XXVIth Annual Meeting of the European Chemoreception Research Organization

7-10 September 2016, Athens – Greece

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Welcome to the ECRO2016 Meeting

It is our privilege and great pleasure to welcome you to the 26th Congress of the European Chemoreception Organization in Athens, Greece, a most famous city since antiquity. The Greek capitol derived its name from the Olympian virgin goddess of wisdom, Athena, who originated as an incarnation of mind fully grown-up and dressed in her armor from Zeus' forehead. Under the aegis of Athena arts and science blossomed out. Athena became the patron of the then nameless city when she entered a contest together with Poseidon, the second mighty god after godfather Zeus. The two competed for the honor of becoming the city's name-giving patron god by offering their gifts to the residents. Poseidon struck the earth with his trident and created a spring, the symbol of fertility and life *per se*. Yet the population preferred the present of Athena, an olive tree, symbol of prosperity and peace and decided to make her the patron goddess. For centuries the goddess held her protective hand over the city. Athens' residents thanked her for the rescue from the Persians after the last Persian war by building and dedicating her the Parthenon temple at the Acropolis citadel. The city continuously exists since the Neolithic and became the largest Greek city state during the archaic period. Attic Greek culture expanded through large parts of Europe and the Middle East during Hellenism and, with the adoption of Hellenic traditions, by the Roman Empire. In 1985 Athens became the first European Cultural Capital and in 1987 the Acropolis UNESCO world cultural heritage. The meeting venue is in short walking distance to the Acropolis. Holding this year's conference close to the sanctum of Athena, the goddess of witness and patron of science, and perhaps under her umbrella is much valued in difficult and uncertain times.

The organisation of the Meeting this year was a team work. It relied a lot to Marika Kapsimali who wished to bring the eager of ancient Greeks for constant learning, improving and discussing fundamental scientific questions inspired by the events of everyday life. It was warmly supported by the ECRO Board who advised and contributed to the successful organisation of the meeting. Local companies assisted Marika to deal with the practical aspects of the Meeting.

Like last year, the program format includes key note lectures, plenary symposia, parallel symposia and poster sessions ensuring that researchers have numerous options to

present their research and learn the latest developments in the field. Prominent leaders in their fields accepted Marika's invitations to deliver the key note lectures or serve as symposia organizers and speakers. The team set up the program to represent topical interests and to be balanced across senses, models, concepts and methods. Based on the excellent experience we made last year in Istanbul we maintained the young scientist symposium which will be chaired by Stefan Fuss and Peter Mombaerts. The scientific program is complemented by a social program that includes a gala dinner excursion on the last day.

ECRO continues to support students and young researchers to present their work. This is visible by the program format giving them the opportunity to present their work but also by financial support in form of travel grants to young investigators. We acknowledge the continuous support by the Polack Foundation that allowed ECRO to honor a number of young scientists also in this year.

The ECRO Congress would of course not be possible without the contributions from our sponsors, i.e., Priority Program 1392 "Integrative analysis of olfaction" of the German Research Foundation, Sarstedt, and Aurora Scientific.

We hope to see you soon in Athens and wish you an exciting, successful and pleasant meeting that provides you with new impressions and ideas that will stimulate your future research.

Marika Kapsimali Wolfgang Meyerhof

European Chemoreception Research Organization

Πάντες ἄνθρωποι τοῦ εἰδέναι ὀρέγονται φύσει. All people, by nature, desire knowledge.
Μεταφυσικά - Βιβλίο Α :Αριστοτέλης. *Metaphysics – Book A: Aristotle.*

ECRO2016 Scientific Organising Committee

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ECRO2016 Practical Organisation

Marika Kapsimali
http://www.ibens.ens.fr/spip.php?article211&lang=en

Symvoli Conference and Cultural Management
www.symvoli.gr ecro2016@symvoli.gr

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Projector Company Events and Congresses technical support
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ECRO travel awards for the ECRO2016 meeting

BANNER Amir
BENDAS Johanna
de GROOT Jasper
HAN Pengfei
JUAN-CORDOBA Beatriz
KOCAGÖZ Yigit
LI Qian
PORADA Danja
RICATTI Maria Jimena
ROBERT-HAZOTTE Aline
SHARMA Kanika
STEIN Benjamin
TSITOURA Panagiota

ECRO BOARD 2016

President: Wolfgang MEYERHOF
Past President: Anna MENINI
Future President: Peter BRENNAN
Treasurer/ Executive Secretary: Krishna PERSAUD
General Secretary: Didier TROTIER
Members: Marika KAPSIMALI, Stefan FUSS, Teun DEKKER

Join the General Assembly on Friday to learn about the new Board elections and how you can contribute to ECRO !

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European Chemoreception Research Organization

CONFERENCE VENUE

The XXVIth Annual Meeting of the European Chemoreception Research Organization 2016 will be held at Divani Palace Acropolis Hotel in Athens.

DIVANI PALACE ACROPOLIS
ATHENS

Within the historical area of Athens, The Divani Palace Acropolis is in walking distance from the Acropolis, the Acropolis museum, the ancient theatres of Herodes Atticus and Dionysos, Philopappou Hill, the Arch of Hadrian, the Temple of Olympios Zeus and Plaka (the oldest section of Athens and the most popular and picturesque area in Athens). The Divani Palace Acropolis provides easy access to all areas both within and outside the city centre as it is very close to Athens Metro "Acropolis" station (200m).

http://divaniacropolishotel.com/
Parthenonos 19-25
11742 Athens - Greece
Tel: +30210 9280100
Fax: +30210 9214993
info@divaniacropolis.gr

AT THE VENUE . . .

CONFERENCE SECRETARIAT

Conference secretariat will be at the coffee break area in the ground floor of the Divani Palace Acropolis, the following hours:

Wednesday	07/09/2016	16:00 - 19:30
Thursday	08/09/2016	08:30- 19:00
Friday	09/09/2016	08:30- 19:30
Saturday	10/09/2016	08:30- 16:00

You can also contact the secretariat at:
T: 0030 6937 327775 | *E-mail :* ecro2016@symvoli.gr

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Βενιζέλου 3, Στοά Λεβή, 546 24 Θεσσαλονίκη
3, Venizelou str., Levi Stoa, GR-546 24 Thessaloniki

www.symvoli.gr

The MEETING

Lectures: Erechtio (mezzanine), Aristotelis A (ground floor)

Posters, Sponsors: Aristotelis B, ground floor

Coffee and lunch breaks: Ground floor (reserved lobby, outdoors around the swimming pool, next to the Posters, next to the Secretariat)

Internet access
High – speed wireless internet is available in all areas of the conference venue. Please contact the conference secretariat for your personal access code.

GALA DINNER

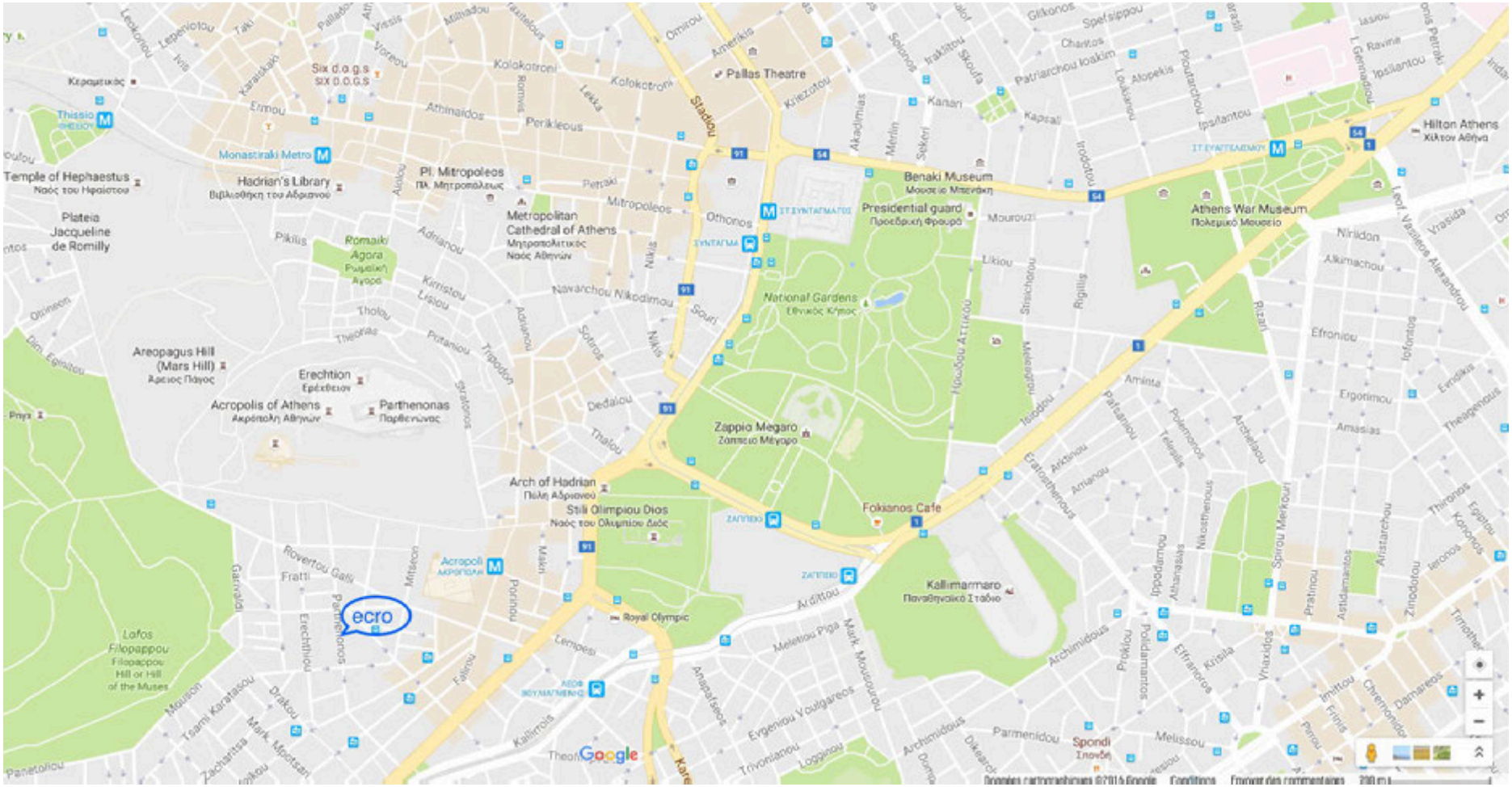
Date: Saturday 10th September 2016

Cape Sounion is the site of an ancient Greek temple of Poseidon, the god of the sea in classical mythology. The remains are perched on the headland, surrounded on three sides by the sea. Enjoy the sunset over the Aegean Sea, as viewed from the ruins, a sought-after spectacle.

** Note:the time of departure may be modified a little, according to the scientific program …*

16:15	departure from Divani Palace Acropolis
18:00	arrival & guided tour
19:40	view of the exceptional sunset
20:00	going for dinner
23:00	return to Divani Palace Acropolis

GETTING AROUND! Metro, Venue location


Pallas Theatre
Six d.o.g.s
Monastiraki Metro
Temple of Hephaestus
Hadrian's Library
Pl. Mitropoleos
Metropolitan Cathedral of Athens
Plateia Jacqueline de Romilly
Romaiki Agora
Areopagus Hill (Mars Hill)
Erechtion
Acropolis of Athens
Parthenonas
Arch of Hadrian
Stili Olimpiou Dios
Zappio Megaro
National Gardens
Benaki Museum
Presidential guard
Athens War Museum
Hilton Athens
Fokionos Cafe
Kallimarmaro
Royal Olympic
Lofos Filopappou
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GETTING AROUND!

The Venue Divani Palace Acropolis (Parthenonos Street, 19-25) is located
- 1.5 km from Athens central **Syntagma** Square
- approximately 35km from the international airport and 10km from the Piraeus port.
Closest Metro : "Acropolis" station (500m).

From/To the Athens AIrport

Distance: approx. 35 km

- **Taxi:** A taxi from the airport to the city center costs a flat rate of €38 from 5:00 a.m. to midnight, and €50 from midnight to 5:00 a.m. To confirm with the driver before starting the journey and ask a receipt.

- **Metro:** Take Metro Line 3 (Blue line / Airport –> Douk. Plakentias –> Aghia Marina), which connects the Athens airport to the city center. Trains run every 30 minutes, 7 days a week from 6:30 a.m. to 11:30 p.m. The trip from the Airport to 'Syntagma' station (Athens center) is 40 minutes. Then, to go to the Venue: change from the blue Line 3 at station 'Syntagma' to the red Line 2 (direction Eliniko) and get off at the station 'Acropolis'.
  Ticket costs 10 euros, valid for 70 minutes after validation.

From/To Syntagma (Central Athens)

- **Walking:** 20min approx, 1.5km to Syntagma. At walking distance from the Venue (less than 1km, 10 min) : the Acropolis, the Acropolis museum, the ancient theatres of Herodes Atticus and Dionysos, Philopappou Hill, the Arch of Hadrian, the Temple of Olympios Zeus, Plaka.
- **By taxi** : Venue - Syntagma square by taxi about 8 min. Taxi station at Syntagma.
- **By metro** : Venue-closest metro station: Acropolis (approx 6 min walk). Direct connection to station Syntagma (red line, M2). More information about Metro at: www.stasy.gr Metro tickets cost 1.40€ for 90 min one way, 4.5€ for 24 hour ticket, 9€ for 5 days and 22€ for 3day tourist ticket
- The Venue offers free transportation to Syntagma with complimentary shuttle bus service. For the exact timetable please ask the concierge.

