Intensity, Liking and Habitual Intake of Commonly Consumed Foods

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Taste intensity and quality affect the liking of foods, and determine food choice and consumption. We aimed to 1) classify commonly consumed foods based on recalled taste intensity for bitter, sweet, salty, sour, and fatty taste, and 2) examine the associations among recalled taste intensity, liking, and habitual consumption of foods. In Stage 1, 62 Canadian adults recalled the taste intensity of 120 common foods. Their responses were used to identify sets of 20-25 foods classified as strongly bitter, sweet, salty, sour or fatty-tasting. In Stage 2, 287 U.S. adults validated these selections, and let us reduce them to sets of 11-13 foods. Ratings of recalled taste intensity were consistent across age, sex and overweight status, with the exceptions that sweet, bitter and fatty-tasting foods were rated as more intense by women than by men. The recalled intensity ratings of the most bitter, salty and fatty foods (but not sour or sweet foods) were inversely correlated with liking and intake. The negative correlation between fatty taste intensity and fatty food liking was stronger among normal weight than among overweight participants. Our results suggest that the recalled taste intensity of foods is associated with food liking and habitual consumption, but the strength of these relationships varies by taste. The food lists based on taste intensity ratings provide a resource to efficiently calculate indices of exposure to the different tastes in future studies.

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#337

Olfactory Bulb Acetylcholine Reinstates Habituated Odor Responses and Drives Behavioral Dishabituation

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The olfactory bulb (OB) receives cholinergic input from the basal forebrain (BF) and expresses a variety of cholinergic receptors. Acetylcholine (ACh) is known to be necessary for proper olfactory perception, however the mechanisms are largely unknown. To explore the effects of cholinergic modulation on odor processing, we used transgenic mice expressing calcium indicators in defined OB cell populations in intact, anesthetized mice. Overall, we find that OB ACh release via electrical BF stimulation increases the gain of glomerular odor responses over a wide odor concentration range. Further, pharmacological experiments demonstrate that this effect is mediated through the activation of OB m2 AChRs. Overall, we find this mechanism increases individual glomerular sensitivity to odors and decreases glomerular activation thresholds. Data from our lab and others suggest that habituation of MT odor responses is due to reduced OSN input and not MT cell responsivity. Thus, controlling the gain of MT cell responses could modulate the magnitude of input adaptation. Since salient stimuli increase BF neuronal activity and OB ACh release increases weak odor responses, we hypothesized that OB ACh release could reinstate reduced OB odor responses after OSN adaptation, a process known as dishabituation. Overall, we find that brief BFS following prolonged odor presentations recovers habituated odor responses to near baseline levels at both the glomerular and MT cell output levels. Based on this, we tested whether OB ACh release can lead to similar behavioral effects in a habituation/dishabituation paradigm using mice expressing ChR2 in cholinergic neurons. Preliminary data suggest that OB ACh release leads to increased investigation of habituated odorants, an effect not seen in wildtype littermates. Overall, these studies establish a novel functional role for olfactory ACh release, whereby odor representation and salience can be rapidly and dynamically modulated.

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FCOI Declarations: None.

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POSTER SESSION III
FRIDAY, 8 – 10:30 AM

Non-selective Enhancement of Olfactory Bulb Glomerular Responses following Olfactory Fear Conditioning in Awake Behaving Mice

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The olfactory bulb (OB) glomerular layer is the first site of sensory input to the central nervous system and contributes to cortical processing of olfactory stimuli and subsequent behavioural output. Recent work in anesthetized animals suggests that olfactory fear learning induces plasticity observed at the glomerular level of the OB, presumably affecting downstream processing, however the extent to which this occurs in awake animals is currently unknown. To address this, odor-evoked glomerular activity patterns were assessed in awake transgenic mice expressing GCaMP3 in excitatory post-synaptic OB cell populations. Before and after associative conditioning with foot shock, representations to a panel of similar and dissimilar odorants were imaged, allowing us to investigate glomerular activation to trained and untrained stimuli. Preliminary data demonstrates that glomerular odor responses in awake animals decrease with subsequent exposures to neutral odorants. However, fear learning produces robust reinstatement of representations for both trained and control odorants. By contrast, there were no representational changes in the control group following shock-only exposure. This suggests that non- discriminative fear conditioning non-selectively enhances odor-evoked activity in awake mice at the level of OB output neurons. This mechanism could potentially underlie behavioural generalization, the transference of fear responses from learned stimuli to related stimuli, previously reported by our lab. We are currently exploring the effects of cholinergic modulation during training on the observed plasticity. Together, these studies will further our understanding of how neuromodulation shapes behavioural generalization and plasticity at the initial stages of olfactory sensory processing in awake animals.

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Effects of Olfactory Training on Olfactory and Non-olfactory Functions and Brain Structure/Neuroplasticity

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Repeated exposure to odorants leads to enhanced olfactory sensitivity. Given that the olfactory areas are closely connected to the limbic structures, the field of action of olfactory training may extend beyond olfaction, and may affect cognitive function related to memory and emotional processing. So far, the efficacy of olfactory training on olfactory and non-olfactory functions in healthy population and its underlying neurobiological basis remain poorly known. This study investigated the effects of olfactory training on olfactory and non-olfactory functions, and brain structure/neuroplasticity. Thirty-six healthy young subjects were recruited and distributed in three groups: 1) 12 subjects were exposed to either phenyl ethyl alcohol, or n-butanol, 2) 12 subjects participated in an equivalent visual training, and 3) 12 control subjects did not participate in any training. Training lasted 6 weeks (20 min/day) during which subjects performed three tasks at the lab (intensity classification, quality classification and recognition tasks). Before and after the training period, all participants performed a series of behavioral tests (olfactory, memory and emotional tasks) and underwent magnetic resonance (MR) imaging, during which structural (cortical thickness, density) and task-related functional (BOLD response) measures were recorded. Preliminary results indicate that trained subjects improved their olfactory or visual performance in their respective training tasks throughout the 6-weeks period. In addition, subjects exposed to odorants exhibited increased olfactory function. However, olfactory training induced no or limited enhancement of non-olfactory function. Finally, ongoing MR imaging analysis will test whether improved odor perception, induced by olfactory learning, may be mirrored by increased structural features and by alterations in functional measures in olfactory processing areas. This research brings a better understanding of sensory processing and neuronal plasticity, and these new insights may serve for clinicians taking care of patients with olfactory dysfunctions.

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FCOI Declarations: None.

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#340

Intentional Learning of Odors and Pictures

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We have previously demonstrated, using an incidental learning paradigm, that olfactory and visual memory are functionally different. In the current study we compared olfactory and visual *intentional* learning with a particular emphasis on memory task. Intentional learning allows for rehearsal of stimuli. We used two types of memory tasks, including one that allows for a signal detection theory (SDT) analysis. METHODS: After a delay (15 mins - 1 wk), 101 participants completed either a yes/no monadic (1-stimulus-at-a-time) or a 3AFC task with common and uncommon odors and pictures. RESULTS: As expected, memory declined with delay in both tasks and was better for pictures than odors and for common than uncommon items. Performance with common odors on the 3AFC task was better than predicted by chance for all three delays, whereas SDT analyses of monadic trials indicated that hit rates (HR) dropped to about 50% (guessing) after a week. Correct rejection (CR) rates were relatively high (67%) and constant for common odors, but were much lower (35%) for uncommon odors. HR for common visual stimuli stayed well above 50%. For uncommon odors, a very high HR (80%) and low CR rate indicate a liberal criterion. DISCUSSION: The results from the monadic task suggest that even when rehearsal is possible, memory for common odors is based on novelty detection after 1 week and that the previously reported longevity of odor memory may be an artifact of using forced-choice tasks. Visual memory, on the other hand, seems to rely on genuine internal representations of memory targets. The results for uncommon odors suggest that these are 'remembered' more by category than by individual properties. CONCLUSIONS: Odor memory is functionally different from visual memory, even when rehearsal is possible.

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FCOI Declarations: None.

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POSTER SESSION III
FRIDAY, 8 – 10:30 AM

The Scent of the Other Woman: Impact of Female Body Odor during Intrasexual Competition

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Consistent with Zajonc's affective primacy theory, sensory input requiring minimal cognitive processing may play a determinant role in human reproductive biology. Previously, it has been demonstrated that women exposed to chemosensory substances in body odors implement derogatory strategies towards potential rivals with specific reference to the processing of facial information. Here we aimed to examine if masked chemosensory cues produced by females may induce intrasexual competition strategies in a group of women in reproductive age. Fifty-four young women, not undergoing hormonal therapy, were exposed to body odors of other females masked with a neutral fragrance or to the fragrance alone while performing an emotional recognition task, consisting of a categorization of morphed faces as either angry or happy. A low and a high competitive state was provoked, in half the group respectively, via verbal instructions. Results indicate that women implement female intrasexual competitions strategies, specifically when exposed to the masked body odor. Females generally provided more intense qualitative ratings (e.g., attractiveness, dominance) towards female rather than male faces, suggesting an increment of attentional resources allocated towards female competitors. This was also confirmed in the higher accuracy during facial expression recognition of female than male faces. Increased attentional resources towards females were also matched with compatible cardiac responses, in the absence of perceptual differences among the chemosensory conditions. The present results provide tentative evidence that body odors are able to communicate aggression-related states and trigger female intrasexual competition strategies. Data are discussed in the context of social interactions among competitors.

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FCOI Declarations: None

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Olfactory Function in Patients with Hyposmia Compared to Healthy Subjects

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Individuals with hyposmia, or the partial loss of smell, represent a large sector of the population that is likely to grow with the current aging population; however, our understanding to how hyposmics centrally process odors is still not clear. Therefore, the aim of this study was to understand olfaction processing differences between patients with hyposmia and healthy controls using functional magnetic resonance imaging (fMRI). Eleven hyposmic and 12 healthy, normosmic subjects participated in the study. Olfactory functionality was assessed using tests for phenyl ethyl alcohol odor threshold, odor discrimination and odor identification. During fMRI sessions subjects were exposed to two different food-related odors (coffee and peach). The activations of the normosmic group were localized in typical olfactory areas (insula, orbitofrontal cortex [OFC], limbic system and amygdala). The hyposmic group showed similar regions of activation (insula, OFC, limbic system), however, less activation was found in the amygdala, left anterior cingulate and right OFC, but higher activation was shown in the right parahippocampal and both the left and right posterior cingulate gyrus which are assumed to play an important role in the processing and remembrance of memories. These results indicate similar central olfactory processing among groups, yet subjects with partial loss may attempt to compensate smell impairment with odor memory or higher motivation to smell.

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FCOI Declarations: None.

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#402

Functional Deterioration of Olfactory Cortex in Early Parkinson's Disease

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Olfactory dysfunction is a prodromal symptom of Parkinson's disease (PD) and prevalent in most PD patients with functional deficits in odor detection, identification, and discrimination. Previous studies have demonstrated functional deficits in the PD patients' olfactory cortex. However, it is not clear if the functional deficits in the olfactory cortex progress with the disease. In this study, we used olfactory fMRI to study the functional changes of the olfactory cortex in the early-stage of disease. Twenty-seven early- stage idiopathic PD patients (H&Y stage 1) participated in a longitudinal study of 4 visits in 2 years (baseline, 12 mo, 18 mo, and 24 mo). In addition, a group of 20 age- and gender-matched healthy subjects were studied as controls. All subjects gave written informed consent that approved by the institutional review board. The olfactory fMRI was conducted on a Siemens 3T scanner with BOLD- sensitive EPI. The odor stimulation paradigm consisted of twelve repetitions of odor-sniffing and odorless -air-sniffing. Lavender oil diluted in 1,2-propanediol at 0.10% (vol/vol) was used as the olfactory stimulant. The fMRI data at the individual level were processed with SPM8 (Wellcome Trust Center for Neuroimaging). Statistical parametric maps were generated at for odor-sniffing and odorless-air-sniffing. Odor-induced activation map was calculated by subtracting odorless-air-sniffing activation from odor- sniffing activation. Analysis of longitudinal fMRI data was conducted using Sandwich Estimator for Neuroimaging Longitudinal Data v0.1 (warwick.ac.uk/tenichols/SwE). Results show: 1) comparing to the healthy controls, odor-induced activation in the patients' olfactory cortex was significantly reduced; and 2) odor-induced activation in the PD patients was significantly reduced in the left primary olfactory cortex, left insular cortex, and left hippocampus during the two years of the disease. In conclusion, in the early-stage of PD, there is a significant functional deterioration in the olfactory cortex over time.

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FCOI Declarations: None.

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#403

A Longitudinal Study of Olfactory Function in Patients with Parkinson's Disease

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Olfactory dysfunction is a very common non-motor symptom in Parkinson's disease (PD), which occurs in more than 90% of the patients and does often precede the typical motor symptoms of bradykinesia, tremor and rigidity. The presence of olfactory deficits can be used in clinical routine for early diagnosis and to differentiate between PD and other diseases. However, it is unknown whether the olfactory deficit changes in relation to disease-related features when the disease progresses. In this study we aimed to evaluate changes in olfactory function in the course of PD. Olfactory function of 120 non-demented patients suffering from PD (79 male, 41 female, mean age 65.1 years, age range 40-83 years) was tested using the "Sniffin' Sticks" test battery. All patients had already been tested before. The period of time between the measurements varied from 4 months to 13 years.We found significantly lower olfactory scores ($p<0.001$) in the second olfactory evaluation. Furthermore, there was a significant decline in olfactory thresholds ($p=0.004$), discrimination ability ($p=0.031$) and odor identification ($p=0.005$). The initial olfactory function (normosmic, hyposmic, anosmic) significantly determined the follow-up performance. Normosmic patients presented with the most noticeable reduction whereas anosmic patients showed slight improvement over time. The decrease of quantitative olfactory function did not correlate significantly with clinical features, medication, sex, duration of illness or time span between testing. The presence of qualitative olfactory deficits (parosmia/phantosmia) did not influence the development of olfactory deficits. There is a significant decline of olfactory function in the course of the disease, which appears to be largely independent from the duration of PD and other disease-related features.

Funding Acknowledgements: TU Dresden.

FCOI Declarations: None.

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#404

Family-based Association Analysis Identifies Candidate Causal Genes for Congenital Anosmia

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Complete loss of smell, termed general anosmia, is a condition which affects over 6 million Americans, of which about 3% are congenital (termed congenital anosmia, CA). Although mutations in the *CNGA2*, a member of the olfactory receptor signal cascade, have been identified in one family with isolated CA, the genetic causes underlying this disorder are largely unknown. By examining how genetic variation associates with disease state in multiple families with segregating CA, we can begin to determine candidate causal genes for this disorder. To this end, we recruited families in which at least two members identified themselves as anosmic since birth. To date, we've collected pedigrees for 17 families from five different countries. For each family, at least two affected and two unaffected individuals completed a brief smell identification test to confirm anosmia or normosmia. We performed whole-exome sequencing for eight families, four of which appear to exhibit a recessive inheritance pattern for CA. Short sequence variants were identified in each subject and compared between affected and unaffected individuals. Family 1 features three living CAs from different generations, and CA appears to be inherited dominantly. In this family, roughly 45 genes have non-synonymous missense mutations in their protein-coding regions in anosmic but not normosmic family members. These genes encode transcription factors, extra-cellular matrix proteins, and intracellular signaling molecules, including ZHX3, a member of the zinc fingers and homeoboxes gene family. In two additional families for which we have completed sequence analysis, CA appears to exhibit an autosomal dominant mode of inheritance; however, the coding region of no single gene is mutated in every affected member of all three families, indicating different genetic causes for CA in these families. Further analysis will reveal additional dominantly- and recessively -inherited candidate causal mutations. As there are currently no viable treatments for anosmia, this work may contribute to the development of future diagnostics and therapeutics for this disorder.

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FCOI Declarations: None.

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#405

Olfactory Training and Cognition in Parkinson´s Disease

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Decrease of olfactory function in Parkinson's disease (PD) is a well-investigated fact whose impact on daily life is often underappreciated. Olfactory training however, has been shown to improve olfactory function in humans. The aim of this randomized, blinded, controlled investigation was whether patients with PD would benefit from "training" with high concentrations of odors in terms of an improvement of their general olfactory and cognitive function. Olfactory training was performed over a period of 12 weeks while patients exposed themselves twice daily to four odors (rose, eucalyptus, lemon, cloves). Investigations were performed at two visits with a detailed assessment of olfactory function and mood. Further, cognitive tests were performed. Sixty subjects trained with either high or low concentrations of four odors. After 12 weeks of training the smell score differed significantly between the 2 groups of PD patients ($p=0.003$) with the most pronounced changes in identification ability ($p=0.01$). The high concentration group also improved significantly in cognitive functioning as measured with the MOCA test ($p=0.03$) and in mood scores ($p<0.001$). Therefore, it might be hypothesized that the structured, short-term exposure to odors has positive effects on cognitive functioning and well-being in PD patients.

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FCOI Declarations: None.

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#406

Taste Function in Early Stage Treated and Untreated Parkinson's Disease

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Since brain stem regions associated with early Parkinson's disease (PD) pathology encroach upon those involved in taste function, the ability to taste may be compromised in PD. However, studies on this point have been contradictory. We administered well-validated whole-mouth and regional taste tests that incorporated multiple concentrations of sucrose, citric acid, caffeine, and sodium chloride to 29 early stage PD patients and 29 age-, sex-, and race-matched controls. Electrogustometry was also performed on the anterior tongue. The PD cohort was tested both on and off dopamine-related medications in counterbalanced test sessions. While whole mouth taste identification test scores for all stimuli were, on average, nominally lower for the PD patients than for the controls, a trend in the opposite direction was noted for the intensity ratings at the lower stimulus concentrations for all stimuli except caffeine. Moreover, regional testing found that PD subjects tended to rate the stimuli, relative to the controls, as more intense on the anterior tongue and less intense on the posterior tongue. No significant associations were evident between taste test scores and UPDRS scores, L-DOPA medication equivalency values, or [^{99m}Tc]TRODAT-1 SPECT imaging of dopamine transporter uptake within the striatum and associated regions. Our findings, albeit preliminary, suggest that suprathreshold measures of taste function are influenced by PD and that this disease deferentially influences taste function on anterior (CN VII) and posterior (CN IX) tongue regions. Conceivably PD-related damage to CN IX releases central inhibition on CN VII at the level of the brainstem, resulting in enhanced taste intensity on the anterior tongue.