Welcome to Athens!

A little guide

Athens is the historical capital of Europe, with a long history, dating from the first settlement in the Neolithic age. In the 5th Century BC (the “Golden Age of Pericles”) – the culmination of Athens’ long, fascinating history – the city’s values and civilization acquired a universal significance. Over the years, a multitude of conquerors occupied Athens, and erected unique, splendid monuments - a rare historical palimpsest.

In 1834, it became the capital of the modern Greek state and in two centuries since it has become an attractive modern metropolis with unrivalled charm. Today it is the political, social, cultural, financial and commercial center of Greece.

Conference’s urban district

The ECRO2016 conference takes place in the area of the Acropolis, which combines numerous advantages rendering it an ideal spot for the venue selection.

Neighbouring areas of Plaka, Monastiraki and Thiseio remain some of the most enchanted and interesting places of Athens. They are located in the very heart of the city, close to Syntagma Square

The most important sightseeing monuments of the capital of Greece are to be found in this area: the Acropolis and the Ancient Forum, the New Acropolis Museum, Herod Atticus Theatre and the Dionysos Areopagitis pedestrian walk. They all preserve and highlight the grand classicality of Athens.

The area is also famous for its taverns and numerous small bars.

http://www.thisisathens.org/

http://www.athensconventionbureau.gr/

http://www.visitgreece.gr/en/main_cities/athens

Police :100, Duty Hospitals and Clinics:1434, Ambulance 166

• Syntagma Square
Syntagma (Constitution) Square is the hub of Athens, close to many sights and transport links. The square, named for the Greek constitution granted after a popular uprising in 1834, is dominated by the imposing neoclassical Old Royal Palace, which has housed the Greek Parliament since 1934. *[access from Venue: 1.5 km, a 20-minute walk, or metro M2 (10 minutes)]*

• Acropolis
This former citadel perched atop a sheer rock contains some architectural gems from ancient Greece, including the Parthenon and the temple to the goddess Athena, dating from the 5th century BC. This is one of the remaining treasures of Ancient Greece, simply a must during any visit to Athens. *[access from Venue - about 500 metres from the hotel, a 7-minute walk]*

• Herodium Theater
It was built in 161 AD by the Athenian magnate Herodes Atticus in memory of his wife, Aspasia Annia Regilla. It was originally a steep-sloped amphitheater with a three-story stone front wall and a wooden roof made of expensive, cedar of Lebanon timber. It was used as a venue for music concerts with a capacity of 5,000. It lasted intact until it was destroyed and turned into a ruin by the Heruli in 267 AD. The audience stands and the orchestra (stage) were restored using pentelic marble in the 1950s. Since then it has been the main venue of the Athens Festival, which runs from May through October each year, featuring a variety of acclaimed Greek as well as International performances *[access from Venue - only 250 metres from the hotel, a 4-minute walk].*

• Plaka
Plaka is the old historical neighbourhood of Athens, clustered around the northern and eastern slopes of the Acropolis, and incorporating beautiful neoclassical architecture. Most of the streets have been closed to automobile traffic. It is now an area of restaurants, Jewelry stores tourist shops, and cafes. Though it is quite commercialized it is still a neighborhood and arguably the nicest neighborhood in central Athens. *[access from Venue– 750metres, about a 10-minute walk]*

• Monastiraki Flea Market
The place to go on Sunday mornings, this colourful area offers all sorts of trinkets and perhaps some real finds. Start at Monastiraki Square and don't be afraid to cut down side streets away from the crowds. *[access from Venue: 1.4 km, about 18 minutes walking]*

Announcement of the 27th Annual Meeting of the European Chemoreception Research Organization

The 27th ECRO annual meeting will be held at the Wellcome Genome Campus Conference Centre, Cambridge, UK.

Organizers : Darren Logan, Greg Jefferis and Simone Weyand (contact : dl5@sanger.ac.uk)

Date : 2-5 September, 2017

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European Chemoreception Research Organization

PROGRAM

Linear B (Mycenes, 1500-1200 BC)

Wednesday 7th September 2016

16.00-19.30 Registration

19.30-22.00 Welcome Reception at Divani Acropolis Roof Garden

Thursday 8th September 2016

09.00 **Welcome to the ECRO2016 meeting**

09.10 PL1 ***Plenary Lecture 1*** ***(Erechtion)***

Mechanisms of olfactory receptor choice
LOMVARDAS Stavros
Dept. of Biochemistry and Molecular Biophysics, M.B. Zuckerman Mind Brain and Behavior Institute, Columbia University, New York, USA

10.00-11.00 ***Symposium 1A*** ***(Erechtion)***
Organizer: Stavros LOMVARDAS
Deciphering olfaction: from the nose to the brain

10.00 S1 A novel mechanism and logic for mammalian olfaction
DATTA Sandeep Robert
Department of Neurobiology, Harvard Medical School, USA

10.20 S2 Stability and plasticity in the Drosophila olfactory system across timescales
VOLKAN Pelin
Dept of Biology, Duke Institute for Brain Sciences, Duke University, USA

10.40 S3 Odorant Receptor Regulation of Gene Choice, Axon guidance and Ligand binding
FEINSTEIN Paul
Hunter College, City University of New York, USA

11.00-11.30 Coffee break

11.30-13.00 ***Symposium 1B*** ***(Erechtion)***
Organizer S. Lomvardas
Deciphering olfaction: from the nose to the brain

11.30 S4 Functional plasticity in the mouse olfactory bulb following motherhood
MIZRAHI Adi
Dept of Neurobiology, A Silberman Inst. of Life Sciences, The Hebrew University of Jerusalem, Israel

11.50 S5 Sensory and hormonal control of social behaviors
SHAH Nirao
Department of Anatomy, UCSF --- University of Stanford, USA

12.10 S6 Olfactory sensation promotes social determination in the mouse
STOWERS Lisa
Dept of Molecular and Cellular Neuroscience, The Scripps Research Institute, La Jolla, California, USA

12.30 S7 Neural Circuit Formation for Odor-Induced Innate Social Behaviors
SAKANO Hitoshi
Department of Brain Function, University of Fukui, School of Medicine, Japan

13.00-14.00 Lunch

13.00-15.00 POSTER SESSION

15.00 PL2 ***Plenary Lecture 2*** ***(Erechtion)***
Interrogating sweet taste cells
MARGOLSKEE Robert
Monell Chemical Senses Center, Philadelphia, USA

15.50-17.00 ***Parallel Symposium 2A*** ***(Erechtion)***
Organizer: Stavros LOMVARDAS
Deciphering olfaction: from the nose to the brain

15.50 S8 Chemoreceptors: from the immune to the vomeronasal system
RODRIGUEZ Ivan
Department of Genetics and Development, University of Geneva, Switzerland

16.10 S9 trans-Tango: Trans-synaptic Mapping and Manipulation of Neural Circuits
BARNEA Gilad
Dept. Neuroscience, Brown University, Providence, USA

16.30 S10 Carbon dioxide sensing in higher brain centers of Drosophila melanogaster
VASCONCELOS Maria Luisa
Champalimaud Research, Champalimaud Centre for the unknown, Lisbon, Portugal

17.00-17.30 Coffee Break

17.30-19.00 ***Parallel Symposium 2B*** ***(Erechtion)***
Organizer: Stavros LOMVARDAS
Deciphering olfaction: from the nose to the brain

17.30 S11 Representations of odor identity and odor intensity in piriform cortex
FRANKS Kevin
Dept of Neurobiology, Duke University School of Medicine, USA

17.50 S12 Cortical neural circuits for olfaction
FLEISCHMANN Alexander
CIRB, Collège de France, Paris, France

18.10 S13 Learning-related changes in Mitral and Tufted cell responses reflect changes in sniff behavior
SCHAEFER Andreas
The Francis Crick Institute, UK

18.30 S14 Phase dependent lateral inhibition in the olfactory bulb
HADDAD Rafi
Gonda Multidisciplinary Brain Research Center, Bar-Ilan University, Israel

15.50-17.00 ***Parallel Symposium 3A (Aristotelis)***
Organizers: Claire MURPHY, Annick FAURION, Eugeni ROURA, David VAL-LAILLET
Nutrional Chemosensing and food intake regulation

15.50 S15 Nutrient and non-nutrient sensing in the gastrointestinal tract
SHIRAZI-BEECHEY Soraya P
Institute of Integrative Biology, University of Liverpool, UK

16.10 S16 Amino acid sensing in the human gut: effect of obesity
DEPOORTERE Inge
Translational research in gastrointestinal disorders Unit, KU Leuven, Belgium

16.30 S17 fMRI activation during hunger and satiety in young and older individuals with metabolic syndrome
MURPHY Claire
San Diego State University, SDSU/UCSD Joint Doctoral Program and the University of California, San Diego, CA USA

17.00-17.30 Coffee Break

17.30-19.00 ***Parallel Symposium 3B (Aristotelis)***
Organizers: Claire MURPHY, Annick FAURION, Eugeni ROURA, David VAL-LAILLET
Nutrional Chemosensing and food intake regulation

17.30 S18 Maternal transfer of food cues
SOLA-ORIOL David
Animal Nutrition and Welfare Service, Dept of Animal and Food Sciences, Universitat Autònoma de Barcelona,Spain

17.50 S19 Oral and extraoral nutritional chemosensing in pigs and humans
ROURA Eugeni
Centre for Nutrition and Food Sciences, Queensland Alliance for Agriculture and Food innovation, The University of Queensland

18.10 S20 Effects of the Stevia-derived sweetener Rebaudioside A on the incretin *hormone* glucagon-like peptide-1
MEIJERINK Jocelijn
Division of Human Nutrition, Wageningen University, Wageningen, The Netherlands

18.30 S21 Imaging brain responses to oral and visceral food stimuli in the pig model
VAL-LAILLET David
INRA Saint-Gilles, France

20.30-23.00 Social event (Dinner, registration during the meeting)

Friday 9th September 2016

09.00 PL3 ***Plenary Lecture 3*** ***(Erechtion)***
Cellular and molecular basis for taste sensation in Drosophila
MONTELL Craig
Dept. of Molecular, Cellular, & Developmental Biology, University of California, Santa Barbara, USA

09.45-10.15 Coffee break

10.15-11.45 ***Symposium 4*** ***(Erechtion)***
Organizers: Maik BEHRENS, Wolfgang MEYERHOF
Orosensory perception beyond taste

10.15 S22 Some like it hot: TRPV1 receptor agonists in health and medicine
SOMOZA Veronika
Dept. of Nutritional and Physiological Chemistry, University of Vienna, Austria

10.35 S23 Pungent sensation through TRP channels in the oral cavity
TOMINAGA Makoto
Division of Cell Signaling, Okazaki Institute for Integrative Bioscience, Japan

10.55 S24 The complexity of orosensory lipid detection in humans
BEHRENS Maik
German Institute of Human Nutrition Potsdam-Rehbruecke, Dept. Molecular Genetics, Germany

11.15 S25 Beyond olfaction: other airway senses
LIBERLES Stephen
Harvard Medical School, Department of Cell Biology, Boston, MA, USA

11.45-12.30 ECRO General Assembly

12.30-13.30 Lunch

13.30-15.00 ***Parallel Symposium 5*** ***(Erechtion)***
Organizers: Marc SPEHR, Yoram BEN-SHAUL
Unique features of processing in the accessory olfactory bulb

13.30 S26 Interdependent conductances drive infra-slow intrinsic rhythmogenesis in a subset of AOB projection neurons
SPEHR Marc
Dept of Chemosensation, Institute for BiologyII, RWTH Aachen University, Germany

13.50 S27 Information Processing in the Accessory Olfactory Bulb
BEN-SHAUL Yoram
The Hebrew University Medical School, Jerusalem, Israel

14.10 S28 Inhibitory control of chemosignal processing in the accessory olfactory bulb
MEEKS Julian
Dept of Neuroscience, UT Southwestern Medical Center, Dallas, Texas, USA

14.30 S29 Persistent firing and synchronous infra-slow bursting in AOB mitral cells: implications for sensory information processing
WAGNER Shlomo
Sagol Department of Neurobiology, University of Haifa, Haifa, Israel

13.30-15.00 ***Parallel Symposium 6*** ***(Aristotelis)***
Organizer: Masha NIV
Variation in chemosensory receptors across individuals

13.30 S30 Variation in sequence, structure and ligands of chemosensory GPCRs
NIV Masha
Institute of Biochemistry, Food Science and Nutrition, The Hebrew University, Israel

13.50 S31 Omics Analyses of Human Olfactory Diversity
LANCET Doron
Weizmann Institute of Science, Israel

14.10 S32 Bittersweet human taste genetics
REED Danielle R
Monell Chemical Senses Center, Philadelphia, USA

14.30 S33 Bitter Taste Receptors in Asthma
DESHPANDE Deepak
Center for Translational Medicine, Thomas Jefferson University, Philadelphia, USA

15.00-15.30 Coffee Break

Friday 9th September 2016

15.30 PL4 ***Plenary Lecture 4*** ***(Erechtion)***

Molecular regulation of taste bud cell renewal

BARLOW Linda

Dept of Cell and Developmental Biology - The Rocky Mountain Taste and Smell Center, University of Colorado School of Medicine, USA

16.15-17.45 ***Parallel symposium 7*** ***(Erechtion)***

Organizers: Matthieu KELLER, Patricia NAGNAN-LE MEILLOUR

Olfactory regulation of reproductive behavior in mammals

16.15 S34 A multitask mammalian chemosignal and the definition of pheromones

SCHAAL Benoist

Centre des Sciences du Goût, CNRS-Université de Bourgogne, Dijon, France

16.35 S35 The multiple roles of MUPs and their volatile ligands in mouse sexual signaling

HURST Jane

Institute of Integrative Biology, University of Liverpool, Liverpool, UK

16.55 S36 A proposed mechanism for coding pheromones in pig

NAGNAN-LE MEILLOUR Patricia

Univ. Lille, CNRS UMR8576, INRA USC1409, UGSF, Unité de Glycobiologie Structurale et Fonctionnelle, Lille, France

17.15 S37 Olfactory communication and reproductive strategies in Old World primates

VAGLIO Stefano

University of Wolverhampton, UK

16.15-18.00 ***Parallel Symposium 8*** ***(Aristotelis)***

Organizer: ECRO2016 chair: Marika KAPSIMALI

16.15 S38 Functional role of the centrifugal feedback to the olfactory bulb: computational model and experimental data

ZHAOPING Li

Department of Computer Science, University College London, London, UK

16.30 S39 Neuropilin-1 and the positions of glomeruli in the mouse olfactory bulb

ZAPIEC Bolek

Max Planck Research Unit for Neurogenetics, Frankfurt, Germany

16.45 S40 Acute SSRI administration modulates taste sensitivity in untreated depressed human subjects

MELICHAR Jan

Ranvier Health Ltd, Bristol UK

17.00 S41 Molecular markers of GC-D-expressing and Grueneberg ganglion chemosensory neurons
MUNGER Steven D
Center for Smell and Taste and Department of Pharmacology & Therapeutics, University of Florida, Gainesville, USA

17.15 S42 Novel Ca2+-activated Cl- Channel of the chemosensory cilia of rat olfactory sensory neurons
BACIGALUPO Juan
Universidad de Chile, Santiago, Chile

17.30 S43 Odorant reception in the diamondback moth Plutella xylostella
WANG Guirong
State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing,China.

17.45 S44 New insights in vertebrate « biotransfolfaction »
HEYDEL Jean-Marie
Centre des Sciences du Goût et de l'Alimentation, UMR 6265 CNRS / 1324 INRA / Université de Bourgogne Franche-Comté, Dijon, France.

18.00-19.30 Light drink

18.00-19.30 POSTER SESSION

20.30-23.00 Social event (Dinner, registration during the meeting)

Saturday 10th September 2016

09.00 PL5 ***Plenary Lecture 5*** ***(Erechtion)***
Sour: More than a Primary Taste
FINGER Thomas
Rocky Mountain Taste & Smell Center, Univ. Colorado School of Medicine, USA

09.45-10.15 Coffee break

10.15-12.45 ***Parallel Symposium 9*** ***(Erechtion)***
Organizers: Jonas OLOFFSON, Jessica FREIHERR
Trends in human chemosensation: olfactory enhancement and Chemosignalling

10.15 **Introduction** by Jonas Oloffson, Jessica Freiherr

10.25 S45 Olfactory training and recovery from smell loss
HUMMEL Thomas
Smell & Taste Clinic, Department of ORL, TU Dresden, Dresden, Germany

10.40 S46 Olfactory experts: Differences in neuroimaging and behavioral assessments
PLAILLY Jane
Centre de Recherche Neurosciences, Lyon, France

10.55 S47 Odor memory training and transfer of learning
OLOFFSON Jonas
Dept of Psychology, Stockholm University, Sweden

11.10 S48 Olfactory neuroplasticity
HOLBROOK Eric
Harvard Medical School, Boston, USA

11.25 **Discussion**

11.35 S49 On the mechanisms of fear chemosignaling: A meta-analysis
DE GROOT Jasper
Faculty of Social and Behavioral Sciences, Department of Social, Health, and Organizational Psychology, Utrecht University, Utrecht, The Netherlands

11.50 S50 Chemosensory danger detection in the human brain
MUTIC Smiljana**, FREIHERR** Jessica
Diagnostic and Interventional Neuroradiology, RWTH Aachen University, Aachen, Germany

12.05 S51 The role of human body odors in ingroup-outgroup relationships
PARMA Valentina
International School for Advanced Studies (SISSA), Trieste, Italy

12.20 S52 The impact of androstadienone on the multifaceted stress response in females and males
DERNTL Birgit
Dept of Psychiatry and Psychotherapy, University of Tübingen, Tübingen, Germany

12.35 **Discussion**

10.15-12.45 ***Parallel Symposium 10*** ***(Aristotelis)***
Organizer: ECRO2016 chair: Krishna PERSAUD

10.15 S53 Functional metabolomics using a microfluidic chemoreceptor cell assay
ROELSE Margriet
Wageningen University and Research, Wageningen, The Netherlands

10.30 S54 A novel functional screening assay to monitor sweet taste receptor activation in vitro
BASTIAAN-NET Shanna
Food and Biobased Research, Wageningen University and Research Centre, The Netherlands

10.45 S55 Biosensors based on immobilised Major Urinary Proteins from the mouse
PERSAUD Krishna
The University of Manchester, School of Chemical Engineering and Analytical Science, UK

11.00 S56 Multiple receptors contribute to the transduction of fat taste
DAMAK Sami
Nestlé Research Center, Vers-chez-les-Blanc, 1000 Lausanne, Switzerland

11.15 S57 CD36 is involved in fatty acid detection in the murine olfactory system
NEUHAUS Eva
Friedrich-Schiller University Jena, Germany

11.30 S58 Neural mechanisms underlying the ageing-associated decline in chemosensory perception
HUSSAIN Ashiq
Max Planck Institute for Neurobiology, Martinsried, Germany

11.45 S59 Oral and intestinal sweet taste T1R2/R3 receptors in mice; effect on consumption, over weight, blood glucose and insulin levels
HELLEKANT Göran
Dept of Biomedical Sciences, Medical School, University of Minnesota, Duluth, USA

12.00 S60 Rise of the urethral brush cells
DECKMANN Klaus
Institute for Anatomy and Cell Biology, Justus-Liebig-University, Giessen, Germany

12.15 S61 Specificity versus Promiscuity: The Ligand-binding Pocket for Bacterial Signal Peptides in Formyl Peptide Receptors
BUFE Bernd
Center for Integrative Physiology and Molecular Medicine, Saarland University, Homburg, Germany

12.45-14.00 Lunch

14.00-15.30 ***Young scientists - Symposium 11*** ***(Erechtion)***
Organizers: Peter MOMBAERTS, Stefan FUSS

14.00 S62 A calcium signaling 'fingerprint' in vomeronasal sensory neurons
NAGEL Maximilian
RWTH Aachen University, Dept. of Chemosensation, Aachen, Germany

14.15 S63 Olfactory sensory neurons transiently express multiple olfactory receptors during development
LI Qian
Dept of Cell Biology, Harvard Medical School, Boston, MA, USA

14.30 S64 Effect of aging in the activity of the posterior piriform cortex of rats during flavor recognition memory
GRAU-PERALES Alejandro
Dept of Psychobiology. Institute of Neurosciences. Center for Biomedical Research. University of Granada. Spain.

14.45 S65 Automated operant olfactory conditioning of group-housed mice
REINERT Janine
Heidelberg University, Institute for Anatomy and Cell Biology, Functional Neuroanatomy Department, Heidelberg, Germany

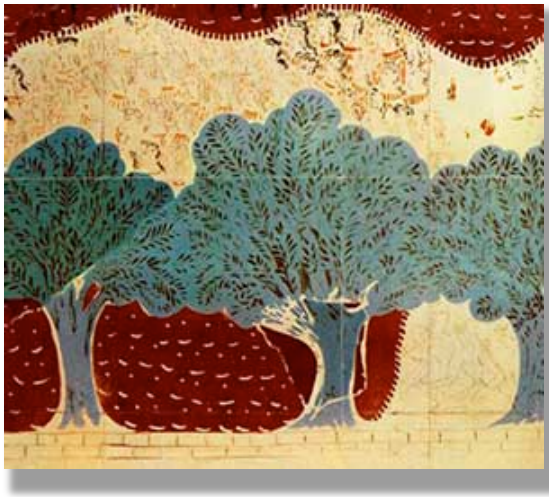
15.00 S66 Odor threshold relates to sexual pleasure
BENDAS Johanna
Dept for Psychotherapy and Psychosomatic Medicine, Dresden University Hospital, Technische Universität Dresden, Germany

15.15 S67 The Effect of Contextual Odors on Emotional Evaluations of Facial Expressions
SYRJÄNEN Elmeri
Stockholm University, Gösta Ekman Lab, Sweden

15.30-16.00 Poster Prize - Closing Remarks

16.00-16.30 Coffee Break

16.45-23.00 Gala excursion at Sounio and dinner (check for the exact departure time)



(Knossos, Crete, 1630-1500 BC)

ORAL PRESENTATIONS

S1
A Novel Mechanism and Logic for Mammalian Olfaction

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The Datta lab studies how information from the outside world is detected, encoded in the brain, and transformed into meaningful behavioral outputs. We address this fundamental problem by characterizing the olfactory system, the sensory system used by most animals to interact with their environment. Here we discuss recent results relevant to understanding sensorimotor coupling — the process of linking sensation to action — in the olfactory system. In particular, we describe a novel molecular mechanism and neural logic that underlies odor perception, one that may be specialized for the detecting and processing of odors with innate meaning. This mechanism defines a new mode of sensory encoding in mammals, and may be relevant to odor perception across deuterostomal lineages, including humans.

S2
Stability and plasticity in the *Drosophila* olfactory system across timescales

Jia W. Pan[1], Scott Barish[1], Tristan Li[1,*], Catherine Hueston[1], Doug Olsen[1], Songhui Zhao[1], Sayan Mukherjee[2], Corbin D. Jones[3], Pelin C. Volkan[1,4]
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[1] Duke University, Department of Biology, [2] Duke University, Department of Statistical Science, Computer Science and Mathematics; [3] University of North Carolina, Chapel Hill, Department of Biology & Carolina Center for Genome Studies; [4] Duke Institute for Brain Sciences; [*] Current Address: Stanford University, Department of Neurobiology.

Most organismal phenotypes result from a trade-off between genetically hardwired genetic and developmental programs to invariably build working cellular systems, and more plastic and variable programs that enable adaptability. In fast-evolving and complex neural systems like the olfactory system, some neuronal lineages and their developmental programs may be more stable to conserve important behavioral functions; others may be more variable to enable better behavioral adaptability to the environment. We pursue this question in both organismal and evolutionary timescales. In the evolutionary timescale, using transcriptional profiling of olfactory system development from 6 Drosophila species, we find that olfactory receptor gene expression, as an output of ORN lineages, and their developmental programs appear strikingly stable across the 6 species. However, specific OR genes normally expressed in large and thin basiconic sensilla ORNs and their combinatorial developmental programs show high variation across evolution. Interestingly, some of these OR genes in large basiconic sensilla neurons are involved in chemosensory behavioral adaptation and show transcriptional, developmental and anatomical convergence in species with host plant specializations. The combinatorial transcription factor code for large and thin basiconic ORN development, which normally show high levels of transcriptional variation across 6 species, also shows convergent transcriptional configuration in feeding specialist. These results identify the large and thin basiconic neuronal lineages and their developmental transcriptional programs as an evolutionary hotspot across *Drosophila* species. Convergence of these transcriptional programs in species with specialized chemosensory adaptations suggests surprising levels of evolutionary constraints at the levels of transcription in the olfactory system.

We observe a very similar compartmentalization of stability and plasticity at the circuit level in each organism. For this, we make use of the ORNs that regulate courtship behaviors in *Drosophila*. Three classes of ORNs (Or67d, Or47b and Ir84a) were shown to be involved in courtship behaviors. Or67d functions to drive innate aspects of courtship, whereas Or47b and Ir84a have more modulatory functions on courtship behaviors. All three ORN classes express the key behavioral switch gene and a molecular marker of courtship circuits in *Drosophila*, *fruitless.* We found that developmentally hardwired programs coordinate *fruitless* expression in these ORNs with appropriate olfactory receptors. However, once the flies emerge, maintenance of *fruitless* expression in Or47b and Ir84a ORNs, but not

Or67d ORNs, require olfactory receptor signaling and histone acetyl transferase p300, in an age-dependent manner. Our results highlight ORN class-specific functional differences in *fru* regulation that rely either on stable or plastic mechanisms driven by olfaction. Such dual mode of *fru* regulation in ORNs might be a trait of neurons driving innate and plastic aspects of sex-specific behaviors, respectively.