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#407

TAS2R38 Genotype-Associated Alterations in Paranasal Sinus Microbiota

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Chronic rhinosinusitis (CRS) is an inflammatory disorder of the paranasal sinuses in which bacteria appear to play a role. The human bitter taste receptor, T2R38, initiates innate mucosal defense systems through detection of bacterial quorum sensing molecules. The genetics of T2R38 are well-described, and it has been hypothesized that a common polymorphism is associated with the inability to clear *Pseudomonas*. Clinical study has demonstrated an increase of the "nontaster" haplotype (AVI) in the CRS population, as well as an increase in culture positivity for *Pseudomonas*. The aim of the current study is to determine if TAS2R38 polymorphisms are associated with alterations in sinus microbiota. Ethmoid sinus samples from CRS patients undergoing surgery were analyzed by 16S rRNA gene sequencing. TAS2R38 genotype was assayed by RFLP. Phylum and genus-level comparisons were made using two-part statistic, ANOVA, and Fisher exact tests. 52 patients were included in the study (PAV/PAV = 10, PAV/AVI = 23, AVI/AVI = 19). Phylum-level comparison showed decreased abundance of Proteobacteria (0.04) and increased Bacteroidetes (p=0.03) in PAV/PAV subjects when compared to AVI/AVI (p=0.03 and 0.04, respectively). Genus-level analysis demonstrated increased abundance of *B.prevotella* (p=0.01) and *F.veillonellaceae* (p=0.04) in PAV/PAV patients. *Pseudomonas* was neither less prevalent nor less abundant in the PAV/PAV group. TAS2R38 polymorphisms are associated with microbiota alterations at the phylum and genus levels, however, *Pseudomonas* did not appear less prevalent or abundant in the PAV/PAV diplotype, suggesting there is a more complex interaction between bitter taste and microbial clearance.

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FCOI Declarations: None.

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#408

Taste Bud Density and Composition of Saliva in Patients Suffering from Taste Disorders Compared to Healthy Individuals

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Previous studies have shown a correlation between fungiform papillae density and acuity of taste. In addition changes of salivary composition in oral sensation complaints were observed as well. Aim of this prospective investigation was to determine correlations between the fungiform papillae density, salivary composition and taste acuity of healthy individuals and patients with idiopathic taste disorders. The present study was performed in 81 patients with a mean age of 58±13 years complaining of gustatory dysfunctions and groups of 20 younger (age 24±3 years) and 20 older healthy subjects (age 55±10 years). Taste perception was measured using taste strips and electrogustometry (ElGu). Fungiform papillae were quantified using the "Denver Papillae Protocol" and dichotomous key. Resting saliva was collected for 5 minutes. Salivary flow, antioxidant capacity, protein amount and carbonic anhydrase VI have been compared between patients and controls. Statistically significant correlations were found between fungiform papillae density, ElGu threshold and the total score of correctly identified taste strips. The average number of counted fungiform papillae in taste disorders (30.4±12.1) was lower than in age-matched controls (32±9.9), without showing statistically significance. Regarding qualitative taste disorders differences in taste acuity were shown. Patients with salty dysgeusia exhibited lower sensitivity (both, with ElGu and taste strips) compared to patients with bitter, metallic or sour dysgeusia.Characteristic changes of saliva were observed in total antioxidant capacity and protein amount. No differences between patients and controls were found for flow and carbonic anhydrase VI. Results indicate that there are changes in fungiform papillae density and saliva composition in idiopathic taste disorders. A follow-up study is underway to examine these changes regarding improvements of the disorders.

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FCOI Declarations: None.

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#409

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

AP1 Transcription Factors Regulate Taste cell Function and Renewal

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It is well established that with age, there is some loss of normal taste function which can have a significant impact on the feeding behavior of older populations. However, very little is known about the underlying mechanisms that link aging and taste function. While some factors have been identified as having a role in taste cell maintenance in adults, there are no comprehensive accounts of the gene expression changes that take place in the peripheral taste system during the aging process. In order to identity cellular factors required for proper functioning and maintenance of the adult taste system, we performed RNA-sequencing analysis on mouse CV/FOL taste cells at different ages and found significant aging-associated changes in gene expression. Interestingly, the Activator Protein-1 (AP1) transcription factors (c-Fos and Jun) were significantly downregulated with age. Since AP1 transcription factors regulate cell differentiation, proliferation and cell death, a reduction in their expression in taste cells with age strongly suggests that they have a role in normal taste cell function. Therefore we generated conditional Fos-knockout mice using Cre-K14-lox system to target the loss of Fos in K14 expressing cells, including differentiating taste cells. We analyzed the effect of Fos-KO on the structural and functional integrity of the taste buds. We found that Fos deletion leads to a significant reduction in the size of taste buds in taste papillae. Using markers for cell proliferation and apoptosis, we evaluated the differences in the taste cell properties in WT and Fos-KO mice. Our observations reveal a new role of AP1 transcription factors in regulating adult taste cell renewal and show that its downregulation leads to taste degeneration. Taken together, our data suggest that downregulation of AP1 transcription factors contributes to the age-associated decline in taste cell maintenance and function. Further studies are ongoing to understand how loss of Fos expression impacts taste cell function.

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#410

Using Cyclophoshamide to Illuminate the Role of Sox2 in the Adult Taste System

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Sox2 is a critical transcription factor required for the growth and development of the taste system, and while Sox2 is found in the adult tongue, it is unclear what specific role Sox2 has in the adult taste system. Sox2 expression in adult mice is affected by cyclophosphamide (CYP), a chemotherapeutic. CYP administration (75 mg/kg, IP), can cause behavioral deficits and cellular damage to the taste system. In this study, CYP was injected IP to determine how Sox2 expression is affected by injury and recovery of lingual epithelium. Animals were perfused with 4% paraformaldehyde each subsequent day, and tongues were extracted, frozen, and sectioned at 6 microns. Immunohistochemistry was used to stain for markers of cell type (PLCβ2; SNAP25) proliferation (Ki67), as well as Sox2. We are comparing the timing of Sox2 expression after CYP injection with markers of proliferation and cell type to determine potential roles for Sox2 in the recovery of taste system tissue as well as identify future experiments into the specific function of Sox2 in the adult tongue.

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FCOI Declarations: None.

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#411

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Long-term Exposure to Taste Stimuli Up-regulates PLCβ2 Expression in Mouse Taste Buds

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Taste buds are the dedicated sensory end organs of taste, comprising a complex and evolving profile of signaling elements. The transcription profile of taste buds has recently been noted to vary, for instance with diet-induced obesity, or satiation. To explore the importance of diet on taste development, we investigated the impact of long-term tastant exposure on taste receptor expression in mice. This was accomplished by exposing mice (n=3-4 for each treatment group) to five basic taste stimulants (0.05 mM quinine, 30.0 mM MSG, 90.0 mM NaCl, 2.0 mM saccharin, and 20.0 mM citric acid) through water supply over a four week period. Following this period, taste cells from the circumvallate papillae and surrounding non-taste tissue were isolated, with receptor expression in taste tissue quantified via qRT-PCR analysis. Data were compared to receptor expression in age-matched control mice (n = 5) exposed to unadulterated water over the same four week period. Results showed that treated mice, regardless of the tastant they were exposed to, expressed the critical taste signaling element Phospholipase-C beta 2 more highly than mice from the control group. This indicates a non-specific impact on taste transduction in treated mice. Interestingly, mice treated with the non-caloric sweetener saccharin displayed a decreased expression of the T1R1 receptor, usually associated with detection of umami taste. As T1R1 associates with T1R3, also a component of the sweet taste receptor, this may imply that the expression of sweet and umami receptors is less independent than is often assumed.

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FCOI Declarations: None.

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#412

Cessation of Hedgehog Pathway Blockade Leads to Restoration of Taste Responses Despite Incomplete Taste Organ Recovery

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LDE225 is a pharmacological Hedgehog (HH) pathway inhibitor used to treat cancers caused by deregulated HH signaling. Patients using LDE225 commonly report taste disturbances. We have shown that HH signaling is active in taste organs and that LDE225 treatment in mice disrupts taste organ morphology and taste sensation. To determine if and when there is recovery from adverse taste effects due to HH pathway inhibition, adult mice were treated with LDE225 for 16 days, and then the drug was discontinued for 7, 14, 21 days, and 3 and 5 months. Chorda tympani (CT) nerve recordings were made in response to lingual taste, tactile and cold temperature stimuli. Tongues were collected to assess: fungiform papilla (FP) morphology, expression of Sonic HH (SHH) ligand and taste bud (TB) cell marker (Keratin 8), individual TB cell types (NTPdase2, PLCβ2, SNAP25) and innervation (Neurofilament, P2X3). Three FP morphologies were quantified: (1) Typical FP/TB, (2) Atypical FP/TB and (3) Atypical FP/No TB. CT nerve responses to tactile and cold stimuli were retained throughout the experiment. In contrast, responses to taste stimuli were virtually eliminated after 16 days of LDE225 treatment but were fully restored by 14 days after drug discontinuation. However, LDE225-mediated alterations in FP/TB morphology did not completely recover. Strikingly, we observed normal taste nerve responses with only about 55% of Typical FP/TB and 40% Atypical FP/No TB from 14 days through 5 months after drug cessation. Innervation was retained in FP with or without TB. Expression of SHH was reduced by LDE225 treatment and increased after the drug was discontinued, reflecting changes in the number of TB cells. All TB cell types were decreased by LDE225 treatment and restored in Typical FP/TB during recovery. In summary, neural taste responses are fully regained after stopping treatment with a HH pathway inhibitor, but even after 5 months a full complement of TB is not restored, perhaps due to elimination of SHH-expressing TB progenitors in a subset of FP. The data indicate an essential requirement for HH signaling to maintain and re-establish taste organ integrity.

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#413

Injury-Induced Plasticity of Taste Nerve Terminal Fields in the Mouse NST Produced by an Initial Chorda Tympani Nerve Cut at Adulthood

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Injury-induced plasticity of central terminal fields occurs in multiple sensory systems when peripheral nerves are cut or damaged at adulthood. However, this plasticity induced at adulthood is not universal. Indeed, the occurrence of the effects appears to relate to the inherent plasticity of the specific sensory system. Moreover, an initial injury to a peripheral nerve may cause a supersensitive response when the nerve is injured later. We show here that a similar phenomenon occurs for the central gustatory system. The organization of the terminal fields of the chorda tympani (CT), greater superficial petrosal (GSP) and glossopharyngeal (IX) nerves, all of which project to the rostral portion of the nucleus of the solitary tract (NST), change in intact mice when experimental manipulations first occur at adulthood. Therefore, the terminal field organization of these nerves may also be especially plastic in response to nerve injury sustained at adulthood. In our first study, we show that cutting the CT (CTX) in adult mice results in a terminal field size decrease of 66% at 30 and at 60 days post CTX. Since our nerve labeling procedure required cutting the CT before applying an anterograde tracer to the same nerve at 30 or 60 days post CTX, this means that the CT was cut twice. For this procedure, the GSP and IX remained intact. In our second study, we did the same procedure as in the first; however, we also cut and labeled the GSP and IX when the second CTX was done. Surprisingly, we found that the CT terminal field size increased from what we found in the first experiment to be like that in control mice. The change in size occurred within 48 hrs. Thus, the CT terminal field nearly doubled in size when the GSP and IX were also cut with the second CTX. These results indicate that the initial CTX alters (i.e., “conditions”) the environment in the NST such that CT terminal field expands dramatically and rapidly. We are currently examining the effects of multiple CTX procedures at adulthood on the terminal field sizes of the GSP and IX, and potential competitive interactions among the three nerves.

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FCOI Declarations: None.

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#414

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Self-Renewal in Globose Basal Cell Cultures Reveals Bmi1 Activity in the Adult Olfactory Epithelium

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Olfactory epithelium (OE) is a self-renewing tissue, due to the presence of basal stem cells. Regulatory mechanisms involved in renewal and lineage commitment among the heterogeneous OE basal cell populations are incompletely understood. We hypothesized that successful culture expansion of purified basal cells may permit the identification of mechanisms important in the subset of basal cells competent for ongoing renewal. For basal cell purification, we utilized the recent finding that the c-Kit receptor is expressed by globose basal cells (GBCs). Here, cultures were established from immunopurified mouse c -Kit+ basal cells. Successful cultures were screened for expression of a panel of candidate regulatory transcripts. We found that GBCs capable of culture expansion are defined by expression of genes inhibiting neurogenesis, including *Hes1* as well as the *Id* gene family. In contrast, the proneural gene *Ascl1*, typical of amplifying neurosphere-forming cells, is down regulated in self-renewing GBC cultures. We also identified expression of the Polycomb group gene *Bmi1*, a key transcriptional repressor. The role of *Bmi1* as a necessary component for maintaining self-renewal properties in intestinal crypt and bone barrow stem cells has been described, but *Bmi1* activity in the OE has not been described. We therefore investigated *Bmi1* expression the OE in vivo. Inducible genetic fate mapping indicates that *Bmi1*+ basal cells function as adult OE stem cells following methimazole-induced lesion, with 44.7 ± 12.4 reporter-labeled cells per 0.5 mm of OE, 12 days following lesion and Cre activation (s.e.m., n=3 mice). Immunohistochemically, Bmi1 protein localizes to Sox2+ basal cells. Of interest, newly produced Gap43+ neurons do not express Bmi1, while the mature OMP+ population is Bmi1+, suggesting a distinct role in neurons following differentiation. In summary, we have identified transcriptional regulators that define self-renewing basal cells in vitro, and conclude that Bmi1+ basal cells are multipotent precursors involved in OE maintenance.

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#415

Mucin Isoform Expression in the Olfactory Epithelium of Mice and Humans

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Olfactory sensory neurons (OSNs) are the only neuronal cells directly exposed to the external environment within the nasal cavity. OSNs extend their axons into the central nervous system (CNS), thus, OSNs are a pathway for CNS infections. OSNs are located within a protective mucus layer that mediates transport of molecules, including odorants. A family of large glycoproteins, mucins, constitute a major protein component within the mucus layer. Mucins are known to mediate the mucus properties and play an important role in response to infections. Of the 21 known isoforms, several mucins have been identified in the sinonasal epithelium, including, mucins 1, 2, 5AC, and 5B. In this study, we utilized immunohistochemistry to observe whether there is differential expression of the mucin isoforms in respiratory versus olfactory epithelium in mice and humans. To observe nasal epithelium in mice, nasoturbinates were dissected from the nasal cavity of four 2-month BL/6 mice. For human studies, superior turbinates were collected during intranasal endoscopic surgeries (n = 4 patients). Membrane- teathered mucin 1 expression was observed in both olfactory and respiratory epithelium in mice, however, the pattern of expression was distinct between the two epithelium types. Intriguingly, in the mouse olfactory epithelium mucin 1 formed a layer approximately 2.4 micrometers below the OSN cilia. Similar to mice, mucin 1 was observed within the olfactory epithelium and respiratory epithelium in humans. In mice and humans, mucin 2 showed a similar staining pattern between olfactory epithelium and respiratory epithelium, and was expressed within secreted mucus and diffusely through the connective tissue of the lamina propria. Mucin 5B and mucin 5AC are secreted by submucosal glands in both olfactory and respiratory epithelium in mice. However, in human samples, mucin 5B and mucin 5AC are not expressed in the olfactory epithelium and appear specific to the respiratory epithelium. These data indicate that mucin isoforms have differential expression patterns in the olfactory versus respiratory epithelium and between mice and humans.

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FCOI Declarations: None.

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#416

Three-layer Regulation Leads to Monoallelic and Diverse Olfactory Receptor Expression

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Olfaction, or the sense of smell, can be essential for the proliferation and survival of an organism. In their Nobel-Prize winning studies, Axel, Buck and coworkers showed that each OR neuron stochastically expresses one and only one type of the ORs. Actually each cell only expresses one of the two alleles, which means two copies of a gene from the two parents, of an OR gene. The studies of Axel and Buck raise an intriguing question that has been puzzling the field since then: how can a cell activate the expression of one and only one allele of a single OR gene out of a large number of different types of ORs, and maintain its stable expression through the life of the cell, which is about 90 days in mice? Decades of studies have accumulated extensive information, but many observations seemingly make the problem even more complex for understanding. In this work we show how olfactory receptor neurons use simple physics and engineering design principles to achieve single allele activation, and several other functional requirements, such as maximizing OR expression diversity, at the same time. Specifically olfactory receptor activation is a multiple-objective optimization problem. Existing models focus only on monoallele activation, and cannot explain recent observations in mutants, especially the reduced global diversity of expressed ORs in G9a/GLP knockouts. Instead we integrated existing information on OR expression, and proposed an evolutionarily optimized three-layer regulation mechanism, which includes zonal segregation, epigenetic and enhancer competition coupled to a negative feedback loop. This model not only recapitulates monoallelic OR expression, but also elucidates how the olfactory system maximizes and maintains the diversity of OR expression. The model is validated by existing experimental results, and particularly underscores cooperativity and synergy as a general design principle for multi-objective optimization in biology.

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FCOI Declarations: None.

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#417

Refinement of Olfactory Receptor Transcribed Regions Improves the Accuracy of In Vivo Receptor Deorphanization

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The olfactory system is essential for survival because it enables animals to find food, detect predators and recognize fellow animals. Matching which odorants bind with which olfactory receptors (ORs) expressed in the olfactory sensory neurons is an essential step toward understanding odor detection and perception. We recently reported a method that enables a large-scale identification of ORs activated by a given odorant (deorphanization) in vivo using the phospho ribosomal protein S6 immunoprecipitation followed by RNA-Seq (Yue Jiang et al., 2015). As OR responses using this method relies on quantification of transcripts mapped on each OR, undefined transcribed regions of OR genes may cause inaccurate identification of ORs responding to the tested odorant. Here we show that an improved definition of the mouse OR transcribed regions allows us to identify more ORs activated by an odorant. We used available RNA-Seq data derived from the mouse olfactory epithelium to map OR transcripts to the genome. We then inspected the read distributions around each OR coding sequence and manually defined transcribed regions of 1111 ORs. Our data was in general agreement with the recently published results by the Logan lab (Ximena Ibarra-Soria et al., 2014), but transcript regions of 69 ORs were inconsistent. To evaluate the new OR transcript definition, we mapped our published RNA-Seq data from phospho ribosomal protein S6-associated mRNAs from olfactory epithelium of acetophenone- exposed mice on the new OR transcribed regions. We found that the number of ORs mapped on the ORs increased, which in turn increased the number of ORs significantly enriched in the acetophenone- exposed samples by more than two fold. In combination with in vitro confirmation, our new analysis resulted in dramatic increase in deorphanized ORs responding to acetophenone. In conclusion, our refined definitions of mouse OR transcripts improved the sensitivity of comprehensive gene expression analysis of ORs, allowing more accurate deorphanization of ORs.

Funding Acknowledgements: NIH R01 DC014423.

FCOI Declarations: None.

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#418

Structure-function of Odorant and Sweet-taste Receptors Based on Molecular Modeling

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No experimental structural data of chemoreceptor is available to date. Structure-function relationships can be obtained through atomic-level insights gained through molecular modeling. Here we have focused on mammalian Odorant (class A GPCRs) and Sweet-taste (class C GPCRs) Receptors. We critically aligned the sequences of receptors with those GPCRs for which a structure is available. Then, we have built the 3D structures of some representative receptors to identify the list of residues involved in their function, from ligand binding to G protein coupling. We have identified the network of conserved residues that drive their function. Above all, we show how the combination of sequence alignment, molecular modeling and the constraints of experimental data leads to identify rules that govern chemoreceptors function.