As a whole, our findings highlight neural diversity as a facilitator to compartmentalize stability and plasticity of transcriptional programs underlying olfactory system function. This allows for stable transcriptional programs to invariably build neuronal lineages and circuits with innate or critical behavioral functions, as well as variable transcriptional programs for neuronal components that enable odor-guided behavioral adaptations to changing environments.

S3
Odorant Receptor Regulation of Gene Choice, Axon guidance, and Ligand binding

Charlotte D'Hulst, Kia Movahedi, Xavier Grosmaitre and Paul Feinstein
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The mouse olfactory system owes its architecture through the regulation and function of odorant receptors (ORs). Initially, ORs are activated in a singular gene choice fashion such that each of the thousands of OR alleles are expressed in a small proportion of neurons. This equal representation of like ORs results in their axons coalescing into homotypic glomeruli. Thus, a low and relative equal probability of choice, sets the system in motion for turning on an OR allele, then the OR protein determines the odorant profile and axon identity of the neuron. We have put forth new principles for the how the olfactory system is organized and utilized through a series of genetic experiments. Using multimers of a gene choice enhancer (the MouSensor Technology), we can alter the representation of any cloned OR. We find that the number of OSNs expressing a given OR determines the threshold towards ligands both in olfactory bulbs and in behavioral experiments (D'Hulst et al, 2016). In a separate series of experiments we have modified cAMP levels generated by OR activation and find that these levels are not required for the regulation of first choice or axonal identity (Movahedi et al., 2016). Taken together our data strongly argue that many developmental functions of the OR are carried out in a G-protein/cAMP independent manner as we originally proposed (Feinstein and Mombaerts, 2004). In addition, our MouSensor Technology will provide a platform to finally crack the human olfactory code.

S6
Olfactory sensation promotes social determination in the mouse

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How individuals are motivated to either strive for social dominance or avoid risk by capitulating to subordinance is largely unknown. Here we utilized an unenforced tube test and studied the effects of olfactory sensation to promote social behavior. In this setting, our study indicates that olfactory cues are essential not to release a specific behavior, but to generate grit and persistence to endeavor in social engagement. We find that variance in social persistence underlies the formation of dominance hierarchies. We additionally find that individual odor scents evoking experience-dependent memories do not serve as a mechanism to generate behavioral differences across the hierarchy. Instead olfactory cues act differentially on high and low ranked individuals to promote action. The unenforced tube test provides a robust platform to begin to identify the mechanisms underlying resilience and determination.

S7
Neural Circuit Formation for Odor-Induced Innate Social Behaviors

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In mammals, neural circuits are formed based on a genetic program and further refined by neuronal activity. Here we report that in the mouse olfactory system, the glomerular map is not merely refined but newly connected to second-order neurons in an activity-dependent manner. It was found that Sema7A/PlxnC1 interaction within glomeruli is key for inducing activity-dependent olfactory synapse formation during the neonatal period. Postnatal blockage of Sema7A signaling perturbs odor perception and some social behaviors in adulthood. Our results provide the molecular basis for the critical period of the mouse olfactory system.

We also studied neural circuit formation for social attraction behaviors in the mouse main olfactory system. It was found that expression of a single axon guidance molecule, Nrp2, in a subset of mitral cells is sufficient to instruct their neural circuit formation from the posteroventral main olfactory bulb (MOB) to the anterior medial amygdala (MeA). By using repulsive interactions with Sema3F, $Nrp2^{+}$ mitral cells are guided to the posteroventral MOB to receive attractive social signals and send their axons to the anterior MeA to elicit attractive behavioral responses. Gene-targeting experiments demonstrate that neural circuit formation of $Nrp2^{+}$ mitral cells and odor-induced social attraction behaviors are impaired in the mitral-cell-specific Nrp2 knockout.

PL2
Interrogating sweet taste cells

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Knockout mice lacking the sweet taste receptor subunit Tas1r3 lose behavioral and nerve responses to non-caloric sweeteners but retain responses to glucose and other sugars. This suggests the presence of a Tas1r3-independent mechanism to detect the sweetness of sugars. We used PCR, in situ hybridization and immunohistochemistry to identify signal transduction elements selectively expressed in sweet-responsive taste cells that might mediate Tas1r3-independent responses to sugars. Glucose transporters (GLUTs), Na-dependent glucose co-transporter-1 (SGLT1) and ATP-gated K^{+} channels (K_{ATP}) are all present in Tas1r3+ taste cells and may constitute a Tas1r3-independent means to detect caloric sugars. In addition, Tas1r3+ taste cells selectively express disaccharidase enzymes sucrase, maltase, trehalase and lactase. In both wildtype and Tas1r3 knockout mice disaccharidase inhibitors significantly reduced gustatory nerve responses to the disaccharides sucrose and maltose, but not to the monosaccharides glucose and fructose or the noncaloric sweeteners. It appears that these orally expressed enzymes act in concert with salivary amylase to generate free glucose from starch, sucrose, maltose, trehalose and lactose that can activate the Tas1r3-independent sugar detection pathway. To identify additional signalling components and regulatory factors selectively expressed in Tas1r3+ taste cells we used single taste cell RNA-Seq (deep sequencing) and bioinformatics to “data-mine” the Tas1r3+ taste cell “transcriptome” (i.e. all genes transcribed in Tas1r3+ taste cells). Gli3, a key transcriptional effector in the sonic hedgehog signalling pathway, was found to be selectively expressed in Tas1r3+ taste cells and Lgr-5+ taste stem cells, but not in type I or type III taste cells. Gli3 conditional knockout mice were more sensitive to sweet, umami and bitter compounds and had increased numbers of type II taste cells. Our results suggest that Gli3 is an important negative regulator of taste bud maintenance that enhances the number of type II taste cells, including those responsive to sweet.

S10
Carbon dioxide sensing in higher brain centers of *Drosophila melanogaster*

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Carbon dioxide generates a strong avoidance response in *Drosophila melanogaster* that at the first olfactory center is processed by a single channel, the V glomerulus. Projection neurons that innervate the V glomerulus make direct connections to the Lateral Horn (LH). It is thought that the innate olfactory responses are processed at the LH however the LH neurons are very poorly described. We did a neuronal silencing screen for LH neurons that are required for CO2 avoidance. We identified two positive lines. Interestingly, the neurons in each line are quite distinct in terms of the areas that they innervate LH and where they project in the brain. Further behavioral testing of the two lines revealed specificity to carbon dioxide response. We are now imaging the neurons to assess their response profile to different odorants. Our findings suggest that there are two sets of neurons dedicated to CO2 processing at the LH that output to different brain areas possibly contributing to different aspects of the behavioral response.

S11
Representations of odor identity and odor intensity in piriform cortex

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The ability to represent both the identity and intensity of an odor are fundamental for olfactory perception and odor-driven behaviors. To determine how these distinct odor features are represented in olfactory (piriform) cortex we recorded odor-evoked spiking activity in large populations of piriform neurons in awake mice. We find distinct coding strategies facilitate non-interfering representations of odor identity and intensity. Odor identity can be accurately decoded using population spike counts. By contrast, intensity is encoded, in part, with spike timing. We find subsets of cells have brief, concentration-insensitive responses, providing a rapid identity code, while others occur later with concentration-dependent response latencies. We also find functionally distinct subpopulations of 'on' and 'off' neurons that preferentially represent different features of the odor. Together, our data support a multiplexed spatial-identity/temporal-intensity cortical odor code in which different subpopulations of neurons represent odor features that may be most salient for their specific downstream targets.

S12
Cortical neural circuits for olfaction

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The olfactory (piriform) cortex is the main target of olfactory bulb mitral and tufted cells and plays key functions in odor perception and memory. To understand principles of odor coding in piriform neural networks we have recorded, using *in vivo* two-photon calcium imaging in anesthetized mice, odor-evoked activity from large ensembles of piriform neurons. We find that odor identity - at a given odor intensity - can accurately be decoded from spatially distributed ensembles of piriform neurons. However, piriform response patterns change substantially over a 100-fold change in odor concentration, apparently degrading information about odor identity. We show that this problem can be resolved by decoding odor identity from a subpopulation of concentration-invariant piriform neurons. We propose that distinct perceptual features of odors are encoded in independent subnetworks of neurons in the olfactory cortex.

S13
Learning-related changes in Mitral and Tufted cell responses reflect changes in sniff behaviour

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Sensory circuit activity has often revealed correlates of information about behavioral context. Such representations could arise either from physiological changes to the network, or from changes in stimulus sampling behaviour. The olfactory bulb (OB) is ideally suited to studying this, with both inputs from higher centers and clear coupling of activity to the sniff cycle. Using whole cell recordings in the OB of passive and behaving mice, we identified learning-related changes in mitral and tufted cell odor responses. Sniffing behavior also underwent parallel changes, which correlated with the changes in odor response. In absence of learning, sniffing could alter both baseline activity and odor responses. Learning-related changes occurred prior to both the typical reaction time and earliest estimates of decision time (a single sniff cycle), while sniff changes correlated with the motivational state of the animal. Therefore, highly motivated mice may modulate their sampling strategy, altering odor representations to facilitate decision. Thus, contextual representations in sensory circuits can be explained by alteration in stimulus sampling behavior.

S14
Phase dependent lateral inhibition in the olfactory bulb

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In the olfactory bulb (OB) the mitral/tufted cells (M/T), receive lateral inhibition from surrounding neurons. However, the spatial organization, density, and strength of this lateral inhibition have not been elucidated. In addition, how different sub classes of M/T cells differ in the amount of their inhibitory input is not known. Furthermore, how this spatial organization improves odor representation without causing a reduction in odor sensitivity is unclear. To characterize lateral inhibition in the mouse OB we recorded M/T cell responses while optogenetically stimulating surrounding glomeruli expressing channelrhodopsin2 in either their olfactory sensory neurons or M/T cells. We found that inhibition strength depended on the preferred phase of the MT cell: Neurons with preferred phase at the second half of the sniff cycle receive only high level of surround inhibition while M/T neurons with preferred phase at the first half of the sniff cycle receive intermingled level of surround inhibition. Furthermore, inhibition and excitation strength depended on the neuron phase and could shift the neuron preferred phase. We also found that proximal neurons receive different levels of inhibition strength. Interestingly, sister M/T cells received relatively similar amount of inhibition strength but from different glomeruli. Plugging these findings into a simple computational model of the olfactory bulb demonstrates that the amount of inhibition and connectivity revealed here are well positioned to decorrelate odor responses while minimizing the reduction in odor sensitivity.

S15
Nutrient and non-nutrient sensing in the gastrointestinal tract

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The intestinal epithelium is a major interface with the outside world. This interface is separated from the body's internal milieu by a single layer of epithelial cells consisting of absorptive enterocytes, goblet, enteroendocrine and Paneth cells. These cells are exposed, at the luminal domain, to an external environment that is continuously changing by types and amounts of nutrients, microorganisms, microbial products, gastrointestinal secretions and potentially toxic chemicals. The intestinal epithelium constantly monitors the composition of its contents in order to optimize nutrient absorption, as well as defending threats to its integrity.

In recent years significant advances have been made in the understanding of the molecular recognition events involved in sensing the luminal contents of the gastrointestinal tract. The sensing of various nutrients in the gastrointestinal tract is accomplished by a number of G-protein coupled receptors, expressed on the luminal membrane of enteroendocrine cells. Sensing of nutrients by these receptors leads to secretion of hormones that control vital physiological functions such as food intake, nutrient digestion and absorption, intestinal barrier function and insulin secretion.
The intestine also contains approximately 1000 different species of bacteria and has to discriminate between pathogenic and commensal bacteria in order to maintain a balance between immune protection and inflammatory over-reactions. A class of proteins known as pattern recognition receptors, in particular toll-like receptors (TLRs 1-10) play a key role in the recognition of microbes via detection of conserved molecular features.
The sensory receptors that face the lumen of the intestine and are responsive to luminal contents provide a unique therapeutic opportunity.
In my talk I will present data on the role of the gut expressed taste 1 receptor (T1R) family in intestinal nutrient sensing and the contribution of TLR9-recognition of bacteria in control of gut hormone release. The impact of these findings in respect to health maintenance and disease prevention will be discussed.

S17
fMRI activation during hunger and satiety in young and older individuals with metabolic syndrome

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Age effects on the human taste system are evident at both peripheral and central levels. Metabolic syndrome is a constellation of risk factors (e.g., abdominal obesity, hypertension) that commonly occur together, increase with age, and heighten the risk for cardiovascular disease, type II diabetes and cognitive decline in old age. Little is known about how age, metabolic syndrome and hunger state interact to influence how the brain processes information about taste, a powerful motivator of consumption. Thus, here we aimed to investigate differences in functional MRI of brain response during the hedonic evaluation of a pleasant, nutritive stimulus (sucrose) within regions critical for taste, homeostatic energy regulation, and reward as a function of the interactions among age, metabolic syndrome, and hunger condition. We scanned young and elderly adults, with and without metabolic syndrome, at 3T twice: once fasted over night and once after a preload. Data were analyzed using voxel-wise 3dMVM (multivariate modeling) in AFNI, and a region of interest analysis in key regions associated with taste, homeostasis, and reward processing. Results indicated significant effects of age as well as interactions between 1) age and metabolic syndrome, 2) age and hunger condition, and 3) hunger condition and metabolic syndrome. The present

findings indicate that metabolic syndrome influences functional activation and that there are significant age related differences in activation that are dependent on the hunger state in regions critical for 1) homoeostatic energy regulation, 2) basic as well as higher-order sensory processing and integration, and 3) cognitive and emotional aspects of eating behavior. The effects of age and metabolic syndrome on activation seen in regions such as the insula, orbital frontal cortex, caudate and the hypothalamus may have particularly important implications for taste processing, energy regulation and dietary choices. Implications for development and management of obesity will be discussed.

Supported by NIH grant # R01-AG004085-26 from the National Institute on Aging to C.M. We thank the participants, Drs. S. Horgan, W. Perry, and C.D. Morgan for patient referral, Dr. T.T. Liu and the UCSD Center for fMRI, and Rochelle Britainy Vertrees, Ekarin Pongpipat, and members of the SDSU Lifespan Human Senses Center.

S18
Maternal transfer of food cues

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The maternal transfer of food cues in perinatal fluids has been widely reported in most mammal species and pigs are not an exception. As omnivorous, pigs share large number of physiological similarities with humans and amongst others, perinatal fluids are sensitive to the uptake of odorants and food cues coming from the maternal diet. Although pigs due to the commercial production practices do not have the opportunity to choice between different diets, it has been observed that slight changes in feed composition or ingredients promote changes in the presence of some odorants in amniotic fluid and milk. However, amniotic fluid and milk are considered as perinatal fluids, there is not compositional resemblance between them as the volatile compound profile and most of the volatiles are not shared between amniotic and milk. Even for those compounds that could be found in both fluids, a higher concentration is found in the amniotic fluid than in milk, in which only traces are observed. For this reason it is reported in pigs that the pre-natal exposure to flavours or odour metabolites that can enter to the fetal environment together with nutrients stablishing a positive rewarding relationship with their mother. Changes in the sow diet during the last third of gestation are enough to change the amniotic fluid volatile compounds (flavors) and even chemicals with distinct taste properties transmitted from the maternal diet that can be detected by the fetus and therefore easily recognized by the after birth by the newborn piglet with a long lasting effect that allows the weaned pig recognise their mother. The response of the offspring to flavours experienced in amniotic fluid, indicate that early learning is a powerful tool based on early sensory experiences that reinforces further food acceptance, food intake and welfare. Studies conducted in our group showed that newborn piglets whose mothers consumed an anise or garlic-flavoured feed throughout late gestation showed a clear preference for the pre-exposed flavours than for a control flavour. Similarly other works with aniseed inclusion in the gestating diets showed a calming effect on piglets after weaning when the same flavour was offered to post-weaning pigs indicating an improvement in animal welfare. In humans the learning about the dietary choices of the mother continues when infants experience the flavours of the mother's diet transmitted in breast milk. However in pigs it has been observed that although for example anethol, cynemaldeide and eugenol pass from the sow to the litter through milk the impact of the post-natal experience as reinforcement for further flavour acceptance is lower than the pre-natal experience. Contrarily to humans in which a large variety of flavors (e.g., anise, garlic, ethanol, carrot, mint, vanilla, bleu cheese) are reported to clearly pass via breast milk, lactation in sow seems to be a less effective way maybe explained by the low concentration detected. Similar findings were observed with the same flavour and protocols in human neonates which reveals a strong similarity in terms of the physiology and behaviour related with maternal transference between pigs and humans.

S19
Oral and extraoral nutritional chemosensing in pigs and humans.

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Nutritional chemosensing is the science studying the perception of nutrients in biological systems. Nutrients are perceived in the oral cavity through the taste system allowing to discriminate relevant foods and reflecting species-specific needs. Pigs have a high number of taste buds (19,904) relative to humans (6,074) but the ratio of taste buds to body weight is similar. The comparison of taste sensory studies between the two species is limited by the methodologies used: threshold concentrations are determined in pigs using preference tests, whereas recognition thresholds are used in human studies. Nonetheless, the molecular mechanisms and taste sensitivity are regarded similar between the two species for simple sugars (sweet), glutamate (umami), citric acid (sour) and fatty acids. However, in contrast to humans, pigs do not seem to taste most high-intensity sweeteners, have a broader sensing of amino acids and appear less sensitive to NaCl (salty). Taste differences between humans and pigs are particularly marked for bitterness. Bitter sensing in pigs and humans is diverse and characterised by specific features evolving from the adaptation to different ecosystems. Recently, evidence for expression of taste receptors outside the oral cavity has emerged. Taste/nutrient sensors seem to be involved in the mediation of the hunger satiety cycle but also in the host response to some bacterial pathogens. Studying gene expression in human tissues can be difficult, particularly outside the oral cavity and when using well-controlled nutrition intervention studies. Laboratory rodents are currently the preferred mammalian model for chemosensory studies. However, the distance with humans in terms of diet, digestive physiology and metabolism is quite significant. The similarity in most nutrient receptors studied to date between pigs and humans advocates for the use of the pig as a model for nutritional chemosensory science.

S20
Effects of the Stevia-derived sweetener Rebaudioside A on the incretin hormone glucagon-like peptide-1

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Gut hormones play a pivotal role in the regulation of food intake and the control of gastrointestinal functioning. Their release is induced by food-derived nutrients and sweeteners involving intestinal sensing by chemosensory receptors and/or transporters. One well-known gut hormone is the glucagon-like peptide-1 (GLP-1), which besides its effects on food intake acts as an incretin by influencing insulin secretion. GLP-1 can signal in an endocrine manner via the circulation and in a paracrine manner via receptors located on sensory nerve endings. Additionally, GLP-1 expression levels can differ along the intestinal tract. Here, we aimed to study the effects of Stevia-derived Rebaudioside A, a low caloric sweetener that is increasingly being used in foods and beverages, on intestinal GLP-1 release.
Methods: To address both the endocrine and paracrine way of GLP-1 induced signalling, a 2D intestinal organoid model was developed. 2D organoids were derived from different intestinal locations in order to investigate location-dependent effects. Also, an ex-vivo intestinal pig model was used (2). Additionally, expression levels of gut hormones and

receptors, including glucagon and the glucagon-like peptide-1 receptor, involved in nutrient sensing processes along the intestinal tract were compared for mice, pig and man.
Results: Rebaudioside A augmented GLP-1 release in a location-dependent manner in 2D-organoids derived from duodenal, jejunal and ileal mouse tissue. GLP-1 release in response to Rebaudioside A (1 hr) was found to be highest in ileal-derived organoids. Similar results were found in the *ex vivo* intestinal pig model. Interestingly, prolonged stimulation resulted in an increase of entero-endocrine markers, including increased expression of glucagon which encodes for GLP-1. Furthermore, by comparing location specific expression markers (*Gcg, Pyy, Gpr120, Slc2a2, Slc30a2* and *ApoA4*) from *in vivo* mice tissue with our cultured organoids, it was established that the 2D organoids keep their regional characteristics. Moreover, the expression patterns of nutrient sensing genes along the intestine were found to be highly comparable for mice, pig and human in the distal ileum of the intestine (1).
Discussion and conclusion: Stevia-derived Rebaudioside A enhanced secretion of GLP-1, a key intestinal hormone known to modulate insulin secretion and satiety, in a developed 2D-organoid model. Rebaudioside A-induced GLP-1 release was found to be location specific with highest secretion in ileal-derived 2D organoids. Similar results were found in the *ex vivo* pig intestinal model (2). Interestingly, expression of entero-endocrine markers, including glucagon expression, was enhanced after long term stimulation with Rebaudioside A. These data highlight potentially new applications for rebaudioside A in metabolic diseases like type 2 diabetes mellitus.
1) van der Wielen N, van Avesaat M, de Wit NJ, Vogels JT, Troost F, Masclee A, Koopmans SJ, van der Meulen J, Boekschoten MV, et al. Cross-species comparison of genes related to nutrient sensing mechanisms expressed along the intestine. PLoS ONE. 2014;9:e107531
2) Ripken D, van der Wielen N, Wortelboer HM, Meijerink J, Witkamp RF, Hendriks HF. Steviol Glycoside Rebaudioside A Induces Glucagon-like Peptide-1 and Peptide YY Release in a Porcine ex Vivo Intestinal Model. J Agric Food Chem. 2014 Aug 7.

S21
Imaging brain responses to oral and visceral food stimuli in the pig model

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Describing brain responses to food signals is important to investigate food pleasure and motivation, or decipher the brain matrices underlying sensory and nutrient perception. Studies using large animal models did not make fully use of the opportunities provided by *in vivo* brain imaging yet. Our group was the first to use the pig model to investigate brain responses to oral and visceral food signals *via* ^{99m}Tc-HMPAO SPECT (single photon emission computed tomography) and ^{18}F-fluorodeoxyglucose PET (positron emission tomography), allowing for mapping cerebral blood flow and brain glucose metabolism, respectively. In a pig model of food conditioning, specific modulations of the brain reward circuit were recorded after exposure to flavours with positive or negative hedonic values. This model might be useful to understand the onset of conditioned aversion and anorexia in human patients treated with chemotherapy or radiotherapy for example. To alleviate these clinical symptoms or improve appetite in the elderly and malnourished people, sensory additives might be used to increase food pleasure and consumption. In a recent study in the pig model, we managed to improve food consumption and modulate the metabolism of the brain reward circuit using sensory food additives, which opens the way to further (pre)clinical trials. In another set of studies, we explored the brain responses to sugar. We showed that both duodenal and portal glucose infusions led to activity changes in brain areas regulating food intake and pleasure, but that these stimuli induced different systemic and central responses. Comparing congruent *versus* dissociated oral and duodenal sucrose perception, we found different brain responses in the limbic and reward circuits. These studies are important to understand the brain correlates of sugar craving in the human, as well as the behavioural and brain changes that could emerge further chronic consumption of non-caloric sweeteners for example. Overall, the pig model is a real asset to perform mechanistic studies in sensory sciences and nutrition, or design preclinical studies relevant to human medicine.

PL3
Cellular and molecular basis for taste sensation in Drosophila

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Insects such as the fruit fly respond to a similar spectrum of tastants as humans, but do so through a distinct repertoire of receptors. The largest family of taste receptors in insects are the so called “gustatory receptors” (GRs). We showed these aversive GRs are highly complex and are ionotropic receptors. Recently, we made the surprising discovery that two members of the Drosophila rhodopsin family are co-expressed in gustatory receptor neurons (GRNs), and enable flies to sense and avoid feeding on a noxious compound. The chemosensory roles played by the opsins are light-independent. A well-known, but poorly understood behavior is that animals are attracted to low Na^+-containing foods and reject foods with high Na^+. We uncovered the cellular and molecular mechanism through which low- and high-salt tastes are differentially encoded in GRNs. We also addressed the question as to how dietary experience can alter taste preferences. We found that dynamic regulation of the TRPL channel comprises a mechanism that allows the animals to alter taste behavior in response to a changing food environment. Food texture influences food preferences. However, the mechanosensory receptors responsible for sensing food texture are unknown. Akin to mammals, we found that flies prefer foods with a specific texture. This taste discrimination depends on the fly homolog of the transmembrane channel-like (TMC) family. TMC is expressed in the primary gustatory organ and defines a previously unknown multidendritic neuron (md-L). We propose that TMC and md-L neurons are molecular and cellular mechanoreceptors through which food mechanics is perceived and encoded by a taste organ.

S22
Some like it hot – TRPV1 agonists in health and medicine

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Heat receptors provide an important early warning system to prevent tissue damage. Capsaicin — the 'hot' component of chilli peppers — evokes a sensation of burning pain by activating one such receptor: The ion channel TRPV1. The TRPV1 is a heat-activated, ligand-gated ion channel of the transient receptor potential (TRP) family, and is further specified as belonging to the subfamily V as member 1 (TRPV1). TRPV1 receptors are predominantly found in nociceptive neurons of the peripheral nervous system, but they have also been described in many other tissues, including the central nervous system. In the presence of capsaicin, i.e., or temperatures higher than 43°C, TRPV1 becomes activated. The phospholipid molecule phosphatidylinositol-4,5-bisphosphate (PIP2) inhibits the activation of TRPV1. In 2003, Prescott and Julius (1) identified a motif in the cytoplasmic domain of TRPV1 that had the structural characteristics of a PIP2 binding site and concluded that a compound`s potency for TRPV1 activation is determined by the strength of its interaction with PIP2. Next to capsaicin, other TRPV1 activators include, e.g. allyl isothiocyanate, the pungent compound in mustard and wasabi as well as the endocannabinoid anandamide, or N-arachidonoyl-dopamine. One of the widely studied competitive antagonist is the synthetic capsaicin-analog capsazepine, whereas numerous TRPV1 antagonists have been developed by pharmaceutical companies for the reduction of nociception from inflammatory and

neuropathic pain. Since TRPV1 receptors are also expressed at high levels in the central nervous system, they have been further proposed as a targets for treatment of other neurological conditions such as anxiety or depression. TRPV1 receptors have also been identified to play a critical role in the development of inflammatory hyperalgesia. Whereas mice lacking the TRPV1 receptor failed to develop thermal hyperalgesia during inflammation (2), pharmacological studies using TRPV1 antagonists such as, e.g. capsazepine, demonstrated that mechanical hyperalgesia and inflammatory processes were attenuated in a variety of pain models, making TRPV1 a potential target for novel analgesic and anti-inflammatory drugs. Recent evidence also indicates functional roles of TRPV1 beyond sensory nerve activity. TRPV1 and has been shown to control vascular responses with impact on blood pressure, and to play a homeostatic role in the regulation of body temperature and body fat (3). Recent studies of our own group have demonstrated a less-pungent TRPV1 agonists than capsaicin, nonivamide, to impact adipogenesis and fatty acid uptake in cells in culture (4,5), to reduce ad-libitum energy intake from a standardized breakfast (6) and to prevent a dietary-induced body fat gain in healthy, moderately overweight subjects (7). However, whether the underlying mechanisms for the role of TRPV1 in thermoregulation are intrinsically linked with cellular mechanisms regulating body fat has to be elucidated in future studies.