Funding Acknowledgements: NeurOlf project (ANR-NSF: Collaborative Research in Computational Neuroscience), Olfactome project (Région Provence Alpes Côte d'Azur,) Giract, Gen foundation Fondation Roudnitska.

FCOI Declarations: None.

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#419

Predicting Olfactory Perception from Chemical Structure using the DREAM Olfaction Challenge Dataset

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In vision and audition the mapping from wavelength to color and frequency to pitch are well understood. In contrast, the mapping from chemical structure to olfactory percept remains unclear. To address this problem, the DREAM Olfaction Prediction Challenge asked participants to build quantitative models that predict odor intensity, pleasantness, and 19 other perceptual descriptors from chemical structure alone. The perceptual data for the competition were collected from 49 untrained subjects who rated 476 different odors along 21 different scales, including intensity, pleasantness and 19 descriptor scales. Preceding this challenge, two published models (Khan et al., 2007; Kermen et al., 2011) predicted odor pleasantness from physicochemical features. These models successfully predicted odor pleasantness in this dataset ($r = 0.49$, $p < 0.001$; $r = 0.29$, $p < 0.001$). Using the extremely randomized trees algorithm, we built models predicting all 21 perceptual ratings using both physicochemical properties and metrics for structural similarity. Our models outperformed previous models for predicting pleasantness ($r = 0.65$, $p < 0.001$). We found that the model predicting odor intensity primarily used physicochemical properties related to hydrophobicity, polarizability, and molecular shape. In contrast, models predicting subjects' ratings of "bakery" and "fruit", relied heavily on structural similarity to prototypical molecule (such as vanillin for "bakery"). These findings indicate that for some perceptual features our machine learning algorithm successfully isolated particular physicochemical properties underlying the partitioning of odors between air phase and the receptor binding pocket, while for other features it instead utilized template matching.

Funding Acknowledgements: R01DC013339.

FCOI Declarations: Ajinomoto

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#420

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Sweet Modifications by Miracle Fruit and Gymnema sylvestre Affect Flavor

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Taste and retronasal olfaction (volatiles perceived from the mouth) interact in the brain to produce flavor. Recent work from our lab has focused on fruit volatiles that enhance sweetness. The present study uses two substances that alter sweetness (*Gymnema sylvestre* and miracle fruit) to examine their effect on the perception of volatiles in sweet and sour foods. Subjects used the Global Sensory Intensity Scale (GSIS) to assess the sweetness, sourness, aroma, and flavor of 10 foods varying in sweetness and sourness. The contributions of taste to the perception of flavor were examined with multiple regression. Specific hypotheses were tested with ANOVA, correlation, and regression. Our results support previous research on sweetness modifiers: miracle fruit added sweetness to acids, which in turn reduced sourness via mixture suppression; *Gymnema sylvestre* reduced sweetness in sweet foods as well as the sweetness induced by miracle fruit. Statistically significant new results were as follows. Multiple regression showed that both sweet and sour contribute to flavor. In predominantly sweet foods (chocolate, maple syrup), *Gymnema sylvestre* reduced sweetness and flavor. In sweet plus sour foods (strawberries, tomatoes), miracle fruit enhanced sweetness and flavor, and *Gymnema sylvestre* reduced them. The effects of miracle fruit on predominantly sour foods was complicated by the dual action of miracle fruit: intensification of sweet and reduction of sour. These foods (vinegar, lemon, mustard, pickle) were sweetened by miracle fruit, but the flavor enhancement associated with added sweetness appears to be countered by the flavor reduction produced by reduced sourness.

Funding Acknowledgements: NIDCD R21DC013751.

FCOI Declarations: None.

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#421

Detection of Glucose Oligomers by Humans is Not through the Human Sweet Taste Receptor

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Previous studies in our lab showed that humans could taste glucose oligomers, but not polymers. The main goal of this study was to determine whether glucose oligomers can be detected through the sweet taste receptor. During pilot study, we learned that lactisole, a sweet inhibitor, causes 'sweet water taste' during rinsing, but that cold temperature eliminates this effect. The first study was thus conducted to determine whether stimulus temperature affects the detection of glucose oligomers. Ss were asked to discriminate 75 mM reducing ends (RE) glucose oligomers (S1: ~75% DP3-8, ~25% DP9+; S2: ~25% DP3-8, ~75% DP9+) and polymer (S3: 100% DP9+) from blanks at room (~22 °C) and cold (~10°C) temperatures. Results showed that Ss can discriminate glucose oligomers but not polymer at both temperatures. In the second study, Ss were then asked to discriminate sugars (75 mM glucose and maltose; 0.025 mM sucralose) and glucose oligomers (75 mM RE S1 and S2) from blanks in the absence and presence of 300 ppm lactisole at ~10°C. All stimuli and blanks contained 5 mM acarbose to prevent hydrolysis of glucose oligomers. Results showed that Ss can discriminate sugars in the absence of lactisole only, while they can discriminate glucose oligomers in both conditions. To confirm these results, we performed CA^{2+} signaling assay in hT1R2/hT1R3-expressing cells glucose oligomers (G3-G8) and sweet tastants (sucrose, cyclamate, Ace-K). Results showed that hT1R2/hT1R3 was activated by sweet tastants but not by glucose oligomers. In addition, the glucose oligomers did not influence on sweet ligands-induced Ca^{2+} influx. Overall, these findings imply that there is an independent pathway or receptor(s) that facilitates the detection of glucose oligomers in humans.

Funding Acknowledgements: This study was supported by OSU Agricultural Research Foundation to Lim and the Korea Food Research Institute (E0111201-05) to Rhyu.

FCOI Declarations: None.

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#422

Interactive Effects of PTC Sensitivity, Papillae Density, Food Neophobia and Food Choice

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Past research has shown that a) the number of papillae an individual has influences their food choice, b) PTC sensitivity is related to both food choice and the number of papillae, and c) the degree of food neophobia (the unwillingness to try new foods) influences diet. The present study assesses the interactive effects of all of these influences. Participants (n=69) completed questionnaires related to their eating attitudes and behaviors. PTC sensitivity ratings were made, tongue papillae were counted, and participants chose a typical meal from among 70 foods in a mock cafeteria setting. A significant positive correlation between papillae count and PTC bitterness rating was found, $r=.65$, $p<.001$. Food neophobics were found to have a greater number of papillae than food neophilics or an average group, $F(2,69)= 4.16$, $p=.02$. Food neophilics chose a larger meal weight (M=671.37 grams) than food neophobics (M=432.97 grams), $F(2,69)= 3.36$, $p=.04$. Food neophobics chose a meal comprised of significantly fewer calories (M=433.34 calories) than the average group (M=666.05 calories), $F(2,69)=3.43$, $p=.04$. Food neophobics chose a meal comprised of significantly less protein (M=24.44 grams) than the average group (M=24.44 grams), $F(2,69)= 3.66$, $p=.03$. Future research should examine such interactive effects on participant health status.

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FCOI Declarations: None.

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#423

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Sensory-specific Satiety Modulates Neural Representations of Expected Food Odor Rewards in Humans

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In order to make adaptive food choices, animals must generate internal representations of expected outcomes that are sensitive to fluctuations in satiety state, which can drastically modulate the value of food items and their associated aromas. While the orbitofrontal cortex (OFC) has been identified as a key neural substrate for updating expected food value after satiety in non-human model systems, the specific neural mechanisms by which signals in this region inform choices in the human brain remain unknown. Here we conducted an experiment in which human subjects (N=17) learned to associate visual symbols with two distinct appetizing food odors. Subjects then performed a task in which they made choices among these symbols in order to receive low-intensity (i.e. low value) or high-intensity (i.e. high value) versions of the odors, while undergoing fMRI. Scanning was conducted first while participants were hungry, and then immediately after they had eaten food corresponding to one of the food odors to satiety. In the hungry state, subjects reliably chose symbols associated with high-intensity odors. However, after eating, subjects preferred low-intensity versions of the sated food odor, while the non- sated food odor choices were unchanged. Analysis of the imaging data revealed that this behavioral switch coincided with modulation of odor identity-specific representations in posterior OFC at the time the choice offer was presented, and odor identity-general representations in ventromedial prefrontal cortex (vmPFC) at the time the decision was made. Taken together, these preliminary findings provide evidence for a neural mechanism in which expected value signals in distinct substrates are updated at specific times in a decision process to redirect choices towards non-sated foods.

Funding Acknowledgements: T32NS047987-09 Training Program in the Neuroscience of Human Cognition.

FCOI Declarations: None.

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#424

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Modulation of the Sweet Water Taste by Sweeteners

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Pre-exposure to certain antagonists (e.g. lactisole) of the hT1R2-hT1R3 sweet taste receptor cause water to taste sweet, and Galindo-Cuspinera et al (2006) have proposed a receptor based mechanism to explain this long-puzzling phenomenon. After we observed in preliminary testing for a different study that mixing sucrose with lactisole reduced the intensity of the Sweet Water Taste (SWT), we conducted an experiment to quantify this effect and to learn whether sucrose also suppresses the SWT when it is presented before or after lactisole. 22 subjects used the gLMS to rate the sweetness intensity of multiple dH2O rinses (1) after lactisole; (2) after a lactisole-sucrose mixture; (3) when sucrose was presented before lactisole; (4) when sucrose was presented after lactisole; and (5) when sucrose was presented after a dH2O rinse had followed lactisole (i.e. when the SWT was established). The results confirmed that sucrose in mixture with lactisole can partially suppress the intensity (and duration) of the SWT, and further showed that when presented after lactisole, sucrose completely suppressed the SWT. The latter surprising result was followed up in a second experiment with 20 subjects which showed that in mixture with lactisole, cyclamate and fructose also partially suppress the SWT, whereas saccharin, sucralose, and neohesperidin dihydrochalcon initially enhance it before partially suppressing it. The same experiment also showed that presenting fructose and cyclamate after lactisole completely suppressed the SWT, whereas the other 3 sweeteners only partially suppressed it. These results imply that dissociation of lactisole has an excitatory effect on hT1R2-hT1R3 that can be differentially modulated by receptor agonists, most likely via positive and negative allosteric interactions.

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FCOI Declarations: None.

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#425

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Female Rats Do Not Discriminate Sucrose from Water as Accurately as Male Rats Despite Showing Similar Asymptotic Performance and Detection Thresholds

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Understanding sex differences in behaviors guided by taste may help achieve more female sex-inclusive health information; however, differences in taste detection between the sexes have not been fully established. To this end, we compared the sucrose taste detection profiles of 6 pairs of male and female Sprague-Dawley rat littermates. Animals were trained to perform a 2-response operant taste detection task in computer-controlled gustometers. In this task, water-restricted rats are trained to associate one response spout with sampling water and the other response spout with sucrose (0.3 M); licking the correct response spout following sample exposure produces a water reinforcer. Once the performance of the rats was high and stable, rats were tested with sucrose concentrations that were systematically decreased (0.3-0.003M) across sessions until their behavior fell to chance accuracy. At the conclusion of testing, some midrange concentrations were retested (0.08 M; average used in analysis) and added (0.11 & 0.057 M). The overall percentage of accurate responses by the rats across sucrose concentrations was analyzed via 2-way repeated measures ANOVA. Curves were fit to the data using a 3-parameter psychometric function to determine asymptotic performance, slope, and taste detection threshold (concentration at half-maximal performance). Overall differences in performance across the sucrose concentrations tested between the sexes were revealed, but detection thresholds and other curve parameters did not differ. This suggests that male and female rats may differ in their ability to discriminate water from sucrose, particularly at midrange concentrations, but that any difference disappears at asymptotic and perithreshold concentrations. Future studies are needed to assess the perceptual and physiological mechanism underlying these differences. While a lack of differences in at least behavioral detection thresholds between the sexes may encourage researchers to use both male and female animals in their future studies, sex differences exist that require additional consideration and assessment.

Funding Acknowledgements: Baldwin Wallace Summer Scholars Program.

FCOI Declarations: None.

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POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Individual Differences in Psychophysical Functions for Glucose, Sucralose, and Rebaudioside A

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Many non-nutritive sweeteners have sweetness thresholds far below those of glucose and sucrose. However, common non-nutritive sweeteners actually have lower maximum sweetness, perhaps due to inhibition of sweetness by increasing bitter side tastes (Antenucci and Hayes, 2015). The purpose of the current study was to determine if suppression of sweetness was apparent with increasing bitterness in psychophysical functions for individual subjects. Healthy adults rated the sweetness and bitterness of 8 concentrations of glucose (0-2500 mM), sucralose (0-187.5 mM), and rebaudioside A (reb A; 0-4 mM) in aqueous solution. Concentrations were selected to achieve a wide range of perceived intensities (reb A was limited due to solubility). Subjects rated intensity using the general Labeled Magnitude Scale. Tests were repeated both within sessions and on separate days to assess stability. The average (group) sweetness function for glucose had no clear asymptote, but sweetness did stop increasing at high concentrations for the non-nutritive sweeteners. At the individual level, no subject rated glucose as bitter, and sweetness vs. concentration functions were monotonic. In contrast, for sucralose sweetness first increased, then decreased for most subjects. The concentrations at which individual subjects began to report bitterness and the concentrations at which sweetness began to decrease corresponded closely. A similar pattern was seen for reb A, though the association between bitterness and suppression of sweetness was perhaps less clear. Interestingly, one subject who never reported bitterness for either non -nutritive sweetener also produced monotonically increasing sweetness functions for all stimuli. Accordingly, individual subject data were consistent with the hypothesis that increasing bitterness plays a role in limiting the maximum sweetness of non-nutritive sweeteners. Further, though the group function for sucralose closely resembled published functions (Schiffman and Gatlin, 1993), the group function was not representative of any individual.

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FCOI Declarations: None.

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#427

A New Paradigm for Human Taste Measurement

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We previously developed a high throughput system for in vivo measurement of taste quality and palatability using rats as subjects (Palmer et al, 2013, PLoS ONE). We now have extended this technology to creation of a new paradigm for taste measurement in humans. The system is fully automated and is comprised of a robotic sample delivery system, a touch-sensitive display (TSD) that records the subjects' responses, and a command computer that runs subject-interactive algorithms and communicates with a database. The algorithms that operate the taste testing method are "gamified" - structured according to game play and game mechanics. Subjects are trained how to use the system through the interactive algorithms and are given minimal instruction. The samples (200-500 ml each) are drawn from a single randomly-selected well of a 96-well plate by an automated pipette clipped to a robotic gantry. The gantry moves the pipette to the subject who then removes the pipette and self-administers the solution. A set of taste-stimulus standards are mapped to specific locations in the visual field of the TSD, and subjects are trained by the algorithms to touch a location on the TSD for a reward after tasting the sample. The rewards are virtual poker chips with point values that are directly remunerated for actual currency at the end of a session. The algorithms are designed to incorporate principles of operant conditioning and signal detection theory and are structured to provide consequences to the subject's response on each trial. Thus taste-testing performance and rate of testing are incentivized. Subjects typically complete a 96-trial session in 45-60 minutes with high performance accuracy and test-to-test reproducibility. The software is designed to accommodate flexibility in experimental design so that a large array of test protocols can be operated under the interactive algorithms.

Funding Acknowledgements: Internal funds from Opertech Bio, Inc.

FCOI Declarations: Kyle Palmer is Co-founder, CSO, and part owner of Opertech Bio, Inc. The abstract describes methodology developed at Opertech Bio that can be used for commercial purposes.

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#428

Simple and Affordable Microcontroller-based Platforms for Quantifying Odor and Taste Preferences in Rodents

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We developed independent platforms for the quantitative measurement of odor and taste preferences in mice. The taste preference platform is based around the well established two-bottle flavor preference task (www.monell.org/MMTPP) developed by Bachmanov & Tordoff and using a contact lickometer (Slotnick, J Exp Anal Behav, 91:253-55) to detect individual lick events. The odor preference apparatus consists of a six-walled behavioral arena with 3D-printed nose poke ports on five of the walls which house plastic odor cups and infrared beams to detect nose-pokes. For both platforms, we developed custom code written for the affordable Arduino line of microcontrollers to monitor the animal's behavior (either licking or nose poking) and store the data onto removable SD (secure digital) cards for later transfer onto computers for analysis. This design allows for multiple platforms to run and acquire data from an unlimited number of animals simultaneously - independent of a computer. Both platforms are affordable and easy to assemble. Experiments using these platforms illustrate the reliability of the lick-detection platform to detect the licking behavior of c57bl6 mice to a variety of tastants and to use the data to quantify the micro-structure of licking. Additional experiments also illustrate the ability to use the olfactory platform to quantify odor investigation behavior in c57bl6 mice. Together, these results demonstrate the development and validation of these microcontroller-based platforms for use in rodents. We intend to share the acquisition code, custom-designed 3D-printed part source files, parts list, and assembly instructions to interested users. Given their ease of assembly and use, as well as low-cost, we believe these platforms will make excellent additions to behavioral neuroscience labs interested in the chemical senses and even lab-based classes to provide students with opportunities to learn simple olfactory and gustatory behavioral testing.

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FCOI Declarations: None.

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POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Consistency of Odor Perception: Comparison by the Number of Components of the Odor Mixtures

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The impression is different every time we smell the odor. It should be considered that unstable perception evoked by various factors such as how to sniff, surrounding environment, and health condition. It might be suggested that olfactory sensory coding hardly match with memory of meaning than other senses. To examine the change of odor impression at the perception level, we simply repeated similarity rating of odor pairs for two times and compared the results. Apple(A), grapefruit(G), peach(P) consisting of 10 components, “binary” mixtures(AG, AP, GP), and “trinary “ mixture(AGP) were used. Thirty participants rated similarity for twenty-eight odor pairs using VAS (0: not similar, 100: similar). The second rating conducted within three days. As results, however correlations between 1st and 2nd similarity rating of individuals were low, the correlation between them of means among participants was significantly high. More precisely, we examined factors which might have affected the low correlation of individuals, and counted the number of ratings which deviated from mean among participants by more than 1SD. However, there were no tendencies in rating times (1st, 2nd), odor pairs and certain individuals. Thus, participants seemed to rate odors precisely, however at certain times rated odors differently than before. Furthermore, we conducted cluster analysis on ratings of same odor pairs, and two groups were made. As a result of MDS, a group which rated same odor pairs similarly (n = 14) showed ratings reflecting the characteristic of each odor in the “binary” mixtures more precisely in 2nd rating, and showed significantly high OAS score than the other group (n = 16). It suggested that greater tendency to pay attention to odors in the environment related to good ability for discrimination of the components in the odor mixture, and consistency of similarity rating might be higher if the contact period setting is more longer.

Funding Acknowledgements: University of Tsukuba.

FCOI Declarations: None.