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S23
Sensation through TRPV1 and TRPA1 in oral cavity

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TRP channels are nonselective cation channels with high Ca^{2+} permeability. They are known to be involved in the taste receptor-independent sensation in the oral cavity, and capsaicin receptor TRPV1 and wasabi receptor TRPA1 are two representatives. Their structures at an atomic level have recently been clarified by the single particle analysis with the cryoEM. We previously reported the physical and functional interaction between TRPV4 and anoctamin1 (ANO1), a Ca^{2+}-activated chloride channel in which Ca^{2+} entering the cell through TRPV4 activates ANO1. Similar physical and functional interaction between TRPV1 and ANO1 was

found in HEK293T cells and mouse sensory neurons. Well co-expression of TRPV1 and ANO1 was observed in the trigeminal ganglion. Large chloride currents were induced in the cells expressing both TRPV1 and ANO1, but not in the cells expressing TRPV1/TRPA1 or ANO1 alone. Capsaicin-evoked inward currents were significantly inhibited by a specific ANO1 antagonist T16Ainh-A01 (A01) in mouse DRG neurons, indicating that capsaicin-induced inward currents are composed of two components; a TRPV1-mediated cation influx and an ANO1-mediated chloride efflux. In addition, capsaicin-evoked action potential was drastically inhibited by A01. These results indicate that the TRPV1-ANO1 interaction is a significant pungency-enhancing mechanism in the trigeminal neurons. The same interaction is true for TRPA1 and ANO1. Thus, TRP channel/anoctamin complex could play an important role in the pungent sensation of capcicum and wasabi.

S24
The complexity of orosensory lipid detection in humans.

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Together with the other two macronutrients, proteins and carbohydrates, dietary lipids are an important source for energy and a supply of essential metabolites in humans and other mammals. While it has been convincingly demonstrated that the building blocks of proteins and carbohydrates, the amino acids and mono- and disaccharides, are recognized in the oral cavity by specialized taste receptors, it is still a matter of debate whether the orosensory perception of lipids also relies on gustatory detection pathways or not. Regardless of the suspected transmission pathway, due to the palatability and high energy density of dietary lipids, the regulation of fat consumption plays an important role for human nutrition. Previous research in human and rodents has identified a number of candidate receptive molecules for free long-chain fatty acids, the prime stimulus for orosensory fat perception, which are currently, alone or in combination, evaluated for their contribution to the detection process.
Using functional calcium-mobilization assays, sensory tests, diverse histochemical and molecular analyses, we investigated the potential of humans to detect fatty stimuli and the occurrence of G protein-coupled receptors specific for free long-chain fatty acids as well as auxiliary proteins in human taste tissue.
Although the majority of data on the ability of mammals to recognize dietary lipids were obtained in rodent models, this presentation will focus on human. We will provide evidence for the expression of fat receptor candidates in human lingual tissue and the pharmacological characteristics of human G protein-coupled receptors involved in fatty acid recognition. The fact that triglycerides, in which fatty acids are bound to glycerol, represent the major form of ingested dietary lipids and that human subjects can detect trioleate, whereas fat receptor candidates do not respond to these molecules, spawned our interest in the occurrence of oral lipases able to liberate fatty acids from triglycerides. Indeed, we detected the presence of lipases in human von-Ebner's gland tissue as well as lipolytic activity in human saliva. In order to identify the cells in human taste buds, which are involved in the detection of fatty acids, we started immunohistochemical experiments using cell type markers combined with GPR120 antibodies. These experiments hint at type I cells as the dominant cell type expressing GPR120 in human taste buds.
Taken together we found that humans, similar to rodents, appear well equipped to recognize dietary lipids in the oral cavity, however, further research to elucidate the detection and transmission of fatty stimuli is necessary.

S25
Beyond olfaction: other airway senses

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The airways contain a myriad of sensory cells that mediate olfaction, control breathing, and protect the respiratory system from damage and disease. The vagus nerve is a major conduit between lung and brain required for normal respiration. Within the airways, vagal sensory neurons detect mechanical stretch of the lung during cycles of inhalation and exhalation, cues associated with inflammation and illness, and irritants that in some species evoke cough. However, mechanisms by which vagal sensory neurons detect and encode respiratory stimuli remain poorly understood. We initiated a molecular and genetic analysis of the sensory vagus nerve, identifying novel vagal receptors, classifying distinct sensory neuron subtypes, and adapting genetic approaches to map, image, and control each neuron population. Using these tools, we identified two vagal afferent subtypes that exert powerful and opposing effects on breathing. One neuron type expresses P2RY1 and consists largely of Piezo2-expressing, fast-conducting A fibers that innervate clusters of pulmonary secretory cells termed neuroepithelial bodies. Optogenetic activation of vagal P2RY1 neurons stops breathing, trapping animals in exhalation, without acutely impacting heart rate or gastric pressure, which are also under vagal control. A second non-overlapping vagal neuron type expresses NPY2R and consists largely of capsaicin-responsive C fibers. Optogenetic activation of vagal NPY2R neurons causes rapid and shallow breathing, similar to some pulmonary defense responses. These findings raise basic questions about the natural stimuli sensed by each neuron type, how signals are transduced, and the necessity of each neuron type in respiratory physiology. Understanding the sensory biology of respiratory control neurons in the vagus nerve may provide therapeutic targets for airway disease intervention.

S26
Interdependent conductances drive infra-slow intrinsic rhythmogenesis in a subset of accessory olfactory bulb projection neurons

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The accessory olfactory system controls social and sexual behavior. However, key aspects of sensory signaling along the accessory olfactory pathway remain largely unknown. Here, we investigate patterns of spontaneous neuronal activity in mouse accessory olfactory bulb mitral cells - the direct neural link between vomeronasal sensory input and limbic output. We identify a subpopulation of mitral cells that exhibit slow stereotypical rhythmic discharge both *in vitro* and *in vivo*. In intrinsically rhythmogenic neurons, these periodic activity patterns are generated in absence of fast synaptic drive. The physiological mechanism underlying mitral cell autorhythmicity involves cyclic activation of three interdependent ionic conductances: subthreshold persistent Na^+ current, R-type Ca^{2+} current, and Ca^{2+}-activated big conductance K^+ current. Together, the interplay of these distinct conductances triggers infra-slow intrinsic oscillations with remarkable periodicity, a default output state likely to affect sensory processing in limbic circuits.

S27
Information Processing in the Accessory Olfactory Bulb

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In my talk, I will discuss three different lines of investigation pursued in our lab, each addressing a different aspect of information processing in the accessory olfactory bulb (AOB). First, I will describe behavioral experiments in which we study whether the AOB can facilitate associative learning. Our findings indicate that rather than playing an exclusive role in innate/hardwired responses, AOB activity, much like that in the MOB, can be associated with arbitrary behavioral responses. Next, I will summarize a set of electrophysiological experiments, in which we show that behaviorally relevant information is encoded by the activity of multiple AOB neurons, each of which can provide only limited information. This finding is inconsistent with the view that readout of a few dedicated "experts" suffices to direct behavioral outputs. Finally, I will show a more refined examination of the temporal evolution of stimulus induced responses in the AOB, as manifest by local field potential oscillations. Specifically, I will describe a unique oscillatory signature in the AOB which is associated with vomeronasal stimulus delivery, and describe our ongoing attempts to better characterize and understand the significance of this unique activity signature.

S28
Inhibitory control of chemosignal processing in the accessory olfactory bulb

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The accessory olfactory bulb (AOB) is a critical computational circuit for chemosensory processing in most mammals. Proper AOB function is required for the expression of behaviors that promote survival and reproduction, including predator avoidance, mate choice, and territorial aggression. The AOB circuit consists of thousands of projection neurons called mitral cells (MCs) and a large and diverse population local inhibitory interneurons. AOB interneurons come in several classes, including juxtaglomerular cells (JGCs), external granule cells (EGCs) and internal granule cells (IGCs). GABAergic inhibition by these interneurons is essential for maintaining proper levels of MC activity. Moreover, MC inhibition has been shown to be plastic, and is increased after salient chemosensory social encounters.

Despite their importance, we know very little about AOB interneuron function. We took a bottom-up physiological approach, with the goal of improving the foundation underlying AOB interneuron research. We discuss two current studies investigating the intrinsic physiology of AOB interneuron classes and their roles in experience-dependent plasticity. In the first study, we made targeted patch clamp recordings from AOB JGCs, EGCs, and IGCs, revealing blends of intrinsic conductances in each that give rise to distinct electrophysiological properties. For example, AOB IGCs have pronounced HCN-mediated I_H currents, and are poorly able to sustain high rates of action potential firing during direct somatic depolarization. In contrast, EGCs have small I_H currents, extremely hyperpolarized resting membrane potentials, and do sustain high firing rates. Quantitative evaluation of AOB interneuron intrinsic properties revealed capabilities and limitations of each class that will be important for understanding their computational roles.

In the second study, we investigated a subpopulation of AOB IGCs that expresses the plasticity-associated immediate-early gene *Arc* following intermale aggressive encounters. *Arc*-expressing IGCs are more strongly excited by sensory input than non-expressing IGCs 4-8 hours after behavior, suggesting enhanced MC-IGC communication specifically in *Arc*-expressing cells. This effect is not the result of an increased number of IGC synaptic gemmules or an increase in the frequency or amplitude of IGC miniature excitatory postsynaptic currents. Instead, Arc-expressing IGCs showed increased intrinsic excitability

and a paradoxical decrease in I_H currents. *Arc*-expressing IGCs strongly inhibit MCs up to one week following behavior, suggesting that these interneurons contribute to experience-dependent MC inhibition. In sum, these results highlight the importance of interneuron diversity and plasticity in shaping MC responses and, ultimately, behavior.

S29

Persistent firing and synchronous infra-slow bursting in accessory olfactory bulb mitral cells: implications for sensory information processing

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Accessory olfactory bulb (AOB) mitral cells exhibit multiple unique features distinguishing them from their counterparts in the main bulb. Among these are prolonged responses to stimuli, infra-slow (<0.1 Hz) rhythmic bursting activity and distinct network connectivity. We combined intracellular recordings, calcium imaging and computational modelling to explore the biophysical basis of these features, as well as their consequences for sensory information processing in the AOB. We found that the prolonged epochs of persistent firing reflect a slow interplay between Na^+ and Ca^{2+} extrusion mechanisms that dictates extremely prolonged dynamics of dendritic Na^+ and Ca^{2+} concentrations. We then examined the bursting activity of AOB mitral cell in light of the slow dynamics of their responses to transient stimuli and the unique network topology of the AOB. We found that infra-slow bursting activity emerges from the interaction between the intrinsic properties and network connectivity of AOB mitral cells. We further show that assemblies of AOB mitral cells are synchronized by lateral connections through chemical synapses and gap junctions. The AOB network topology, where a mitral cell receives input from multiple glomeruli, thus enables integration of chemosensory stimuli over long time scales by inter-glomerular synchrony. Therefore, we provide a possible functional significance for the distinct AOB physiology and topology.

S30

Variation in sequence, structure and ligands of chemosensory GPCRs

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Bitter taste perception can be elicited by dramatically dissimilar compounds[1] and is mediated by bitter taste receptors (TAS2Rs), a subfamily of GPCRs[2]. TAS2Rs present an intriguing case for studying receptor promiscuity: some of the receptors are still orphan, or have few known ligands, while others can be activated by numerous, structurally dissimilar compounds. Furthermore, some compounds are selective towards a single TAS2R, while others activate multiple TAS2Rs. We show that TAS2R-promiscuous and TAS2R-selective bitter molecules differ in size, globularity, and other properties; and develop a ligand selectivity predictor. Selective TAS2Rs, with the exception of hTAS2R5 and hTAS2R49, are activated by promiscuous compounds, which are already recognized by additional TAS2Rs. Thus, unique ligands, that may have been the evolutionary driving force for the development of selective TAS2Rs, still need to be unraveled[3]. The ability of Family A GPCRs and of TAS2Rs to recognize chemically diverse ligands, can be predicted based on the hydrophobicity and accessibility of the canonical ligand-binding site[3,4]. TAS2R receptors have lower affinities and higher agonist/antagonist ratios than typical Family A GPCRs, possibly due to differences in some of the structural motifs[2]. SNPs are less abundant in the transmembrane regions of both TAS2Rs and Family A GPCRs, and some regions of TAS2Rs seem to be more SNP-prone. These results are discussed in the context of tastants discovery, protein evolution and protein engineering.