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POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

What is the Functional Significance of Odor Response Maps in the Olfactory Epithelium? A Critical Test of the Sorption Hypothesis in the Mouse

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Sensory epithelia contain arrays of receptor cells that transform specific types of stimuli into neural signals that form an organism's 'knowledge' of their environment. The spatial distributions of receptors on these surfaces (e.g. retina and skin) typically possess nearest-neighbor relationships and are often dramatically non-uniform to gain efficiency in information capture and neural processing. By contrast, odors, unlike visual and tactile stimuli, have no obvious spatial dimension. What need then could there be for either nearest-neighbor relationships or anisotropic distributions of receptor cells in the olfactory epithelium (OE)? More than a half century ago, Adrian provided the only widely debated answer to this question when he posited that the physical properties of odors, such as volatility and water solubility, determine a spatial pattern of stimulation across the OE that could aid in odor discrimination, a still controversial and actively investigated idea often referred to as the 'sorption hypothesis.' Here we report recent results in our ongoing effort to test this hypothesis by mapping mouse OE responses using the electroolfactogram (EOG) in an advantageous open preparation with fresh postmortem tissue. This effort employed 23 different odorants and includes data from 91 subject and 32 recording loci across all regions of the OE resulting in over a 1,000 EOG traces. We have correlated these results with our computational fluid dynamics (CFD) simulations of airflow velocities and odorant sorption patterns in the mouse nasal cavity. Consistent with previous studies, we found pronounced spatial heterogeneity of EOG responses across the OE. However, we *did not* find evidence of sharp zonal boundaries (map fractures) and we found scant support for the sorption hypothesis, despite the inclusion of extremely hydrophobic and hydrophilic odors in our stimulus set. Theoretical objections to the sorption hypothesis and alternative explanations for the map of odor responses in the OE will be explored.

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FCOI Declarations: None.

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#431

Smelling is Telling: Human Olfactory Cues Influence Interpersonal Judgments

Jessica M. Gaby, Vivian Zayas
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How does a person's smell affect others' impressions of them? Studies employing axillary odor on t-shirts or pads have found that natural body odor (no deodorant or perfume) conveys cues related to emotion, genetics, health, and mate fitness. In real life, however, people encounter very different olfactory information: perfume, deodorant, hygiene products and dietary choices (typically eliminated in body odor studies) modify natural body odor to create "diplomatic odor." Here, we aim to establish that body odor influences social judgments in realistic interactions, to examine the social differences between natural and diplomatic odor, and to compare our method to traditional t-shirt collection methodology. We first examined whether olfactory cues reliably affect interpersonal judgments in real life interactions. Blindfolded, earplugged raters smelled and made judgments about the diplomatic odor of live odor donors in a repeated measures design. We found that raters' social judgments based on these odors were highly consistent across time ($p < .01$). We then examined 1) whether liking of natural odor predicts liking of diplomatic odor; and 2) whether real-life olfactory judgments relate to t-shirt based judgments. In a similar design, we asked raters to judge the odors of both live donors and their t-shirts, smelling natural odor (following a 2-day washout) and diplomatic odor on two separate weeks. As in study 1, raters' repeated judgments based on live odor in both natural and diplomatic conditions were highly consistent across time ($p< .05$). Interestingly, 1) judgments based on natural odor did not consistently predict judgments based on diplomatic odor. Finally, 2) live judgments within odor condition predicted t-shirt based judgments for approximately half of our rating questions ($p<.05$). Our results show that humans reliably use olfactory information to inform their social judgments of others, but suggest that natural and diplomatic odor convey different social information. Future studies should account for personal odor influences in order to best represent realistic social interactions.

Funding Acknowledgements: Cornell Institute of Social Sciences.

FCOI Declarations: None.

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Stranger's Body Odor Selectively Reduces Response Time to Threatening Faces

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Threat detection capitalizes on sensory mechanisms enjoying prioritized processing. Besides visual signals, recent evidence suggests that chemical signals may also be used in threat detection. Indeed, smelling the body odors of strangers, a potential source of threat throughout human history, activates the cerebral network processing threatening stimuli. However, whether and how the body odor of strangers modulate behavior in line with threatening stimuli is still unclear. Here, we tested the effect of strangers' body odor in modulating motor responses to faces with different threat levels. To minimize purely perceptual effects (e.g., odor pleasantness) body odors were masked with a neutral common odor, which alone served as a control condition. Results indicate that participants reacted significantly faster to highly threatening faces only in the presence of the mixture containing the body odor. Taken together, our findings demonstrate that, like other animals, humans are able to extract chemical information about the presence of unknown individuals and their threat-potential as well as accordingly modulate behavioral processing. Importantly, these effects occur outside of conscious awareness.

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FCOI Declarations: None.

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#433

Increased Sensitivity to L-Felinine in Mice Correlated with Elevated Fos-Immunoreactivity in the Accessory Olfactory Bulb

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Unique for Felidae family sulfur-containing amino acid felinine and its volatile derivatives may be used by the house mouse to recognize potential predators, their physiological status and may affect reproductive output in mice (Voznessenskaya, 2014). Our earlier studies showed that regardless of the odorant, early exposure to it resulted in an increase in a rodent's sensitivity to that stimulus; exposure to the odorant during two weeks after the pups opened their eyes appeared to induce the greatest level of sensitization, suggesting a sensitive period to such stimuli (Voznessenskaya et al., 1995; 1999). Specific aim of our study was to examine whether early olfactory experience of mice with cat chemosignals may affect sensitivity to target odors later in adulthood and whether these changes in sensitivity correlated with neural activation in olfactory bulbs. We measured olfactory thresholds (OT) to cat urine/ L-felinine using an 8-channeled olfactometer (Knosys, USA). Exposures of mice to cat odor (urine or L-felinine) during two weeks after eyes open, significantly lowered the OTs to cat urine ($n=10$, $p<0.05$) as well as to L-felinine ($n=10$, $p< 0.01$) relative to controls. We performed immunohistochemical studies to identify neural substrate involved in reception and analysis of L-felinine. Mice were exposed intermittently (50% duty cycle each minute) to 0.05% L-felinine ($n=8$) or clean air ($n= 8$) for 45 minutes prior to perfusion. We sectioned olfactory bulbs at 30 μ and stained (c-Fos (4) sc-52, Santa Cruz Biotechnology; Alexa Fluor ®594, Life technologies). Sections were analyzed using All in-One Fluorescence Microscope Keyence Bz -9000 (Keyence, Japan) with software. We recorded specific pattern of activation in accessory olfactory bulb (AOB). Neonatal exposures to L-felinine (0.05%) caused significant increase in number of Fos-positive cells in AOB in response to stimulation with L-felinine ($n=8$, $p < 0.01$) as well we recorded an increase of activated area ($n=8$, $p<0.001$). Sensitization to L-felinine in mice correlated with elevated Fos-immunoreactivity in AOB in response to stimulation with the compound.

Funding Acknowledgements: Russian Foundation for Basic Research 14-04-01150 to VVV.

FCOI Declarations: None.

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#434

Modulation of Medial Amygdala Circuits involved in Chemosignal Processing

Don Tucker Award Finalist

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Florida State University, Program in Neuroscience and Dept. Biological Science, Tallahassee, FL, United States

Many species use chemosignals to convey social information. Chemosignals are detected by the vomeronasal organ and main olfactory epithelium, which project to the medial amygdala (Me) via the accessory olfactory and main olfactory bulb respectively. Within Me, immediate early gene responses have previously shown significant differences in responses of distinct cell populations to chemosensory signals, including conspecific and heterospecific stimuli. Me projects to downstream hypothalamic targets which produce appropriate social behaviors. Several GABAergic intercalated (paracapsular) nuclei in other areas of the amygdala have been implicated in the circuitry involved in fear conditioning and extinction. These paracapsular cells modulate activity in the adjacent basolateral and central amygdala in order to produce appropriate fear responses. The main intercalated nucleus cell-group (mICN) is adjacent to posterior Me (MeP) and appears to be involved in chemosignal processing. Previous immediate early gene studies have shown a negative correlation between mICN and MeP responses to various odors and electrophysiological studies in hamster horizontal and coronal slices now show functional inhibitory connections from mICN to MeP. In the fear conditioning circuit, activity of the paracapsular ICN neurons can be modulated by dopamine (DA) and by infralimbic (IL) cortex input, in different phases of conditioning and extinction. Using whole cell patch clamp electrophysiology, we find DA and DA receptor specific agonists have a hyperpolarizing effect on mICN neurons and reversibly reduce the inhibitory effect of mICN extracellular stimulation on MeP principal neurons. Further, *in vivo* tract tracing from IL showed fibers projecting from IL to mICN, suggesting a potential role for IL in the Me/mICN circuit involved in chemosignal processing.

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FCOI Declarations: None.

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#435

Effects of Congruent vs. Incongruent Product Scent Administration on Online Purchasing Behavior

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The present study investigated whether the administration of a congruent vs. incongruent product scent during an online shopping session influenced participants' product ratings and purchasing behavior. Participants (n=80) rated 10 on-line products. Three of these products constituted the experimental conditions (leather jacket, fruity cereal, fresh brewed coffee) for which the congruent scent accompanied the online shopping experience via a Aroma-Crystal ultrasonic piezo crystal essential oil diffuser attached to the USB port of the computer. Participants were asked to rate a) the quality of the product and b) the maximum amount they would be willing to pay for the product. Independent t-tests were performed between the item ratings in the control condition (no scent) vs. the item ratings when a congruent product scent was administered. Consistent trends were noted such that congruent scent administration increased quality ratings and the amount participants were willing to pay for these products (such as coffee product with coffee scent). Further, if the product was related to the scent being administered (such as coffee with breakfast foods or sneakers with leather), the participants also rated those products as having a higher quality and cost. Given the continuing increase in online shopping, the administration of congruent product scents could further bolster ratings of product quality and revenue. Future research should examine actual purchasing behaviors in-store vs. online during congruent scent administration.

Funding Acknowledgements: Funding was provided by the Wheeling Jesuit University Department of Psychology.

FCOI Declarations: None.

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#436

The Association of Weight Change in College Freshmen with Taste Change

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Human and animal studies report blunted taste sensitivity in the overweight and obese, with heightened sensitivity reported following weight loss, but whether a modest increase in weight can induce taste changes is unknown. We hypothesized that a moderate increase in weight experienced in the freshman year of college would result in decreased ratings of taste intensity perception. A longitudinal study assessed within-person suprathreshold taste intensity changes in 89 male and female college freshmen, given that 75% of college students experience weight gain. Appetitive (sweet, salty, and umami) taste intensity was measured upon college entry, and after 3 and 9 months, using the general Labeled Magnitude Scale. Relative taste change was computed by taking the difference in log taste intensity ratings across two time periods (baseline to 3 months, baseline to 9 months). Ordinary least square regression models assessed the association of weight change with the concentration-dependent taste function change, adjusting for confounding dietary factors, gender, and ethnicity. Participants gained an average of 1.8 kg (4.0 lbs) in the first 3 months and 2.3 kg (5.1 lbs) over the 9-month academic year; weight change ranged within the entire sample from -4.0 to +9 kg. Focusing on sweet taste, which was assessed at 3 concentrations and by the area-under-the-curve (AUC) to integrate across concentrations, sweet taste AUC increased by 7% over the 9-month period. Models assessing the association of weight change with sweet taste change found statistically significant differences in this association by gender ($p=0.043$). There was little to no association in females, but nine-month weight gain in males associated with decreasing sweet taste function such that a 1 kg increase in weight was associated with a 16% decrease in sweet taste AUC ($p=0.016$). This trend was evident in both the concentration-dependent and overall sweet sensitivity models. These results suggest that moderate weight gain may associate with taste loss, and that males may be more susceptible to sweet taste loss than females.

Funding Acknowledgements: Rose Marie Pangborn Sensory Sciences Scholarship Fund.

FCOI Declarations: None.

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#437

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Effects of Gastric Bypass Surgery on Oral Fat Preference and Lingual Fatty Acid Receptor Expression in High Fat Diet-Induced Obese Female Rats

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Roux-en-Y gastric bypass (RYGB) surgery has been used successfully as a treatment for obesity. Following RYGB, preference for dietary fat and sweet tastes is reduced. Animal research on RYGB- elicited effects on taste has been limited to male rats despite the fact that >85% of RYGB patients are women. Furthermore, lingual mechanisms of altered dietary fat detection have not been investigated. Thus, the current study assessed the effects of RYGB on 1) lick responses to sweet or fat oral stimuli in high fat diet (HFD)-induced obese female rats, and 2) CD36 and GPR40/FFAR1 expression in the circumvallate papillae (CP). All rats undergoing RYGB or Sham surgery were fed a HFD (60%kcal from fat) for 14 weeks prior to surgery. A control group fed a standard chow diet was included. Lick-responses to water and various concentrations of sucrose solutions or Intralipid (containing primarily long chain fatty acids) were assessed in brief-access (10 s) tests using Davis rigs. For the molecular study, separate cohorts of RYGB or Sham-operated rats were sacrificed following an overnight fast. CD36 and GPR40/FFAR1 mRNA levels were assessed by RT-PCR. RYGB reduced body weight by 20-25%, alike in male rats and human subjects, and the weight loss was maintained throughout the study (for 3 months). Similar to previously published data in male rats, female RYGB HFD rats displayed significantly fewer lick responses ($p<0.05$ vs. Sham) to higher concentrations of Intralipid (1-10%) and sucrose (0.6 -1.5 M) solutions, but unchanged responses for the lower concentrations. CD36 mRNA levels in the CP were increased by the consumption of the HFD in the Sham-operated rats compared to the control rats. RYGB attenuated the HFD-induced increase in CD36 mRNA expression. In contrast, RYGB surgery increased lingual GPR40 expression, which was not affected by HFD alone. Thus, changes in lingual CD36 expression in female rats coincide with changes in body weight and taste responses to oral fat following consumption of a HFD and RYGB surgery, suggesting that lingual CD36 contributes to RYGB-induced changes in dietary fat preference, and in turn, to sustained weight loss.

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FCOI Declarations: None.

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#438

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Taste Responses in the Nucleus of Solitary Tract of Freely Licking Rats with Roux-en-Y Gastric Bypass Surgery

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Roux-en-Y gastric bypass surgery (RYGB), the most effective treatment for morbid obesity, produces changes in food preference. In particular, studies show that there is a shift in preference toward less calorie-dense foods after surgery. While several mechanisms have been proposed, the underlying brain and behavioral changes remain unclear. Here, we recorded responses from the nucleus of the solitary tract (NTS), the first neural relay of the gustatory system, in rats that underwent RYGB surgery. Rats were implanted with an 8-wire bundle electrode in the NTS. Following recovery, rats were then mildly water deprived and placed in an experimental chamber containing a lick spout for fluid delivery. Gustatory stimuli consisted of prototypical tastants: sucrose (0.1 M, 0.05 M, 0.025 M), NaCl (0.1 M, 0.05 M), 0.1 M MSG, as well as naturalistic stimuli: grape juice (100%, 25%, 12%), 75% clam juice (0.12 M NaCl), 25% cream. Artificial saliva (AS) was used as a rinse and also to dilute the tastants. Taste stimuli were presented for 5 consecutive licks separated by 5 licks of AS rinse presented on a variable ratio 5 schedule. Preliminary recordings of taste-responsive NTS cells (n = 6) from 7 RYGB rats show that the concentration-response functions for sweet stimuli were not monotonic. There was also no correlation between the responses to sucrose and those to grape juice. In addition, there was only one response to cream, despite the observation that nearly every NTS cell in lean rats responds to cream. Though preliminary, results suggest that NTS responses to sweet and fatty tastes may reflect a compromised gut-brain communication and synaptic reorganization in the NTS following RYGB surgery.

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FCOI Declarations: None.

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#439

Profound Taste/Nutrient Preference Differences between CAST/Ei vs. C57BL/6 Mice

Anthony Sclafani, Soner Vural, Karen Ackroff
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Inbred mouse strain differences in taste and nutrient preferences are well known. Prior results indicate that C57BL/6 (B6) and CAST/Ei mice show high and low preferences, respectively for fat-rich vs. carbohydrate-rich diets and high and low preferences for maltodextrin. However, they show similar preferences for sucrose, consistent with having similar sweet-sensitive T1r3 receptors. We confirmed and extended these strain differences in a series of 2-day nutrient vs. nutrient (2N) and nutrient vs. water (NW) preference tests. In an initial 2N test between isocaloric fat (3.2% Intralipid) and sugar (8% sucrose), B6 and CAST mice displayed fat preferences of 71% and 4%, respectively. After separate NW tests with fat and sugar, which facilitates post-oral nutrient conditioning, their fat preferences were 36% and 6%, respectively. In NW tests with 8% maltodextrin and 8% sucrose, B6 mice consumed similar amounts of the two carbohydrates (28 g/day each) whereas CAST mice consumed more sucrose than maltodextrin (27 vs. 17 g/day). In a subsequent 2N test, B6 and CAST displayed maltodextrin (vs. sucrose) preferences of 74% and 6%, respectively. In a 2N test with 8% starch vs. 8% maltodextrin, both groups preferred maltodextrin. Both groups also preferred 8% starch to vehicle (gum) but the CAST mice had a weaker preference and consumed less starch than B6 mice (6.8 vs. 14.2 g/day). In NW tests with 8% glucose and 8% fructose, B6 mice consumed much more glucose (30 vs. 14 g/day) whereas CAST mice consumed similar amounts of the two sugars (27 vs. 25 g/day). In a 2N sugar test, the B6 and CAST mice displayed glucose preferences of 98% and 30%, respectively. These profound preference differences may represent strain differences in fat, maltodextrin, and glucose taste receptors and/or differences in post-oral nutrient sensors. Further testing (nerve recordings, brief lick tests, intragastric conditioning) is required to reveal the genetic/physiological bases for these disparate nutrient preferences.

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FCOI Declarations: None.

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#440

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

Piriform Cortex Response to Milkshake is Negatively Associated with Saturated Fat Intake

Don Tucker Award Finalist

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Food intake is regulated by homeostatic mechanisms as well as hedonic and sensory signals, including olfaction. Amassing evidence links excess consumption of palatable foods high in saturated fat and refined carbohydrates to perturbations in metabolic and hedonic systems involved in the control of feeding. Moreover, high-fat diet was recently shown to produce olfactory sensory neuron loss and alter olfactory- and reward-driven behaviors in rodents (Thiebaud et al., 2014). However, the impact of such diets on chemosensory system function in humans remains a matter for debate. As part of a clinical weight loss trial, we assessed the relationship between perceptual and hedonic ratings for food stimuli, neural response to receipt of a sweet and fatty liquid, and dietary intake of saturated fat and free sugar in overweight and obese participants (11M, 29F; BMI=33.6 kg/m^2; age=31.1). At baseline, participants tasted a series of flavored puddings and jellos which varied in either fat or sucrose content (puddings: 0%, 3.1%, 6.9%, and 15.6% w/v fat; jellos: 0M, 0.1M, 0.56M, and 1.0M sucrose). Liking and wanting as well as perceived intensity, sweetness, saltiness, fattiness, creaminess, and oiliness were rated using Labeled Magnitude and Visual Analog Scales. In a separate session, we used functional MRI to measure BOLD response to oral receipt of milkshake (MS) vs. a tasteless, odorless solution (TS). Dietary data were collected using a validated semi-quantitative food frequency questionnaire. Preliminary analyses revealed a positive correlation between saturated fat intake and wanting ratings for puddings but not jellos. Greater fat intake was also associated with decreased BOLD response to MS vs. TS in the piriform cortex. This effect was not dependent on age, sex, or BMI, but was abolished when rated wanting for puddings was included as a covariate in the design. Consistent with previous work in animals, these preliminary findings suggest a link between dietary fat intake, chemosensory processing, and food reward.

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FCOI Declarations: None.

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#441

POSTER SESSION IV
SATURDAY, 8 – 10:30 AM

WITHDRAWN

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#442

Gustatory Information Processing in the Nucleus of the Solitary Tract in the Awake, Freely Licking, Diet-induced Obese Rat

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Previous studies have shown that obesity changes both behavioral and brainstem electrophysiological responses to sweet tastes (e.g. Hajnal et al., *Am J Physiol Regul Integr Comp Physiol* 289: R1675-R1686, 2005). The aim of the present study was to determine how obesity changes the taste response profiles of neurons in the nucleus of the solitary tract (NTS) of the diet-induced obese (DIO) rat. Following a minimum of 5 weeks on a high fat diet (60% fat), obesity was verified via DXA body-composition scans. DIO rats were implanted with a chronic 8-channel electrode bundle in the rostral NTS. Following recovery, rats were water deprived for ~22 hrs and placed in an operant chamber where they were allowed to lick freely. Tastants were presented in 5 consecutive lick trials interspersed with 5 licks of artificial saliva (AS) presented on a variable ratio 5 schedule. Tastants (dissolved in AS) were: NaCl (0.05 M), sucrose (0.1 M, 0.5 M), citric acid (0.016 M), caffeine (0.002 M), grape juice (0.1 M, 0.5 M), clam juice (0.05 M), heavy cream (25%), coffee (with 0.002 M caffeine), lemon juice (with 0.016 M citric acid) and AS. Preliminary results from 15 taste-responsive NTS cells suggest that NTS taste cells in DIO rats were more narrowly tuned than those in lean rats (Roussin et al., *J Neurosci*, 32(31):10494 -10506, 2012): e.g. 43% (6/15) of cells only responded to 2 tastants in DIO rats, 16% (2/31) in lean rats. Further, there was no difference in mean response magnitude between the high and low concentrations of either sucrose or grape juice. Interestingly, 44 non-taste-responsive cells were recorded from the same electrode arrays where a taste responsive neuron was recorded. Though preliminary, these data suggest that NTS taste responses may be sparse but narrowly tuned in DIO compared with lean rats. In addition, there may be a flattened concentration- response function for sweet tastants. The results reflect compromised vagal gut-brain communication and intraneuronal reorganization produced by diet-induced obesity.

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FCOI Declarations: None.

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#501

The Transcriptional Factor Ap2e Controls the Expression of the V2r Vomeronasal Receptor Superfamily in Mouse

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The vomeronasal organ is a specialized sensory system responsible for the detection of pheromones. Pheromone signals play a pivotal role in social interaction in a large number of mammals. The vomeronasal organ of mice is composed of two classic types of vomeronasal neurons that selectively express receptors encoded by one of the two vomeronasal receptor (Vr) gene super families: V1r and V2r. The V1r and V2r expressing neurons respectively localize in the apical and basal areas of the vomeronasal organ, express different types of G-protein subunits, and project to spatially distinct areas of the accessory olfactory bulb. How vomeronasal neurons differentiate into one of the two neuronal cell types and how the vomeronasal receptor superfamily genes expression is controlled is still largely unknown. A potential role for the transcriptional factor, Ap2-epsilon (Ap2e), has been previously suggested to be involved in the dichotomy of vomeronasal sensory neurons (Enomoto et al. 2011, Suarez 2011). We analyzed Ap2e expression and genetic lineage in the vomeronasal organ, by exploiting a knock- in/knock-out Ap2eCre mouse line, from embryonic development to postnatal life. Our data indicates that, in the nasal area, Ap2e is selectively expressed by V2r basal vomeronasal precursors and mature neurons. Gene expression profiling, in-situ hybridization and histochemical analysis revealed that that loss of Ap2e expression negatively affects the expression of the entire V2r superfamily in the basal neurons but does not affect the specific neuronal fate choice, proliferation or survival of these neurons. Aknowledgments: Support: Dept. startup package SUNY, Albany; Trevor Williams, Department of Craniofacial Biology, University of Colorado Denver.

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FCOI Declarations: None.

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#502

The Neuronal Recognition Molecule, Neurexin 1, a Potential Coordinator of Innervation in Taste Buds

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Taste bud cells utilize a plethora of receptors to recognize tastants. Taste buds are also innervated by neurons, but how a neuron distinguishes between different subsets of taste cells (e.g. between a *T1R2*- and a *T2R5*-expressing Type II cell) is unclear. In the central nervous system, neurexins are a family of transmembrane proteins that are used by neurons to distinguish between different potential targets. Due to their abundant molecular diversity through alternative promoters and alternative splicing, neurexins are attractive as possible regulators of taste cell innervation. We performed RNA-seq on mouse fungiform taste buds and pools of Type II, Type III and other cells (identified via GFP labels). In this RNA-seq, *Neurexin 1* (*Nrxn1*) was detected in taste buds, was absent from adjacent non-taste epithelium and was prominent in pools of Type III and Type II taste cells. We validated the expression pattern seen in the RNA-seq by performing RT-qPCR. Relative to non-taste epithelium, *Nrxn1* is expressed at 200-fold higher levels in fungiform and palatal taste buds and 500-fold higher in circumvallate taste buds. We validated anti-Nrxn1 by immunohistochemistry on cerebellum. In taste tissue, as in cerebellum, two types of Nrxn1 signals were detected: punctate and more evenly distributed across the plasma membrane. In cryosections of circumvallate papillae and palate, anti-Nrxn1 puncta were located exclusively inside the taste bud. Nrxn1 immunostaining was also detected in the plasma membrane of some taste cells. By triple immunofluorescence in two experiments, 34 of 48 (71%) of Type III cells showed plasma membrane staining. This staining pattern was completely absent in Type II cells (0 of 232 cells). We observed occasional Nrxn1-positive cells that were neither Type II nor III. We propose that taste cells and their innervating neurons utilize the molecular diversity of Nrxn1 to coordinate innervation. By RT-qPCR, we are investigating whether multiple forms of *Nrxn1* are found in taste bud cells and whether such isoforms vary across cell types or across buds in different oral fields.

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FCOI Declarations: None.

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#503

Is There a Change in Olfactory Information Processing During Development?

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Information processing requires morphological structures. The major structure in olfactory information processing is the olfactory bulb; its cells are arranged in specific layers. During postnatal growth and development, especially in altricial animals, the body mass increases several magnitudes, as does the neural system. Taken the fact, that the olfactory sense is functional already from birth on, the question arises: Are the proportion of the olfactory bulb layers kept constant during postnatal development? We therefore analyzed morphometrically 36 female American minks (*Neovison vison var. atratus*) for the layer composition from birth (postnatal day 0, P0) up to P210. Major changes occur within the first month of life but are not restricted to this time window. The fila-layer increases from about 15% to over 20% during adolescence to fall to 18% in adults. The glomerular layer comprises about 12% in young animals (P15-60) to increase significantly to about 15% afterwards. The external plexiform layer as major information processing layer, is only 3% in newborns, indicating not much of information filtering in neonates, and dramatically increases to more than 20% in adults. In contrast, the mitral cell layer is 15% in neonates and falling to values of about 4% in older animals, reflecting the fact that mitral cells are born before birth. The internal plexiform layer increases from 1.5% up to 4.7% in juveniles to fall to 2.7% in adults. The granule cell layer accommodating the information processing neurons, is the major part of the olfactory bulb in most age groups with more than 20% increasing to nearly 30% in adults. The stratum album (P0: 16%) decreases continuously to 9% (P210) as does the subependymal layer (P30: 10%, P210: 1%). The results indicate, that although the olfactory system is functional at birth - in contrast to the visual and auditory system -, the information processing changes during postnatal development by establishing a filtering system, according to the necessity of increasing olfactory challenges to identify milk, social cues, family members, enemies, food and sexual partners.

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FCOI Declarations: None.

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#504

Sex Differences in Maternal Care Determine Effects of Infant Odor Memory on Anxiety-related Behaviors in Adult Rats

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Odors associated with traumatic events during the neonatal period affect emotionality and anxiety-like behaviors in adulthood. We have reported that these effects vary by sex: females are more likely to be affected by an odor experienced during neonatal maternal separation (MS) than are males. The quantity and quality of maternal care that infants receive from their dam may influence the sex differences in odor effects. In this study, we observed maternal behaviors immediately after MS. Wistar-Imamichi dams (n = 7) and their pups (n = 4 of each sex per litter) were divided into two groups [MS, no maternal separation (NMS)]. Pups in the MS group were separated from their mothers for 3 h daily during postnatal days (PNDs) 1-14. Immediately after MS, on PNDs 3, 6, and 9, pup-retrieving behaviors by mothers were observed. The pups were placed in diagonally opposite corners to the nest separately by sex, and the rank order of pup retrieval by their mother based on sex was recorded. Maternal behaviors after MS, including nursing, licking, and nest building, were measured for 10 s every 2 min for 60 min throughout PNDs 1-14. In the Pup-Retrieving test, male pups were more likely to be retrieved before females on PND 3, but not on PNDs 6 and 9. MS increased the amount of nursing and kept it higher until PND 14. In addition, mothers in the MS group showed more licking of male pups than they did of females. These findings suggest that sex differences in maternal care could result in sex differences in the effects of MS on anxiety-related behaviors in adult rats.

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FCOI Declarations: None.

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#505

The Influence of Olfactory Stimulation on Oral Food Intake Examined on Newborns and Premature Infants

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It is assumed that the olfactory sense has some impact on food intake of newborns. Especially premature infants have problems drinking enough and are fed, in parts, by nasogastrale tube. The purpose of this study was to investigate the influence of olfactory stimulation on oral food intake and drinking behavior of newborns. It was examined, whether the lengths of tube feeding can be reduced by olfactory stimulation.57 children, born after 27 weeks of gestation, were included in this study. Before feeding an odor (odor group) or not odor (control group) was presented under the child's nose. Children were randomly divided into the groups. As odors a food associated odor "vanilla" as well as a non-food associated odor "rose" was used. For odor presentation "Sniffin' Sticks" were used. The control group received an empty "Sniffin' Stick". When all children were included no difference between the control group and odor groups were found regarding lengths of tube feeding, weight gain or age at complete oral food intake. After adjusting for percentage of odor presentation before feeding a trend towards shorter tube feeding was found between the control group (15.8±11.4 days) and the odor groups (11.2±7.7 days) (t=1.72; p=0.091). Further analysis revealed a significant effect of odor presentation on the lengths of tube feeding when only children born after gestation week 32 were selected (13.1±10.0 vs 7.3±2.8 days t=2.20; p=0.034). The two groups did not differ in terms age, weight or gestation age when included in this study. No difference between the two odors used was found (t=0.96; p=0.35). The results show that odor stimulation can reduce the lengths of tube feeding in newborns significantly. This effect was only seen when the odors were presented on a regular basis.

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#506

Visuo-olfactory Social Affective Matching in Childhood

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Recognition of emotional facial expressions is a crucial skill for adaptive behavior. Affective matching tasks have been used to investigate across development how facial information is integrated with other sensory information. Considering the suggested affective power of olfaction and its relevance in mediating social information since birth, we assessed olfactory-visual matching abilities in a group of 140 children between the ages of 3 and 11 years old. We presented one of three odor primes (pleasant, unpleasant, and no odor) before a facial preference task (happy vs. disgusted face). Children were instructed to select one of two faces. As expected, children of all ages preferred the happy faces. However, children younger than 5 years of age were biased towards choosing the happy face, irrespective of the valence of the odor prime, whereas from age 5, an affective matching strategy guided the choice of children. Indeed, the odor considered pleasant significantly predicted the choice of happy faces, whereas the odor considered unpleasant predicted the choice of disgusted faces. The present study fills a gap in the developmental literature by demonstrating olfactory-visual affective strategies that affect decision making and it represents an important step towards understanding the underlying processes that shape the typical social mind.

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#508

The Molecular Basis of Individual Differences in the Taste of an HIV Treatment Drug

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Worldwide, 220 thousand children are infected with the human immunodeficiency virus (HIV) annually. As part of first-line therapy for early childhood HIV infection, current international guidelines recommend the co-formulated protease inhibitor lopinavir/ritonavir (Kaletra®). Kaletra ® is given to young children as a liquid formulation or as "sprinkles" that are mixed with small amounts of food. Both formulations are unpalatable to most children, making adherence challenging for many. Sustaining high levels of adherence is critical for good treatment outcomes. As a first step in determining if there are individual differences in the hedonic response to the taste of this drug and, if so, whether such differences in taste perception are related to taste genetics, 84 unrelated women tasted the liquid formulation of Kaletra® and rated its flavor (using the hedonic general Labeled Magnitude Scale which frames affective experience in terms of the strongest imaginable liking (94) to strongest imaginable disliking (-94) of any kind) and provided salivary samples for genotyping (HumanOmni2.5m-8v1-1bead microarray). A genome-wide association analysis was conducted with PLINK and genotypes were imputed using reference panels from the 1000 Genomes Project. We found varied, but reliable, individual differences in taste ratings (i.e., ratings ranged from 33 to -87 which was from strong liking to strongest imaginable dislike), which were explained in part by genetic variation from six regions of the genome (chr 1, 5, 10, 13, 14 and 22). Together, this accounted for the vast majority (71%) of the personal taste ratings. The hedonic ratings of taste of this drug and the genotype- taste relationships were reproducible for five of the six regions in the subset of individuals (N=73) who were retested, using identical methodologies. Knowledge of taste genotype may help tailor future treatment recommendations for antiretrovirals and other medications since even the most powerful medications are not effective if patients reject its taste.

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#509

Development Of Hedonics: Experience-Dependent Ontogeny Of Circuits Supporting Maternal Odor And Predator Odor Responses In Rats

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A major aspect of odor perception is hedonic valence - odors are generally perceived as pleasant or unpleasant. Early olfactory experience shapes odor preferences, suggesting developmental plasticity in circuits mediating odor hedonic responses. Here, using 2-deoxyglucose autoradiographic mapping of neural activity, we identified circuits differentially activated by biologically relevant preferred and avoided odors across rat development. Using both region of interest (ROI) and functional connectivity analyses, we identified specific regions within primary olfactory, amygdala/hippocampal and prefrontal cortical networks that were differentially activated by maternal and male odors. While some activated regions remained stable across development (PN7-23), there was a developmental emergence of others that resulted in an age-dependent elaboration of hedonic response-specific circuitry, despite stable behavioral responses (approach/avoidance) to the odors across age. In addition, we explored the effects of modifying the hedonic response to these odors for example, by co-rearing with a male, which modifies behavioral responses to the odors. This allowed assessment of hedonic circuits in isolation of the specific odor quality and/or intensity. Early experience significantly modified odor-evoked circuitry in an age-dependent manner. For example co-rearing with a male which induced pup attraction to male odor, reduced activity in amygdala regions normally activated by unfamiliar male odor, making this region more consistent with maternal odor. Understanding the development of odor hedonics, particularly within the context of altered early-life experience, provides insight into the development of sensory processes, food preferences, and the formation of social affiliations, among other behaviors.

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#510

The Role of Olfaction in Mate Selection

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Although a number of factors that influence mate selection overlap for men and women, some of them do differ. For example, the importance of physical attributes in selecting a potential mate differs between the sexes, with men valuing visual attractiveness most in a partner and women reporting smell as the most essential feature (Herz & Cahill, 1997). Olfactory information may be important in mate selection because it can be a cue to the reproductive fitness of a potential mate (Trivers, 1972), but it is also possible that the appropriateness of a potential partner in terms of their sexual orientation is also being communicated (Lübke & Pause, 2015). One way to distinguish between these ideas is to examine the importance of olfactory cues in selecting a mate in homosexual individuals, for whom mating is dissociated from reproduction but who would presumably find information about sexual orientation valuable. The present study examined the value of olfactory information in mating by presenting the Romantic Interest Survey (Herz & Inzlich, 2002) to 453 individuals, 150 of whom were gay men. Regression analysis indicated that gay men did not value the sense of smell in mate selection as much as heterosexual men (β = -.39, p =.003), and valued vocal quality more than heterosexual men (β = .29, p =.022). Heterosexual men and women did not differ by sex, with results indicating that both groups value olfactory aspects of a person highly in a potential mate. These findings support the idea that olfaction in mate selection acts as cue to reproductive fitness, since gay men did not value this information to the same extent as heterosexual individuals.

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#511

POSTER SESSION V
SATURDAY, 8 – 10:30 PM

The Influence of Gender and Age on Human Axillary Odor

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There are many anecdotes and "folklore" surrounding human body odor. For example, it is widely thought that aged individuals are more odoriferous than younger people. The results of the current study lend support to the contrary. Axillary odorants from 48 males and females between the ages of 21- 80 were analyzed using gas chromatography/mass spectrometry (GC/MS). The results reveal that younger individuals exhibited higher levels of axillary odorants than older individuals. In fact, younger males were found to produce the highest levels of 17 of the 19 axillary odorants examined, while older females produced the lowest levels for 13 of the 19 compounds. The analytical results are in excellent agreement with sensory results that found age group and gender could be readily discriminated by the majority of 24 sensory panelists (12 of each gender; all relatively young). In addition, odors from the older donors were found to have a lower impact and were less unpleasant than were samples from the younger group. These data facilitated the creation of model mixtures that simulate age and gender related odors. The synthetic models will aid in the creative process of age specific fragrance mixtures for use in personal care products.