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S32
Bittersweet human taste genetics

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The human sense of taste is intensely diverse which is due in part to inborn variation in sensory receptor genes such as the T1R and T2R family that produce proteins in the sweet/umami and bitter sensory cascade. While the classic example of individual differences in bitter taste perception is for the receptor T2R38 and its ligand phenylthiocarbamide, we and others have discovered additional examples of extremes in perceptual differences, for example bitter compounds found in plants used for foods or medicines and high-potency sweeteners. In addition to inborn genotype, gene expression and epigenetic modifications of genomic DNA that affects the abundance and timing of gene expression and receptor abundance may partially determine individual differences in taste perception. Not all genes and their protein products that participate in the taste signaling cascade are known; thus, genome-wide association methods, commonly used to identify genes relevant to human disease, can be harnessed to point to novel genes and their variants, e.g., for taste qualities less studied using genetic approaches, such as sourness and saltiness. These genome-wide association methods may also point to transcribed sequences that are not protein coding genes, e.g., non-coding RNAs. While the relationship between taste perception, liking and intake of foods and medicines is not always direct, these basic sensory processes contribute to behaviors like food choice and the acceptance of medicines.

S33
Bitter Taste Receptors in Asthma

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Multiple type 2 taste receptors (TAS2Rs) are expressed in human airway smooth muscle (HASM). TAS2R agonists decrease airway constriction and invoke a bronchodilatory response, suggesting that TAS2Rs could be novel targets for obstructive airways diseases such as asthma. In addition, airway diseases are characterized by airway inflammation, remodeling and mucus production. Extensive in vivo studies using murine model of asthma suggested anti-inflammatory and anti-remodeling effect of TAS2R agonists such as chloroquine and quinine. Furthermore, in vitro studies using human ASM cells demonstrated that bitter tastants inhibit mitogen-induced airway smooth muscle proliferation, and the antimitogenic effect of TAS2R agonists on HASM involves inhibition of cell cycle progression and induction of autophagy. Human peripheral blood mononuclear cell migration is significantly attenuated by chloroquine and quinine supporting our findings from in vivo studies. Twelve TAS2R genes expressed in HASM and known to contain common, potentially functional non-synonymous coding variations were systemically evaluated in 1048 subjects with asthma (41 % severe) for correlation with pre-drug % predicted FEV_1 and post-

bronchodilator max % predicted FEV_1. Coding variations with significant correlations with one or both lung function measures were further evaluated for associations with additional asthma phenotypes and asthma severity. All associations were adjusted for ethnicity, age, and gender. The G allele for the non-synonymous *TAS2R42* coding change $Tyr^{265}Cys$ (TAC>TGC; rs1451772) was found to be associated with higher pre-drug % predicted FEV_1 (ANOVA; p=0.012), post-bronchodilator max % predicted FEV_1 (ANOVA; p=0.009), and log PC_{20} (ANOVA; p=0.015). The SNP rs1451772 is in complete linkage disequilibrium (LD) with the *TAS2R42* coding change $Arg^{292}Gln$ (rs1669412), and there are four additional common non-synonymous coding changes in *TAS2R42*, suggesting that multiple isoforms of this receptor are expressed in human airways. These findings demonstrate the clinical importance of TAS2R genotypes in asthma severity and bronchodilatory responsiveness. Collectively, TAS2Rs have emerged as novel, promising drug targets in obstructive airway inflammatory diseases such as asthma.

PL4
Molecular regulation of taste bud cell renewal

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Taste is a fundamental sense and is crucial for human health. Like our other primary senses, we consider our ability to appreciate sweet, sour, salty, bitter and umami tastes to be relatively constant, even though taste bud cells that transduce these stimuli are renewed rapidly and regularly. The importance of the sense of taste is particularly evident for cancer patients receiving a range of radiation and chemotherapies, as these individuals often experience significant taste loss or dysfunction, and as a result, a significantly diminished quality of life. In the past decade, understanding of cellular and molecular mechanisms governing taste bud renewal has expanded significantly. In numerous renewing epithelia, the Hedgehog (HH) and Wnt/beta-catenin signaling pathways are key regulators of homeostasis. Using an arsenal of multi-allelic mouse models, we have found that HH and Wnt signaling also control taste bud cell renewal. Specifically, Wnt signaling is required to maintain taste cell renewal and is key for continued proliferation of taste progenitor cells, while the level of Wnt/beta-catenin signal impacts the cell fate of new cells generated from the progenitor population. Our working model of HH pathway function is that it promotes taste bud cell differentiation from progenitors, but does not regulate progenitor proliferation. Going forward, it will be important to understand how these pathways function together, as well as how these may be impacted by cancer therapies.

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S34
A multitask mammalian chemosignal and the definition of pheromones

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The concept of pheromone, construed on the basis of insect research (Karlson & Lüscher 1959), designates a *chemically identified* mono- or multi-molecular compound that *selectively* releases in a *conspecific* a *behavioural* or *physiological response* that is formally stereotyped and reproducible with the synthetic version of the compound. Since this minimal conceptualization, research on olfactory communication has been generalized to all animal phyla, from unicellular organisms to tetrapods. The original pheromone concept has fueled an active debate when it was first applied to mammals. Mammalian actions were then thought to

be determined by highly complex mechanisms, especially through widespread learning and context effects. Thus, the original definition evolved to integrate the fact that the stimulus-response coupling should *not depend on previous exposure effects* or *learning*. In view of the huge challenge to discard such exposure/learning effects in the explanation of mammalian behavior, but also in view of the indiscriminate use of the concept by scientists and of its operational emptiness, it was proposed that this concept is tossed out. We will however highlight the potential usefulness of the concept of pheromone in relying on one mammalian case.

The work that led to the identification of a pheromone emitted by lactating rabbit females and acting on newborn rabbits will be reviewed. Extensive investigations by German groups (P Schley, R Hudson) led to reliable bioassays making possible chemical analyses to pinpoint the effective compound in rabbit milk. The milk headspace was analyzed using gas chromatographs equipped with an olfactory port to which pups were presented to concurrently monitor peak elution and pup behavior. This approach led to evidence a key-compound, 2-methyl-but-2-enal (2MB2), in rabbit milk. As synthetic 2MB2 selectivity released pup grasping in both species general and species-specificity ways, and as behavioural activity of 2MB2 developed without prior direct exposure to it, it was then construed as a pheromone and denoted as the 'rabbit mammary pheromone' (rMP). The rMP is obviously involved in female-offspring exchanges, as it elicits immediate arousal in pups, searching/grasping of nipples and ingestion of milk. Pup initial reactivity to the rMP is prognostic to their long-term viability. The behavioral effectiveness of the rMP on pups matches the period when they depend on milk, especially the first 10 postnatal days, after which automatic rMP-induced responses come to be modulated by multisensory and metabolic influences. The response to the rMP can, however, take different forms in different contexts, such as in the nest right after satiation where it controls contact and first ingestion of non-milk items. During this period of the first 10 days post-birth, the rMP fulfils thus multiple tasks: In addition to its releasing effects on various forms of behaviors, it operates i) as a strong reinforcer of olfactory learning during a sensitive period, and ii) as a potential synchronizer, as it appears to be involved in setting circadian rhythmicity in neonate pups.

These multiple functions of the rMP and their relative timing during postnatal development will be discussed in the context of the developmental shift of rMP-induced responsiveness from automatic to integrated will be discussed. Finally, the case of the rabbit pup will be used to propose operational steps in the identification of pheromones in mammals.

S35
The multiple roles of MUPs and their volatile ligands in mouse sexual signalling

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Abstract: Major urinary proteins (MUPs) are excreted in the urine of house mice, together with the low molecular weight volatile sexual pheromones that they bind. MUPs and their ligands play a surprisingly large number of roles in sexual signalling in the mouse and teasing apart the specific roles of each member of this complex multicomponent system is challenging. Components are not expressed independently and, most importantly, act in concert in the natural scent marking system of the mouse. MUPs are encoded by a cluster of over 20 functional genes, with distinct differences in sequence similarity and function between MUPs encoded in the periphery of the cluster or by central genes.

Males compete to attract female mates through urinary scent marking and females must be able to recognise the scent mark owners. The male-specific peripheral MUP darcin plays a particularly important role as a sexual pheromone and is highly potent in inducing learned attraction to the location of male signals and to the odour of specific males. Highly similar central *Mup* paralogues encode a stable individuality signature; this genetic signature also underpins avoidance of inbreeding and preference for more heterozygous males. Individuality arises from a combination of (i) variation in central MUP sequences at specific sites on the MUP surface and within the ligand-binding cavity, and (ii) differential expression resulting in substantial combinatorial diversity between individuals. The genetically

determined signature of central MUPs and darcin determines the volatile signature used to recognise individuals through differential ligand binding by the MUPs.

Under competitive reproductive conditions, investment in these MUP-ligand signals increases substantially among mice of both sexes. While little is yet known about the roles of MUPs in female sexual signalling, female MUP output is elevated slightly around oestrus, and is strongly related to body mass and competitive aggression. Breeding females with the highest MUP output in semi-natural populations are monopolized by males with the highest overall mating success. This suggests that female MUP investment may allow them to mate with the highest quality males and gain heritable fitness benefits for their offspring.

S36
A proposed mechanism for coding pheromones in pig

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Pheromones involved in reproductive behaviours in domestic pig are well known for the 60s. These well-identified molecules open the way to molecular studies of the coding of pheromone compounds by odorant-binding proteins (OBP). In particular, huge amounts of OBPs allow characterisation of post-translational modifications (PTM), as modified proteins represent only a small fraction of total OBPs. Recently, we have identified the olfactory secretome of the pig by using two-dimensional electrophoresis coupled to mass spectrometry (Nagnan-Le Meillour *et al*., 2014). We have shown that 30 OBP isoforms are generated by post-translational modifications, namely phosphorylation and glycosylation, issued from only three genes coding for OBP, VEG and SAL. These PTM are involved in the modulation of binding specificity of OBP isoforms, suggesting that they constitute a dynamic mechanism for the coding of pheromone components.

If the localisation of phosphates sites on Ser/Tyr/Thr residues was successfully achieved by mass spectrometry, those of *O*-GlcNAc were a little bit more complicated. This post-translational modification is unusual on secreted proteins and Sakaidani *et al.* characterised the responsible enzyme in 2011 in Drosophila. This enzyme, eOGT, is resident in endoplasmic reticulum where secreted proteins are elongated and matured. In pig, we have characterised the homolog enzyme, eOGT that modifies secreted OBPs by adding a single sugar on Ser/Thr residues (Nagnan-Le Meillour *et al*., 2014). To assess the glycosyltransferase function of pig eOGT, we expressed the recombinant enzyme in CHO cells and followed its cellular trafficking by fluorescence microscopy in both living cells and fixed cells. This new cellular pathway is important not only to better understand olfaction mechanisms but is also involved in heavy pathologies such as Alzheimer's disease and Adams-Oliver syndrome.

S37
Olfactory communication and reproductive strategies in Old World primates

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It is becoming increasingly clear that olfaction plays an important role in primate sexual behaviour. Female primates signal impending ovulation with a suite of sexual signals. Studies

of these signals have focused on visual, and to a lesser extent, acoustic signals, neglecting olfactory signals, despite the fact that male primates clearly pay attention to female olfactory cues. This is particularly the case for Old World monkeys and apes (catarrhines), which have traditionally been considered as "microsmatic" (*i.e.*, olfactory sense reduced). I will present a study that investigates the role of chemical signals in social and sexual behaviour in one species of catarrhine, the olive baboon (*Papio anubis*). This study aims to investigate the information content of female olfactory signals and to relate these to the female sexual cycle and the fertile period, other female sexual signals (visual, acoustic and behavioural), and male behaviour. This is the first detailed study of olfaction in sexual communication in Old World primates, and the first to integrate information concerning all the potential signals that females exhibit. Our analyses of the volatile profile of baboon vaginal secretions show that they differ with sexual cycle status, suggesting that odour plays a role in signalling the timing of ovulation. This study of olfactory communication provides a crucial missing piece of the puzzle of how females advertise their sexual receptivity and contributes to an improved understanding of the role of odour in reproductive strategies, sexual selection and signalling in primates.

S38
Functional role of the centrifugal feedback to the olfactory bulb: computational model and experimental data

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The centrifugal feedback to the olfactory bulb is mostly targeted on the granule cells, the inhibitory inter-neurons in the olfactory bulb. Using a neural circuit model of the bulb, Li (1990) proposed that such feedback to the granule cells serves the following roles in a context and task dependent manner: (1) odor segmentation by suppressing bulbar responses to ongoing, but already recognized, odors so that a subsequent addition of a foreground odor object can be singled out for recognition, (2) target odor seeking by enhancing bulbar sensitivities to a particular target odor object. I examine the emerging experimental data related to the model predictions: odor adaptation and segmentation in the bulbar activities, input dependency of the centrifugal feedback to the bulb, effect of odor adaptation on odor segmentation and perception, effect of expectation on odor detection and odor perception, effect of odor familiarity on bulbar responses. In addition, the model can be applied to explain how task-dependent feedback can enhance sensitivities in fine odor discrimination (Zhaoping 2016). I will also discuss further experimental investigations that can investigate the role of the centrifugal feedback.