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#512

TAAR5-mediated Behavioral Responses to Trimethylamine are Altered by Social Status and Estrous State

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Trace amine-associated receptors (TAARs) are a small set of evolutionarily conserved main olfactory receptors that respond preferentially to amines and that contribute significantly to amine aversion in mice. We observe that the TAAR5 ligand trimethylamine is an exception to this observation as both male and female mice display variable attraction and aversion to this odorant. We sought to understand the parameters that might contribute to the variability in these responses. Trimethylamine is a putrid odorant that is found in decomposing fish, but is also enriched in the urine of adult male mice. We hypothesized that if trimethylamine were a social cue that signifies maleness, then behavioral responses to this odorant may vary with social status in males or estrous cycle in females. To examine the possible effects of social status on trimethylamine valence, adult male mice were paired and their social status determined by examining scent mark patterns that are indicative of either dominant or subordinate animals. Responses of mice to trimethylamine were tested in a place preference assay. Subordinate males were consistently averse, while dominant males were attracted to trimethylamine. We then tested trimethylamine valence in diestrus and estrus female mice. Diestrus females were consistently averse to trimethylamine while estrus females failed to display overt attraction or aversion. Importantly, the behavioral responses to trimethylamine were abolished in both male and female mice that lacked TAAR5, regardless of social status or reproductive condition—demonstrating that both aversion and attraction were mediated by TAAR5. The behavioral response to another amine, isopentylamine, was not affected by either social or reproductive status. Our results indicate that trimethylamine, like other amines, elicits TAAR dependent aversion in mice, but that this presumably innate aversion can be modified by social interactions and reproductive status. Our findings support the idea that behavioral responses elicited by activation of a TAAR can be context dependent and modified by experience.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Evidence of Evidence for Human Chemosignaling: Applying a Novel Meta-Analytic Technique

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Humans have been shown capable of communicating emotional states from a sender to a receiver via so-called chemosignals (i.e., axillary odors). Although research showing this remarkable capacity has gradually expanded over the years, the question is: What is the current state of affairs in terms of actual evidence? To assess this, we relied on a novel and practical meta-analytic procedure, namely *p*-curve analysis (Simonsohn, Nelson, & Simmons, 2014). *P*-curve analysis does not suffer from publication bias and does not have the same flaws as merely counting the number of successful observations in the literature (Borenstein, Hedges, Higgins, & Rothstein, 2006). The main idea behind *p*-curve analysis is that the distribution of significant *p*-values (between .00 and .05) is significantly right-skewed (positive skew) when there is a true effect, uniformly distributed (flat) when the null hypothesis is true, and significantly left-skewed (negative skew) when researchers used questionable research practices (e.g., *p* -hacking) to obtain significant results (Simonsohn et al., 2014). We performed a *p*-curve analysis over all significant key analyses (N = 38, < .05) in 23 emotional chemosignaling studies, as well as on *p*-values of groups of outcome variables (e.g., facial EMG, fMRI) for illustrative purposes. *P*-curve analysis revealed that emotional chemosignaling studies do contain evidential value, Z = -8.24, p < .001 (M replicability index: 65%; M statistical power: 73%). Moreover, there was no "lack of evidence", Z = 3.94, p > .99, and there was no evidence of questionable research practices, Z = 8.24, p > .99 (data did not contain insufficient variance: $\chi^2(42) = 38.82$, p = .39). Some outcome measures (e.g., facial EMG) were nevertheless more robust than others (e.g., perception tasks). In sum, there is evidence for the human capacity to communicate emotional states to other humans by means of chemosignals, a remarkable and apparently robust research field.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Olfactory but not Taste Dysfunction Among Chronic Smokers: Baseline Results from an E-cigarette Intervention

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We examined baseline chemosensory function of chronic smokers enrolled in an e-cigarette intervention versus controls and preliminary NHANES results. **Methods:** Chronic smokers were recruited into the intervention. The baseline sample was 79 adults (mean age=32±10 y) who averaged 18 years smoking over 12 cigarettes/d. The smokers self-reported their smell and taste function (NHANES protocol) and participated in measures of smell (16-item olfactometer identification task) and taste (quinine and NaCl intensities, NHANES protocol) function as well as assessment of propylthiouracil (PROP) bitterness. The smoker's taste function and PROP bitterness were compared with age- and sex-matched non-smokers (n=311). Self-reported taste and smell function as well as measured smell function was compared with 2011/2012 NHANES. **Results:** Twenty-five percent of smokers self-reported a smell alteration, which is similar to the prevalence reported in NHANES (Rawal et al, 2015). However, a greater percentage measured with smell dysfunction (22% mild, 35% moderate, 3.8% severe microsmia,1% anosmia), which exceeds NHANES 2011/12 rates. Taste alteration was reported by 15% of smokers, also similar to NHANES. From taste testing, smokers did not report lower tongue tip quinine or NaCl intensities than controls. For the whole mouth, smokers reported greater 1 M NaCl intensity but no difference in intensity of 0.32M NaCl, quinine or PROP. Although the ratio of 3.2 mM PROP to 1 M NaCl suggested fewer supertaster smokers [chi square (2) =6.99, $p<0.05$], it may be an artifact due to the elevated NaCl intensities among smokers. **Summary:** This sample of chronic smokers showed high rates of olfactory dysfunction, which will allow us to test the effects of switching from cigarette smoking to e-cigarettes on olfactory function. Our data did not support that chronic smokers had depressed ability to taste bitterness.

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#515

Influence of Mild Head Injury on the Olfactory Function in a Pediatric Population

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A close relation between head injury and olfactory dysfunction has been reported. In addition previous work has identified several factors e.g. severity and location of trauma, lengths of posttraumatic amnesia, which are related to olfactory dysfunction. Most of these studies were conducted in adults. Only a few studies have targeted the influence of head trauma on the olfactory function in children. Aim of the current study was to evaluate the influence of mild head injury on olfactory function in a pediatric population. A total of 114 patients age 4-17 years (mean 8.7±3.5 years) suffering from mild head injury (GCS 13-15) were included in this study. In addition 56 age-matched healthy children without a history of head trauma were include as control group. Olfactory function was measured using parts of the "Sniffin' Sticks" test-battery - an olfactory threshold as well as an odor discrimination test. Patients were tested three times with an interval of 4 months during one year after head trauma. Olfactory test scores were compared between patents and the control group. Patients scored significant lower on the olfactory threshold test (7.33±1.82 vs 6.50±1.86; t=2.23, p=0.02). No difference was found in odor discrimination scores between the patients and the control group. Although patients had a significant lower olfactory threshold score, the results were still within normal range. Neither olfactory threshold score nor odor discrimination score changed significantly over the study period of one year. The results show that even mild head trauma has an impact on olfactory function in children. Although olfactory function in patients was reduced it is unlikely that children who suffered mild head trauma will become hyposmic or anomsic.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Olfaction and Depression Characteristics in Patients with ESRD

William J. Heaton[1], Robert F. Gormley[1], Katherine A. Pendergast[1], Naomi Whitson[1], Irene N. Ozbek[1], James Tumlin[2]
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Depression has been tied to deficits in olfactory sensitivity and olfactory discrimination (Croy et al., 2014; Lombion-Pouthier, Vandel, Nezelof, Haffen, & Millot, 2006). Individuals with end-stage renal disease (ESRD) tend to suffer from both depression and olfactory loss (McCurdy, M., Ozbek, I.N., Jones, J.,Tumlin, J., 2014). This study examines how the interaction of depression and ESRD affects olfactory ability in these patients. Patients with ESRD were given the Center for Epidemiological Studies Depression scale (CES-D). They were also given the Wheeler-UTC olfactory threshold test (WUTC) and the University of Pennsylvania Smell Identification Test (UPSIT). The WUTC consists of a range of step-wise concentrations for the odorants of vanilla, pinene, banana, and ethanol. The concentrations were administered to each participant twice in a randomized order by a trained research assistant in order to determine olfactory thresholds. Preliminary data analysis reveals a significant correlation between the depression measure and the UPSIT score (r=.654, p=.000, N=28), threshold of ethanol (r=.735, p=.006, N=12), the olfactory threshold of banana (r=.462, p=.053, N=18), the sensitivity to pinene (r=.503, p=.006, N=28), and the sensitivity and specificity of pinene using Youden's J (r=.421, p=.026, N=28). The relationship between depression and olfactory ability in individuals with end-stage renal disease (ESRD) appears to vary depending on whether olfactory identification is being measured or olfactory thresholds.

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Diabetes, ESRD, and Olfactory Sensitivity

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Variability in olfactory sensitivity has been established among individuals with end stage renal disease (ESRD), even though olfactory sensitivity, on average, is greatly diminished in this population (Jones,J, McCurdy,M., LeMay,C., Ozbek,I.N., Tumlin,J., 2014).Diabetes is the most common cause of ESRD (Evans,T. and Cappell,P. 2000). However, diabetes has also been tied to olfactory deficits independent of kidney disease. This prompted inquiry into the specific role of diabetes in olfactory deficits in ESRD patients. In the current study, ESRD patients were given the Wheeler-UTC olfactory threshold test (WUTC) and separated by whether they had a diabetes diagnosis or not. The WUTC consists of a range of step-wise concentrations for the odorants of vanilla, pinene, banana, and ethanol. The concentrations were administered to each participant twice in a randomized order by a trained research assistant in order to determine olfactory thresholds.Olfactory thresholds were compared for through an independent samples Mann-Whitney U test. Diabetic patients had lower thresholds than non-diabetic patients for pinene (p= 0.013) and banana (p= 0.012). Ethanol trended toward significance (p= 0.064), and there was no significant difference found for vanilla (p= 0.260). Evidence suggests that patients with diabetes were able to smell the odorants better than ESRD patients without diabetes. This suggests the need for future research to determine the factors responsible for the differences between diabetic and non-diabetic ESRD patients.

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Chemosensory Disorders, Burning Mouth Sensation, and Halitosis in Patients with Sjögren's Syndrome

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Primary Sjögren's Syndrome (pSS) is characterised by dryness of the eyes and the oral cavity as well as systemic manifestations. It is an auto-immune disease that primarily affects women and patients may complain of cognitive, neurological and psychiatric disorders in addition to sicca symptoms (xerostomia and xerophthalmia). Research on pSS patients has mainly focussed on the symptoms of xerostomia and xerophthalmia, and little is known about chemosensory function in this group.This study investigated the prevalence of taste and smell disorders and other oral complaints in pSS patients. Twenty patients with pSS (women, mean age 53.75 ± 12.9) and 32 controls (women, mean age 50.47 ± 12.6) were recruited to this study. The gustatory and olfactory function was assessed using «taste strips» and «sniffin' sticks», respectively. Stimulated and unstimulated saliva secretion rates were measured and all participants answered a questionnaire to provide detailed medical history.The results showed that the pSS group was more prone to taste and smell disorders when compared to controls. Statistical analyses showed that the pSS group had significantly lower gustatory ($p=0.002$) and olfactory ($p=0.036$) scores. Dysgeusia was one of the most frequent taste disorders detected in the pSS group compared to controls ($p<0.0001$). In addition, pSS patients complained more often of burning sensation of the tongue and halitosis when compared to controls ($p<0.0001$). There was no significant correlation between saliva secretion rates and the above- mentioned disorders. These results show that pSS patients have different kinds of oral health complaints including chemosensory disorders, burning mouth sensation, and subjective halitosis. Patients' quality of life may be seriously reduced because of these complaints. More research is therefore, needed in this field to understand the cellular mechanisms behind these disorders.

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Olfactory Function in Patients with Smell Disorders before and after Topical Treatment with Drops Containing Retinoic Acid

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Vitamin A plays a decisive role in the regeneration of olfactory receptor neurons. In this retrospective study we investigated the effectiveness of topical vitamin A in patients with post-infectious and post-traumatic smell disorders. Clinical study. A total of 170 patients (age range 18-70 years, mean age 52 years) participated, 46 of whom were treated with smell training only and the remaining 124 patients additionally received topical vitamin A. Olfactory function was measured using the Sniffin' Sticks test kit, a validated technique to investigate odor thresholds, odor discrimination, and odor identification. For two months vitamin A containing drops at a dose of approximately 10.000 IU/d were applied topically by tilting the head backwards. Follow-up testing was performed approximately 6 months after the first investigation.Thirty-seven percent of all postinfectious patients treated with vitamin A exhibited recovery of their sense of smell, whereas only 23% improved in controls. In addition, when analyzing the two groups separately for the postinfectious group odor thresholds and odor discrimination improved significantly.The topical application of vitamin A at a dose of 10.000 IU per day for two months appears to be useful in treatment of olfactory loss.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Odor Localization in the Absence of the Corpus Callosum

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Contrary to all other sensory systems, olfactory information is processed predominantly in an ipsilateral manner. Furthermore, stimulus localization, based on inter-nostril differences, is impossible for pure odorants. These two observations suggest information exchange between both cerebral hemispheres in the olfactory system, but the exact anatomical substrate is hitherto unknown. This study aimed at identifying this anatomical substrate of odor localization, assuming a role for interhemispheric communication in this process. We tested the ability to localize pure olfactory and mixed olfactory/trigeminal stimuli in six participants with different forms of structural interhemispheric deficits (agenesis or surgical transection of corpus callosum and/or anterior commissure). We compared their results to those of 46 healthy participants. The study was approved by IRB. Of the six participants in the experimental group, three were not able to localize any of the two stimuli, while two participants had scores significantly different from chance performance for both pure and mixed stimuli. Only one participant exhibited the typical pattern (an ability to localize the mixed olfactory/trigeminal stimulus, combined with an inability to localize the pure olfactory stimulus), which we observed in the majority of the control participants. These results suggest that localization of chemosensory stimuli depends, at least in part, on the corpus callosum.

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Optical Interrogation of Olfactory Tubercle Connectivity with the Piriform Cortex

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How second-order olfactory structures innervate and subsequently modulate one another has important implications for how olfactory information is distributed throughout the brain. Axonal association fibers from piriform cortex (PCX) pyramidal neurons innervate a number of olfactory structures, including the lateral entorhinal cortex, anterior olfactory nucleus, and olfactory tubercle (OT). Evidence for a functional role of the corticostriatal connections between the PCX and OT is not available. Thus, the aim of this study is to determine whether or not PCX pyramidal neurons modulate the encoding of odors within the OT. To accomplish this, we transduced PCX neurons with an adeno-associated viral vector expressing humanized channelrhodopsin under control of the CamKIIα promoter, allowing us to specifically target PCX pyramidal cells. We histologically verified that PCX cells were transduced with the vector, with dense reporter fluorophore expression observed in layers ii and iii. In these cases we also observed that association fibers from the PCX innervated layer i of the OT, as previously reported. Having validated this approach, mice were shaped to perform an olfactory task in which they were presented with an array of odors as extracellular OT activity was recorded either with or without concurrent optical stimulation of the PCX. Our results show that stimulation of PCX neurons elicits excitation within a subset of OT neurons. Some OT neurons display a sustained increase in firing rate throughout PCX stimulation, while others display shorter, transient bursts of firing. Preliminary data also suggest that stimulation of PCX neurons may modify how odors are encoded among OT neurons. Together, these results show that the PCX modulates the activity of the OT, possibly in a manner important for olfaction.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

A Computational Role for Cortical Feedback in Odor Detection in Natural Scenes

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Rodents use a repertoire of ~1,100 odorant receptors (ORs) to represent several orders of magnitude more olfactory objects. Despite large changes in concentration and turbulent odor flow, they readily identify weak target odors in rich sensory scenes, where several strong background odors can be present. To date, the neural mechanisms underlying this complex computation remain unknown. Here we present a novel algorithm that creates an estimate of odor input identity using as few elements (non-zero contributions) as possible from a large, previously learned dictionary representing possible odor sources. Our algorithm uses as cost function the sum of the squares of the contributions (L2 minimization) to find a sparse solution, as opposed to the widely used sum of absolute values (L1 minimization). It is implemented as a real-time predictive coding scheme, where the current estimate of the sources present, combined with the current odor input create iteratively a new estimate of the odor input. The resulting estimation error is further used to update the estimate of which sources are present. The model mirrors biologically the olfactory bulb (OB)-to-piriform cortex circuit, and assigns a critical role to cortical- bulbar feedback signals. The model predicts: **1)** existence of two distinct feedback channels that differ in response polarity to odor stimulation (enhanced vs. suppressed); **2)** existence of two OB output channels, one that represents the estimation error, and is suppressed by cortical feedback, and a second channel that broadcasts incoming sensory input to the cortex, and is independent of cortical feedback. We successfully cross-validated these predictions using multiphoton calcium imaging in awake head- fixed mice and monitoring: **1)** corticalbulbar feedback boutons and **2)** dynamics of mitral and tufted cells before and after suppression of cortical feedback. Our model represents a major advance in understanding cortical feedback, creating a computational framework that is closely corroborated by experiments.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Advances in SenseLab's Interoperable Neuroinformatics Databases: FunctionalMicroconnectomeDB and ModelDB

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SenseLab's suite of interoperable databases (senselab.med.yale.edu) supporting research in olfaction and other brain regions continues to grow. Here we highlight several current advances in microconnectomics and computational models. Microconnectomics: To understand the function of the brain it is critical to know 1) which cells are the sources and targets of the connections, 2) where the connections are made on the dendrites, and 3) the type of synapse (excitatory, inhibitory, etc.). The functional consequences of the spatial placement of connections and the type of receptors involved is critical to forming the input/output operations of a cell or circuit. SenseLab integrates this research by providing the new ability to browse between the different spatial scales of brain region connectivity, cell sources and targets, and the synaptic types, and also links to models that have been built at their different spatial scales. This new microconnectomics resource can be searched, extended as new data is obtained, and browsed in new versatile multi-scale visualization tools at a new FunctionalMicroconnectomeDB. Models: ModelDB archives over 1,000 models which an integral part of research on cells and systems. ModelDB provides several search tools (full text plus database keywords) to identify models of interest. As an example, searching on "olfactory" creates a pop-up menu whose full text menu items at the top lead to 55 models entries (currently all olfactory related models) while database keyword results narrow these results to 12 Olfactory Cell Types (Main Mitral cell, the most frequent, has 30 models), 2 Regions (Olfactory bulb (20) and Olfactory cortex (4)) and Olfactory Receptors (3). User functionality will be demonstrated at the poster.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Respiration Gates Sensory Input Responses in the Mitral Cell Layer of the Olfactory Bulb

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In the olfactory bulb, unique interactions occur between respiration- and odor-evoked glomerular inputs. Baseline burst firing during the transition from inhalation to exhalation is followed by synchronized lateral inhibition that quiets mitral and tufted cell (MTC) activity. Periglomerular cells (PG) tune sensory inputs in the glomerular layer. Inputs that reach the MTC somas backpropagate into the lateral dendrites where they are balanced by feedback and lateral inhibition from granule cells (GC). *In vivo* optogenetics and electrophysiology and neural network modeling were used to provide mechanistic explanations of how the bulb may process discrete dorsal glomerular inputs. MTC processing of olfactory sensory neuron inputs during a respiratory cycle was analyzed. Respiration was found to gate excitatory sensory inputs to periods preceding baseline burst firing, causing net shifts in MTC activity across the cycle. Computational models found that PG and GC inhibition, as well as respiration-mediated burst firing, shaped tuning of sensory inputs across the respiratory cycle. Respiration modulates temporal processing of sensory inputs by excluding them from late bursting phases of the breath cycle, which may aid odor computation by resetting the network on each sniff.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Possible Roles for Dopamine and Vasoactive Intestinal Polypeptide in Circadian Rhythms of the Olfactory Bulb

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Circadian rhythms affect nearly every aspect of biological function from behavior to genetics. The suprachiasmatic nucleus (SCN) controls these rhythms throughout the body, and these rhythms are lost when the SCN is destroyed. Two notable exceptions to this are the retina and the olfactory bulb (OB), both of which, while normally influenced by the SCN, maintain rhythms in the absence of the SCN. In the OB, this manifests as rhythms in clock genes, neuronal activity, and odor sensitivity. However, the cellular and molecular mechanisms underlying these phenomena are not well understood. Previous results from our lab demonstrate that OB neuronal excitability may be influenced by the rhythmic expression of gap junctions, glutamate receptors, melatonin receptors, and monoamine transmitters including dopamine (DA). It has been previously demonstrated that DA plays a critical role in regulating the circadian release of prolactin from the hypothalamus. This, in turn, is regulated by vasoactive intestinal polypeptide (VIP), and our pilot data indicate that VIP regulates firing frequency of hypothalamic DA neurons. Our pilot immunocytochemical OB data indicate that VIP receptors are highly expressed in juxtaglomerular neurons (JGNs) including OB DA neurons. Because VIP also has been implicated in regulating circadian activity within the OB, we are currently exploring the electrophysiological effects of VIP on JGNs as another possible contributor to the cellular and molecular mechanisms underlying circadian olfactory function.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Investigating Attentional Modulation of Odor Coding in the Olfactory Tubercle

Don Tucker Award Finalist

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Depending on the internal state of an individual, the same sensory stimulus may be perceived differently at different times. Attentional processes are especially well known to allow an individual to filter out irrelevant stimuli to focus on aspects of the environment that are relevant for survival. What are the underlying neural processes that contribute to this difference in perception? While this question has been investigated at the cellular level in sensory systems such as vision and audition, very few studies have explored attentional modulation of olfaction at this level. Human functional imaging research has, however, uncovered evidence that odor-directed attention modulates the representation of odors in the olfactory tubercle. While the rodent olfactory system lends itself well as an ideal model to explore the influences of attention at the cellular level, behavioral tasks involving manipulations of selective odor- directed attention in rodents are unavailable. To overcome this, we designed a novel two-alternative choice behavioral task to precisely and systematically manipulate odor-directed selective attention, in which the subject must shift its attention to the relevant (rewarded) modality when presented with simultaneous olfactory and auditory cues. Our results to date display the role of attention in modulating odor-guided behavior, and provide initial insights into the attentional modulation of odor coding in the olfactory tubercle. Together, these results enhance our understanding of how attention shapes sensory processing and perception, which are impaired in a variety of neurological disorders, including Alzheimer's disease.

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FCOI Declarations: None.

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Size-Dependent Characteristics of Glomerulus in Mammalian Olfactory Bulb

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The neuronal architecture of mammalian olfactory system has been extensively studied through identifying the topographical projection of olfactory sensory neurons (OSN) to its glomeruli. The size and density of each glomerulus was suggested to be correlated by the olfactory receptor (OR) expression level in each OSN. However, the volume-dependent identity of molecular and anatomical characteristics of each glomerulus has been poorly understood. Here, we presented the quantitative analysis of about 125 OR mRNAs expression in OE and OB which revealed different expression level. Next, histological property was examined through olfactory marker protein (OMP) and cresyl violet (CV) staining. Interestingly, each glomerulus set was distinctively distributed in region specific manner by their volume and density. Further, different number of mitral cell (MC) and tyrosine hydroxylase (TH)-positive periglomerular cell (PC) was presented by small- versus larger-glomerulus size. The axonal innervation of PC to each glomerulus also differed by the size of each glomerulus. Taken together, we suggest that characteristics of different glomerular size may support the process of different olfactory cognition.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Glutamatergic Modulation of Mitral Cells by Pre-proglucagon Neurons (dSACs) in the Olfactory Bulb

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Mitral cells (MCs) constitute the principal relay of olfactory information between olfactory sensory neurons and higher processing areas of the brain. We previously characterized neuromodulation of MCs by the incretin (gastrointestinal) hormone glucagon-like peptide-1 (GLP-1), which increased the firing frequency of evoked action potentials (APs) by shortening the interburst interval. Using a transgenic mouse model expressing YFP under the control of preproglucagon (PPG), the precursor of GLP-1, we have discovered a population of GLP-1 producing neurons located in the inner part of the granule cell layer (GCL). Histologically, the PPG neurons are characteristic of deep-short axon cells (dSACs) or Cajal cells having stellar dendrites and one axon projecting to the MC layer and the external plexiform layer. To optogenetically activate this class of PPG-neurons, we crossed PPG-Cre mice to floxed ChannelRhodopsin-2 mice. The resultant progeny were used in acute slice preparations of the olfactory bulb whereby light-activation parameters for PPG-neurons were empirically determined by voltage- clamp. Light-activation of the dSACs evoked simultaneous inhibitory (IPSCs) and excitatory (EPSCs) postsynaptic currents in MCs that were abolished after application of glutamatergic inhibitors (APv, NBQX). In current-clamp, light-activation produced a biphasic inhibition-excitation control of AP firing in the MCs. Light-activation of the dSACs, while recording alternatively from granule cells, elicited glutamate-induced EPSCs, supporting a di-synaptic origin of IPSCs recorded from the MCs. This was further corroborated by confocal localization of the glutamate transporter VGLUT2 in the dSAC synaptic terminals. Taken together, these results demonstrate that this class of dSACs/PPG-neurons constitutes a unique population of glutamatergic neurons within the GCL that form a local microcircuit controlling MC activity.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Hedonic Valence Guides Convergence of Oral Trigeminal and Taste Signals in the Mouse Parabrachial Nucleus

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Neural convergence of gustatory and oral trigeminal signals is discussed to play a role in flavor integration and intraoral clinical disorders in humans. Yet the logic of taste and trigeminal interaction remains enigmatic. Limited data in rodents suggest central trigemino-taste convergence involves brain stem nuclei including the parabrachial nucleus (PbN). Here, we used electrophysiologic methods in anesthetized C57BL/6J mice to identify and study orosensory PbN neurons receiving dual input from taste and trigeminal pathways. For individual cells, taste sensitivity was indexed through oral delivery of temperature-controlled solutions of (in M) 0.1 NaCl, 0.5 sucrose, 0.01 quinine, 0.0001 cycloheximide, 0.01 citric acid, and an umami mixture. Receipt of synaptic input from trigeminal circuits was evaluated by monitoring for orthodromic spikes that followed electrical stimulation of the spinal trigeminal subnucleus caudalis (Vc), which projects to the PbN and is implicated for oral nociception. Neurons were also tested for oral sensitivity to thermal stimuli (5° and 55°C) and chemical agonists of transient receptor potential (TRP) ion channels expressed by trigeminal fibers and involved with nociception including (in M) 0.001 allyl isothiocyanate (AITC; TRPA1) and 0.001 capsaicin (TRPV1). Across 33 PbN neurons, all responded (spikes) to at least one taste stimulus, with 24 (73%) of these cells capable of generating spikes that followed stimulation of the Vc with synapse-like lag (Vc+ cells). A subset of Vc+ taste neurons responded to oral delivery of the nociceptive stimuli 55°C, capsaicin, and AITC, indicative of input from TRPV1 and TRPA1. Sensitivity to nociceptive stimuli was associated with aversive gustatory tuning (*P* ≤ 0.01) and emerged selectively in Vc+ PbN neurons that responded, taste-wise, to only quinine and cycloheximide ($n = 6$). These nociceptive-bitter neurons appeared to signal negative affect shared across modalities. Our data show multisensory convergence of taste and trigeminal signals in the brain stem is guided by hedonics and reveal involvement of "taste" cells in this process.

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FCOI Declarations: None.

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#530

Chemesthesis in the Earthworm, Lumbricus terrestris

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Earthworms are often used as biomarkers for healthy soil - the more worms present, the healthier the soil. They can have significant impact on soil properties (e.g. gas, water and solutes transfer), on soil processes (e.g. biogeochemical cycles) and on soil structure. As a result, earthworms improve the conditions of soil, which leads to improved plant growth. Chemicals in the soil which repel earthworms could therefore have a deleterious effect on plant growth. Currently, very little is known about earthworm chemoreception in general, let alone how they respond to chemical irritants. Perhaps the most relevant research done to date involves using chemical expulsion to sample the number of earthworms in an area of soil. Allyl isothiocyanate (AITC) or mustard oil is often the chemical used for chemical expulsion, which demonstrates that earthworms can detect and are repelled by a known chemesthetic irritant. In the current study, we used a T-maze assay consisting of a funnel connected to a T-connector to examine the earthworm's ability to detect irritants. A worm is placed into the funnel and a bright light causes it to move into the T-maze, which has filter paper in both arms, one soaked with water or mineral oil, the other with test compound. Worms significantly avoided all concentrations of AITC and cinnamaldehyde tested (10mM - 100mM) (TRPA1 agonists) but not menthol (TRPM8 agonist) or capsaicin (TRPV1 agonist). Using immunohistochemistry on earthworm skin epithelium, we examined antibody binding to 5 different homologs of TRPA1 channels [*C. elegans* TRPA1 (A), fruit fly dTRPA1 (B), Painless (C), Pyrexia (D), and Human TRPA1 (E)]. Some epithelial cells displayed positive immunoreactivity for A, B, and E. Positive immunoreactivity for D was found in the muscle layer and no immunoreactivity was seen anywhere for C. We are continuing our experiments to determine what other compounds, both natural and man-made, repel earthworms and what cellular mechanisms are responsible for this repellency.

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#531

Taste Buds and Solitary Chemosensory Cells in the Infant Human Larynx

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Laryngeal inflammation can impede vocal production, swallowing, and respiration, thereby affecting communication, quality of life, and overall health. Disorders associated with laryngeal inflammation are commonly treated with proton pump inhibitors to target suspected reflux; however, in many cases this treatment demonstrates limited efficacy, warranting continued examination of the underlying pathophysiologic mechanisms of inflammation. Recent findings in the nasopharynx indicate that detection of chemical irritants - by either free nerve endings or solitary chemosensory cells (SCCs) - induces immediate, local neurogenic inflammation. To date, the role of SCCs in the larynx is unknown. While previous studies in humans have identified laryngeal taste buds, little is known about taste cell types associated with these structures. Here, we characterize chemoresponsive elements, including taste buds, SCCs, and polymodal nociceptive nerve fibers, in the laryngeal epithelium of infants undergoing routine surgical management of laryngomalacia. Using immunohistochemistry and RT-PCR we examine the structure and density of SCCs and taste buds and identify their downstream signaling effectors. Preliminary findings show in 3 of 5 samples, numerous densely innervated taste buds, including Type II taste cells. Ongoing analysis will identify markers for Type III cells and polymodal nociceptive nerve fibers. These findings will contribute to an understanding of the structure and distribution of SCCs and taste buds in the human larynx.

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FCOI Declarations: None.

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A Study of Solitary Chemosensory Cells in the Lamprey

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The lamprey is an excellent basal vertebrate model for studying sensorimotor functions. In this species, putative solitary chemosensory cells (SCCs) have been reported on the surface of the body, including on finger-like extensions named "papillae" that are abundant around the gills. The objectives of this study were to investigate changes in the density of gill SCCs during the life cycle, and to characterize the biochemical properties of these cells. Scanning electron microscopy was utilized to identify and to quantify SCCs on the gill papillae. The highest SCC density was observed during the late stages of the life cycle, when lampreys were migrating upstream, selecting nesting sites and spawning. SCC density was lower during the earlier stages, when lampreys were feeding. These findings suggest that SCC inputs might be important for upstream migration, nest site selection and/or spawning. Western immunoblots and immunocytochemistry were applied to examine the biochemical characteristics of the SCCs. The gill SCCs were calretinin- and serotonin-positive, as previously shown in lamprey pharyngeal taste cells and in the SCCs of other vertebrates. Labeling for phospholipase C was also observed, suggesting that chemosensory signal transduction occurs by an IP_3 mediated cascade, as is the case in mammalian SCCs. Taken together, our findings suggest that SCCs are particularly important during the late stages of the sea lamprey life cycle and lamprey SCCs may be homologous to SCCs described in other vertebrate species.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

A Role for PROP Status in the Salivary Protein Response to Astringency

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Astringency is a complex sensation associated with drying, puckering and roughing of the oral surfaces that is mediated by the interaction of polyphenols (tannins) with salivary proteins. Individual differences in astringency perception are well known, but poorly understood. We recently reported that male PROP non-tasters perceived less astringency from and gave higher liking ratings to cranberry juice cocktail supplemented with 1.5% tannic acid than did male medium and super-tasters (Mattes et al., 2015- abstract). Here, we examined the salivary protein response to 1-min of oral stimulation with 20 mL of 2g/L tannic acid or cranberry juice (prepared from fresh berries according to a standard recipe) in 77 young, adults classified by PROP status. Five families of proteins (basic proline-rich proteins (bPRPs), acidic proline-rich proteins (aPRPs), Histatins, Statherin, & Cystatins) were identified from whole saliva by HPLC-ESI-IT-MS and quantified using the area of the extracted ion current (XIC) peaks. Compared to baseline (resting), tannic acid stimulation reduced the levels of aPRPs and bPRPs ($p<0.0001$ for both) in agreement with reports showing that these proteins bind polyphenols to form large aggregates. However, no effect of PROP status was observed. In sharp contrast, cranberry juice stimulation increased the levels of aPRPs, bPRPs and Cystatins ($p< 0.03$-0.0001); this was likely due to its high pectin content, which interferes with protein binding. Unexpectedly, there was a strong effect of PROP status on levels of aPRPs and Cystatins after cranberry juice, where levels rose sharply in medium tasters and super tasters ($p<0.001$-0.0004) but not in non-tasters. These proteins have anti- microbial properties and have been implicated in the host defense system against oral disease. These novel findings are consistent with a proposed protective role for PROP status in oral health. The present findings will be discussed in the context of current theories of astringency development and oral health.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Hot as a Jalapeño, Cool as a Cucumber: Trigeminal Ganglion Neuron Responses to Capsaicin, Allyl Isothiocyanate (AITC) and Menthol in Mice

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Certain plant-derived compounds such as capsaicin from chilis, allyl isothiocyantate (AITC) from mustard, and menthol from mint add pungency and change the perception of oral thermal sensations. For instance, warm foods/beverages seem hotter after eating chilis and mint feels cool. We have begun to explore the neural basis of these effects using confocal Ca^{2+} imaging of trigeminal ganglion cells in mice that express the Ca-indicator GCaMP3 in sensory neurons (Kim et al, *Neuron* **81**:873; 2014). Mice were deeply anesthetized and cortical tissue aspirated via a lateral window in the cranium to expose area V3 of the trigeminal ganglion. We identified this area as containing neurons with receptive fields in the oral cavity. Aqueous solutions of capsaicin, AITC, and menthol at physiologically appropriate concentrations, and distilled H_2O at 2° to 45° were delivered retrogradely as 10 sec oral lavages via a catheter inserted through the esophagus. Capsaicin evoked robust responses in progressively larger numbers of neurons when applied at 3 to 100 µM. For example, imaging fields of 200 to 800 trigeminal neurons showed that 10 µM capsaicin stimulated 0.2-0.5% of the cells; 100 µM, 2-5% of cells (16 mice). AITC (20 mM) stimulated 2-3% of cells (4 mice). Menthol at 1 mM activated ~0.4% of trigeminal neurons; at 10 mM, 2.5% (6 mice). Each of these three stimuli elicited responses in distinct populations of trigeminal neurons. Capsaicin exerted a long-lasting enhancement of responses to stimulation with warm (37°) and hot (45°) aqueous lavage. That is, the numbers of responding neurons and the amplitudes of Ca^{2+} responses to warm and hot temperatures increased after prior oral lavage with capsaicin. Our data replicate what is commonly experienced when consuming pungent spices and demonstrate that thermal perceptions from chemical stimuli can be generated at peripheral sites independent of higher levels of cortical integration and processing. The preparation promises to be a powerful tool for studying somatosensations, including pain, generated during eating and swallowing.

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FCOI Declarations: None.

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POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Conditioned Oral Behavioral Responses to Trigeminal Stimuli in Mice

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The oral trigeminal system is usually thought of as serving an innate protective function in analyzing ingested food. Trigeminal stimuli, for example, capsaicin and mustard oil, are classified predominantly as irritants and aversive to animals. However, there has been little investigation on how animals react to trigeminal stimuli in orosensory conditioning paradigms. Pilot work from our lab revealed mice do not show orosensory avoidance to micromolar concentrations of menthol, a TRPM8 agonist with therapeutic properties, albeit these concentrations strongly activate trigeminal afferents. Menthol, therefore, allows for a potentially unique way of understanding trigeminal stimulus processing in terms of conditioned responses. In the present study, mice (n=6) were differentially conditioned using a brief-access licking aversion paradigm in the Davis Rig. Stimuli were 0.64mM menthol (CS+) and 0.3M sucrose (CS-). CS+ conditioning sessions consisted of partially water-deprived mice licking menthol in brief access trials followed by injection of either 0.15M LiCl or physiological saline over the course of three days. For two days between CS+ conditioning sessions, animals were presented with 0.3M sucrose (CS-) followed by no injection. Mice were then tested with either water and 0.64mM menthol or water and 0.3M sucrose. During conditioning, a reduced lick rate and number of licks indicated animals conditioned with LiCl showed decreased intake of menthol compared to saline- conditioned animals ($p<0.05$). Test data indicated that LiCl conditioned animals significantly avoided menthol compared to saline conditioned animals ($p<0.05$). Furthermore, LiCl conditioned animals did not show avoidance generalization to sucrose during testing. These data suggest that micromolar concentrations of menthol are perceived as innately innocuous and that animals form an aversion to them through conditioning. Work is currently ongoing looking at conditioned aversive responses to trigeminal-taste mixture objects.

Funding Acknowledgements: NIDCD DC011579.

FCOI Declarations: None.

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#536

POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Dose Response Function for Capsaicin in the Context of Reported Liking and Intake of Chili Peppers

Don Tucker Award Finalist

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Capsaicin is a widely consumed ingredient that elicits sensations of heat and burning. It is well documented that rated intensity varies as a function of prior use; however, less is known about the liking of sampled capsaicin. Here, we revisit the relationships between reported intake with burning, and bitterness, while also investigating reported pleasantness over a wide range of capsaicin concentrations. Eighty-three participants (34 males) completed a two-day study, which involved rating 8 capsaicin stimuli (0.11, 0.275, 0.55, 1.1, 2.75, 5.5, 11 and 22ppm) diluted with water (and 0.1% ethanol). To minimize fatigue, 4 stimuli were presented each day in alternating ascending order, with a 2.5-minute break between stimuli. Intensity ratings were made on general labeled magnitude scale and pleasantness on a bipolar generalized hedonic scale. Additional questions included: willingness to try novel foods (VARSEEK questionnaire); and a variety of questions regarding intake frequency and liking of foods.

Burning was the dominate sensation, increasing with concentration; bitterness also increased, but reached a much lower maximum. Reported intake of various capsaicin containing foods were annualized and pooled, resulting in an intake group (low and high) based on a median split (134 times/year). ANOVAs revealed intake was associated with ratings of pleasantness ($F(7,566)=2.6$, $p=0.01$) and burning ($F(7,566)=2.8$, $p=0.007$) across concentrations. Additionally, stated preference for heat level (mild, medium, etc.) was associated with pleasantness of sampled capsaicin ($F(14,559)=2.3$, $p=0.004$). VARSEEK scores were significantly correlated with pleasantness for 2.75ppm and 11ppm capsaicin (R^2 = 14.0 and 9.7%, respectively), but not burning. The dose response function generated for capsaicin will aid in the design of future experiments. It is apparent, from data presented here and elsewhere that both perceived intensity and pleasantness of capsaicin differs by a variety of factors, including personality measures, and prior intake of spicy foods.

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FCOI Declarations: None.

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#537

Putting Out The Fire: The Efficacy Of Common Beverages For The Mitigation Of Oral Burn From Capsaicin

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Capsaicin is known for its ability to elicit warming and burning sensations. While many people enjoy this burn, it may be overwhelming if chili containing foods are over consumed, necessitating rapid relief. While there is substantial folklore on the ability of specific beverages to mitigate burn, there is minimal data to support such claims. Here, we explored the ability of various common beverages to reduce burn following consumption of spicy tomato juice. Participants (n=72, 42 females) rated the oral burn of room temperature tomato juice (5-Pepper Spicy Bloody Mary Mix, Master of Mixes, 30 mL) on a general Labeled Magnitude Scale (gLMS) immediately after swallowing, and again every 10 s for 2 min. On seven of eight trials, a test beverage (40 mL) was consumed after the tomato juice was swallowed. These included skim milk, whole milk, seltzer water, Cherry Kool-Aid, non-alcoholic beer, cola, and water. Participants also answered questions regarding intake of spicy food. The initial burn of the tomato juice alone was rated near “strong” (33.4+- 2.4) and continued to decay over the two minutes to a mean just above “weak” (9.03 +-1.4). All beverages except seltzer significantly reduced the burn ($p<0.05$) of the tomato juice immediately after consumption. To test efficacy over time, ANOVA was conducted on area under the curve (AUC) values: the burn over time was significantly reduced for whole milk, skim milk, and Kool-Aid. Contrary to expectations, no evidence of a frequent chili user / nonuser effect was seen for rated burn. More work is needed to determine mechanism(s) behind these data: they may be partitioning due to fat, binding by protein, or endogenous opioid release due to sweetness. Empirically, we conclude milk is the best choice to mitigate the burn following the consumption of spicy foods.

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FCOI Declarations: None.

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#538

Fetal Ethanol Exposure Reduces Taste and Trigeminal Responses of Adolescent Rats to Ethanol and Chemical Surrogates for its Flavor Components

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Fetal ethanol exposure leads to increased intake of ethanol in adolescent rats and humans. We asked whether the elevated intake of ethanol by adolescent (P30) rats stems in part from diminished responsiveness of the peripheral taste and oral trigeminal systems to its flavor components. To study taste, we recorded responses of the chorda tympani (CT) and glossopharyngeal (GL) nerves to lingual stimulation with ethanol, NaCl (a control stimulus), and chemical surrogates for ethanol's bitter and sweet taste quality components (quinine and sucrose, respectively). To study trigeminal orosensation, we collected isolated trigeminal ganglion neurons, and measured changes in intraneuronal calcium levels during stimulation with ethanol, capsaicin (a selective TrpV1 channel agonist), mustard oil (a selective TrpA1 channel agonist), and KCl (a control stimulus). We exposed the experimental rats to ethanol *in utero* by administering ethanol to dams through a liquid diet; we exposed the control rats to an iso-caloric iso-nutritive liquid diet. As compared with control rats, the experimental rats exhibited significantly diminished CT and GL nerve responses to ethanol, quinine and sucrose (but not NaCl). The experimental rats also exhibited significantly reduced trigeminal neuron responses to ethanol, capsaicin and mustard oil (but not KCl). The lack of change in NaCl and KCl responsiveness indicates that fetal ethanol exposure selectively attenuated specific response pathways within the peripheral taste and trigeminal systems. These findings uncover epigenetic orosensory mechanisms by which maternal patterns of ethanol consumption can be transmitted to offspring.

Funding Acknowledgements: NNIH-NIAAA grant AA017823.

FCOI Declarations: None.

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#539

Cross-modality Selective Adaptation of Chemosensory Binary Mixtures by Humans

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In natural settings, taste and odor identification depends on sensory adaptation, a loss in distinctiveness over time. In selective adaptation a binary mixture is first adapted to one component, which loses distinctiveness, while identification of the un-adapted 2nd component improves, if separate receptors are involved. Previous studies on selective adaptation used compounds of the same modality (Frank *et al.*, 2012). In the current experiment we adapted salt and sugar tastes to vanillin and phenethyl alcohol (rose) odors and vice versa. We hypothesized sweet odors like vanilla and the congruent sweet taste of sucrose would be reciprocally selectively adapted. Twelve volunteers, aged 18-29 with no history of taste or smell disorders, participated in 4, 1-hour, sessions. Subjects were first trained to identify all stimuli, liquid: 0.1M NaCl-N, 0.15 M sucrose-S and water; volatile: 5mM vanillin-V, 5mM phenethyl alcohol-R and water. Subjects protruded their tongues to accept tastes and squeezed flip-cap bottles to sniff odors. The key 30 adapt-test pairings were delivered in 1 hour for identification of 2nd bottle contents. Single modality binary mixture component identification confirmed previous studies. Proportions correct for N before NS were 0.17-N, 0.92-S and S before NS 0.92-N, 0.13-S ($p < .0002$). However, after any odor (including volatile water), identification of salt decreased ($p = .02$) and sucrose increased ($p = .008$) compared to mixtures adapted to liquid water. Vanilla's sweet odor did not selectively adapt sucrose's sweet taste. Rather, cross-modality adaptation (sniffed volatile before liquid taste) enhanced sugar taste at the expense of salt taste. The mechanism that suppresses salty tastes and enhances sweet tastes immediately after voluntary sniffing is unknown.

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FCOI Declarations: None.

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#540

POSTER SESSION V
SATURDAY, 8 – 10:30 PM

Temperature Affects Bitter Taste Sensitivity and Adaptation in a Stimulus-dependent Manner

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Studies of the effect of temperature on bitter taste have not always agreed, and no prior study had investigated the possible effect of temperature on bitter taste adaptation. Interest in a possible effect on bitter taste adaptation arose from our recent finding that temperature affects sweet taste adaptation differentially across sweeteners. In the present experiment we measured suprathreshold bitterness for 2 stimuli (caffeine and QHCl) at 4 different temperatures (10°, 21°, 30° and 37°C) in 2 conditions. In the baseline condition 25 subjects dipped the tongue tip into one of the bitter stimuli for 3 s before rating bitterness intensity on the gLMS. In the adaptation condition subjects dipped the tongue into either dH_2O (temperature-only pre-exposure) or one of the bitter stimuli (temperature + stimulus pre- exposure) until signaled by the experimenter (after 3 or 10 s) to dip the tongue into a second cup containing the same bitter stimulus at the same temperature. After 3 s the subject lifted the tongue from the second cup and immediately rated the bitterness of the solution while keeping the tongue tip outside the mouth. The results showed that baseline sensitivity to both stimuli followed an inverted U-shaped function of temperature. However, adaptation to QHCl was minimal at 21°C and maximal at 37°C, whereas adaptation to caffeine was rapid and independent of temperature. These data indicate that like sweet taste, temperature affects bitter taste via at least 2 mechanisms, and that the effect on adaptation varies across stimuli. The stimulus dependence of thermal effects on adaptation implies that for both sweet and bitter taste, the mechanism of short-term adaptation resides at a very early stage in the receptor transduction cascade.

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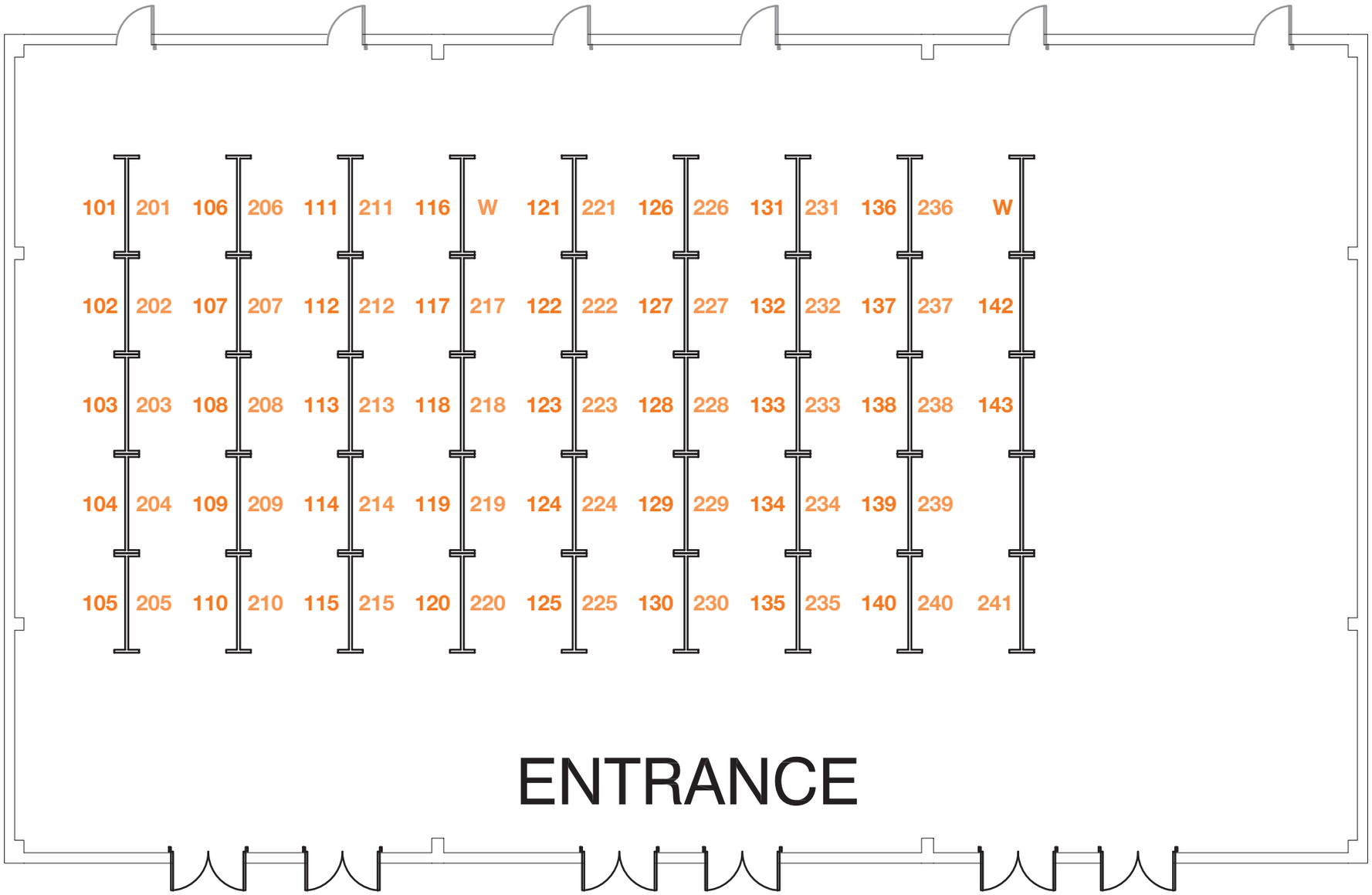
POSTER HALL THURSDAY

ESTERO BALLROOM

100's
Poster Session I: 8 - 10:30 am

200's
Poster Session II: 8 - 10:30 pm



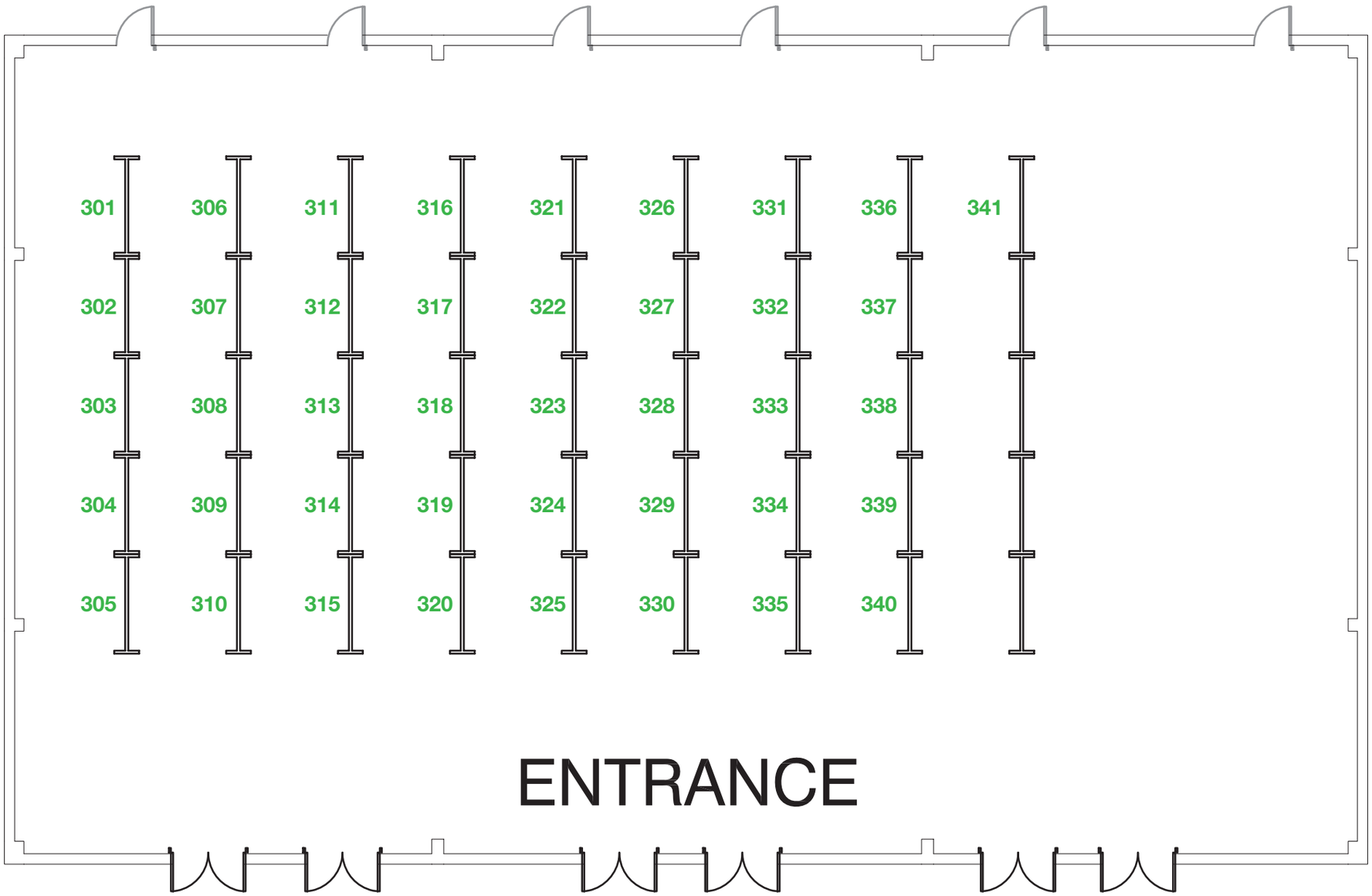
POSTER HALL FRIDAY

ESTERO BALLROOM

300's

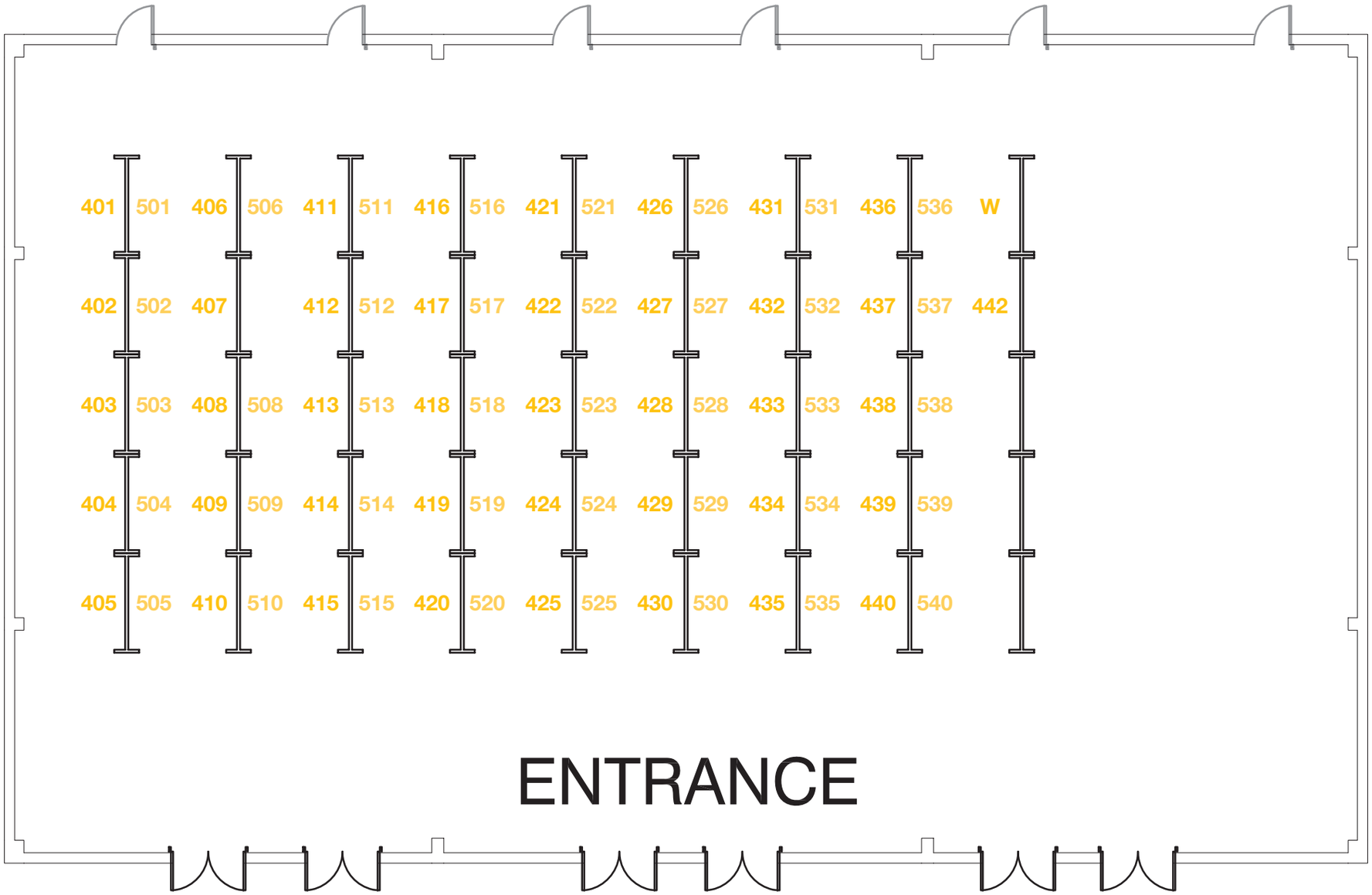
Poster Session III: 8 - 10:30 am

No PM session



POSTER HALL SATURDAY

ESTERO BALLROOM

400's
Poster Session IV: 8 - 10:30 am

500's
Poster Session V: 8 - 10:30 pm