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S39
Neuropilin-1 and the positions of glomeruli in the mouse olfactory bulb

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It has been known since 1996 that mouse odorant receptors (ORs) are involved in determining the positions of the sites of coalescence of axons of olfactory sensory neurons (OSNs) - the thousands of glomeruli in the olfactory bulb. But the mechanisms remain unclear. In 2006 and 2009 a model was proposed by Imai et al. whereby OR-derived cAMP

signals, rather than direct action of OR molecules, determine the target destinations (glomeruli) of OSNs in the bulb. The model hypothesizes that OR-derived cAMP signals determine the expression levels of neuropilin-1 (Nrp1) in OSN axon termini; that levels of Nrp1 in glomeruli form a gradient from anterior-low to posterior-high throughout the bulb; and that these Nrp1 levels determine anterior-posterior patterning of glomeruli. Here, we describe the first independent assessment of the Nrp1 model since 2006. We test the model with the strains used by Imai et al. to formulate their model, and independently, for the well-characterized mouse OR M71 with publicly available gene-targeted strains. Our results do not support the generality of the Nrp1 model. Combined with our analyses of Nrp1 levels in 3D reconstructed bulbs by serial two-photon tomography and a survey of the literature about Nrp1 patterns in the bulb, we conclude that it is unlikely that Nrp1 levels determine positioning along anterior- posterior axes for all 3,600 glomeruli throughout the bulb.

S40

Acute selective serotonin re-uptake inhibitor (SSRI) administration modulates taste sensitivity in untreated depressed human subjects

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Serotonin has been postulated to act as a neurotransmitter in taste buds [1]. In healthy subjects, acute administration of drugs that modulate serotonin (SSRI) or noradrenaline (nor-adrenaline re-uptake inhibitor, NARI) lower taste recognition thresholds after 2h in a taste-modality specific manner [2], suggesting that changes in peripheral rather than central monoamines modulate taste threshold. In patients with depression refractory to treatment, taste is reportedly blunted and resolves with treatment [3]. We sought to determine whether a single acute SSRI could also modulate taste threshold in untreated subjects with depression.

Baseline taste recognition thresholds were measured at the tip of the tongue in an initial sample of healthy (M:F - 6:8; median age 32, range 19-56, n=15) and depressed subjects (M:F - 9:6 (p=0.47); median age 32, range 23-55, n=15 (p=0.8)) recruited from new registrations at a psychiatric clinic (Hyderabad, India) after diagnosing depressive disorder as per International Classification of Disorders-10th revision (ICD-10, WHO, 1992) criteria. In depressed subjects only, thresholds were measured both before and 2h after acute administration of 20mg paroxetine (SSRI). The severity of depression was rated on the Montgomery–Åsberg Depression Rating Scale (MADRS) before taste testing. Ethical approval for the study was given by Maarg Independent Ethics Committee, Hyderabad, India. Two subjects withdrew after the initial taste measurements. Different concentrations of sweet (sucrose, 100-0.1mM), salt (NaCl 100-0.1mM), sour (citric acid 100-0.1mM), and bitter (quinine HCl 1mM – 1μM) solutions were presented to each subject, using cotton buds soaked in each solution. Psychophysical taste functions were constructed to calculate taste thresholds for each volunteer before and after the intervention.

In moderate/severely depressed subjects (MADRS score > 20, n=11), baseline taste recognition thresholds were significantly greater for sweet (25.7±4mM, $p<0.001$), salt (18±3mM, $p<0.001$), sour (18±6mM, $p<0.01$) and bitter (171±50μM, $p<0.01$), compared to healthy subjects (sweet: 2.3±0.3mM, salt:4.7±0.7mM; sour: 1.3±0.2mM; bitter: 16±5.8μM). In depressed subjects (n=13), a single 20mg dose of paroxetine significantly lowered sweet taste recognition thresholds: 12±3mM ($p=0.0017$ compared with before paroxetine, Wilcoxon test), and tended to lower sour thresholds (7.6±2.2mM, $p=0.06$).

In previously untreated depressed subjects, paroxetine lowered sweet and sour thresholds, in a manner similar to that seen in healthy subjects [2]. These findings suggest that although SSRIs have delayed efficacy on symptoms of depression, they can have much more rapid effects on physiological markers such as taste recognition, in previously untreated depressed subjects.

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S41

Molecular markers of GC-D-expressing and Grueneberg ganglion chemosensory neurons

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The main olfactory bulb (MOB) can be differentiated into subregions based on their innervation by molecularly distinct chemosensory neurons. For example, while olfactory sensory neurons (OSNs) of the main olfactory epithelium (MOE) type I odorant receptors (ORs), type II ORs and trace amine-associated receptors (TAARs) share a common cAMP-mediated sensory transduction cascade, they target glomeruli found in distinct domains across the anterior ~90% of the MOB surface. Similarly, OSNs that utilize a cGMP-mediated transduction cascade – guanylyl-cyclase D-expressing (GC-D+) OSNs of the MOE and chemosensory neurons of the Grueneberg ganglion (GGNs) – project to distinct groups of "necklace" glomeruli encircling the caudal MOB. To better understand the unique functionality and neural circuitry of the necklace glomeruli and their associated sensory neurons, we sought to identify additional molecular markers that would differentiate GC-D+ OSNs and GGNs. We found in mouse that the Ca^{2+}/calmodulin-dependent phosphodiesterase Pde1a is expressed in both GC-D+ OSNs and GGNs. Pde1a and immunolocalized throughout the dendrites, somata and axons of these neurons but is not seen in canonical OSNs. Stronger Pde1a immunolabeling in necklace glomeruli innervated by GGNs than in those innervated by GC-D+ OSNs suggests either greater Pde1a expression in individual GGNs than in GC-D+ OSNs or a difference in sensory neuron innervation density between the two types of necklace glomeruli. Supported by NIDCD grant DC005633.

S42

Novel Ca^{2+}-activated Cl^- Channel of the chemosensory cilia of rat olfactory sensory neurons.

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The signaling pathway mediating odor transduction in the chemosensory cilia of olfactory sensory neurons involves cationic cyclic nucleotide-gated channels (CNGs), opened by cyclic AMP. Ca^{2+} entering through CNG opens Ca^{2+}-activated Cl^- channels (CaCCs). A Cl^- efflux through such channels is generally thought to amplify the CNG-dependent inward current. The main component of the Ca^{2+}-activated Cl^- current corresponds to Anoctamine 2 (ANO2), a ~1 pS conductance channel with $K_{0.5}$ for Ca^{2+} >1 µM. Another two CaCCs found in olfactory cilia are Bestrophin-2 (Best2), of similar conductance and $K_{0.5}$, and ANO6, likely combined with ANO2 forming a heteromultimeric channel. ANO2 ablation suppresses the CaCC current in isolated mice OSNs, but has a limited effect over field potential odor responses and does not impair smell behavior. Best2 KO exhibits no cellular and behavioral alterations. The evidence suggests an additional olfactory CaCC. We report a novel CaCC recorded in inside-out patches from olfactory cilia, with substantially higher conductance (>10-fold) and Ca^{2+} affinity (>5-fold). This channel was also found in HEK293 cells transfected with ClCa4l, a CaCC previously detected in olfactory cilia, but not in the non-transfected cells. This channel co-expresses with ANO2 in the cilia, as shown by immunocytochemistry. A similar channel

was previously recorded in toad cilia. The evidence demonstrates a new olfactory cilia Cl^- channel, supporting the involvement of multiple CaCCs in chemotransduction. It also reveals the ciliary expression of ClCa4l, which may correspond to the channel or a functionally related protein.
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S43
Odorant reception in the diamondback moth *Plutella xylostella*

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Olfaction is critical for insect survival, mediating key behaviors such as host preference, mate choice, and oviposition site selection. We identified the olfactory gene subfamilies of the important vegetable pest, the diamondback moth *Plutella xylostella* through *de novo* transcriptomic analysis. These genes include 25 OBPs, 54 ORs, 16 IRs and 15 CSP. We used fluorescence spectrophotometer to measure the binding affinity of PBP to sex pheromone and its analogs. We also used heterologous expression system *Xenopus* oocyte combined two-electrode voltage clamp to deorphanized PRs in *Plutella xylostella*. However, electrophysiological and behavioral assays didnot match with molecular studies. Further investigation revealed that PBP increased the sensitivity of PR to sex pheromone components. Behavioral assays suggested larvae of the diamondback moth *Plutella xylostella* were attracted to their natural sex pheromone and to their major sex pheromone component. However, all three PBPs were detected not to be expressed in the larval antennae. Instead, two general odorant-binding proteins, abundantly expressed in the three major sensilla basiconica of the larval antenna, involved in sex pheromone component reception. We also provided evidence that GOBP2 was a narrowly tuned binding protein, whose affinity could be easily switched from linear pheromones to branched plants terpenoids.

S44
New insights in vertebrate « biotransfolfaction »

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In the peripheral olfactory process, odorant biotransformation is an enzymatic mechanism which modulates the availability of odorant for olfactory receptors. It involved, in particular, Odorant Metabolizing Enzymes (OME) which, alike olfactory receptors, have a preferential expression in olfactory epithelium, a high number of isoforms and a cross-reactivity toward odorants. Our recent studies demonstrated that "biotransfolfaction" is much more than an expected enzymatic mechanism of odorant elimination in order to terminate the olfactory signal and maintain the sensitivity of the peripheral olfactory process. We showed thus, using an *ex vivo* analytical method (headspace gas chromatography), that the availability of odorants in the epithelium environment is strongly impacted by their metabolism. Moreover, we demonstrated using real-time *ex-vivo* analysis (proton-transfer-reaction mass spectrometry) that the fast kinetic of this odorant enzymatic metabolism is in total agreement with the dynamic of the olfactory reception. "Biotransfolfaction" can instantaneously lead to volatile metabolites with odorant properties, potentially interacting with receptors. Additionally, we evidenced, *in vitro* and *ex vivo,* an odorant-odorant metabolic competition mechanism demonstrating that in odorant mixture, the availability of an odorant in the receptor

environment can be strongly modulated by the presence of an other odorant. Strikingly, in the European rabbit, this odorant-odorant metabolic interaction induces perceptual and behavioral consequences, i.e. led to the significant enhancement of the mammary pheromone perception in newborns and of sucking-related orocephalic movements displayed in response to the pheromone. We also showed that each EMO may take in charge dozens of different odorants.

The metabolic events would constitute a supplementary level of sophistication in the complex stimulus peripheral processing: within the OE, odorant-odorant interactions would contemporarily occur at ORs and OMEs' levels, and "biotransfolfaction" would offer to the olfactory peripheral system an additional way to specifically and transiently modulate the peripheral and behavioral response to odors.

PL5
Sour: More than a Primary Taste

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When acidic solutions are taken into the oral cavity, people and animals show avoidance responses, which are commonly attributed to the negative hedonic value of sour taste. Our data brings into question whether this avoidance should be attributed to taste per se, i.e. activation of acid-responsive cells in taste buds -- or whether the avoidance is also due to direct activation of mucosal, e.g. trigeminal, acid-sensitive nerve fibers. In the anterior tongue, the chorda tympani nerve is a pure taste nerve innervating taste buds, while the trigeminal nerve provides general mucosal innervation. Conversely, the glossopharyngeal and superior laryngeal nerves contain two sets of axons, ones that innervate taste buds within the posterior tongue and larynx, respectively, and others that innervate the non-taste epithelium. These general mucosal fibers include polymodal nociceptors many of which express the neuropeptide CGRP, as well as a host of pH-sensitive ion channels including TrpV1, TrpA1, ASICs and ENAC.

The sour-sensing taste cells are Type III cells, which release serotonin (5-HT) upon acidification. Our recent studies (Larson et al *J. Neurosci.* 2016) show that this released 5-HT activates gustatory nerve fibers via 5-HT_{3A} receptors. More recently, we have found that taste fibers expressing 5-HT_{3A} specifically innervate Type III but not Type II taste cells -- and therefore 5-HT_{3A} serves as a marker of sour-responsive nerve fibers. Using transgenic mice in which GFP is driven by the 5-HT_{3A} promoter, we mapped the distribution of the 5-HT_{3A}-expressing (sour) nerve fibers in the primary taste nucleus of the brainstem, the nucleus of the solitary tract (nTS). These GFP-labeled nerve fibers terminate preferentially in the lateral tier of subnuclei within rostral and intermediate levels of the nTS. In order to test whether sour stimulation tends to activate neurons within this 5-HT_{3A}-recipient zone, we mapped citric acid-induced c-Fos in the brainstem orosensory complex including the nTS (nTS) and the adjoining trigeminal nuclei. As described by others, intraoral infusion of citric acid (30mM) evokes substantial c-Fos activity in the lateral and middle segments of the rostral/intermediate (gustatory) nTS – exactly wherein the 5-HT_{3A} fibers terminate. In addition, substantial acid-induced c-Fos activation occurs in the mediodorsal spinal trigeminal nucleus (DMSpV), laterally adjacent to the rostral nTS. Since the DMSpV does not receive input from taste nerves, the c-Fos activation there must arise from trigeminal rather than taste inputs.

In order to test this proposition, we examined acid-induced c-Fos in P2X2/P2X3 double knockout (P2X-dblKO) "tasteless" mice, which lack neural taste responses to acids and all other taste modalities. In these P2X-dblKO mice, intraoral infusion of citric acid evoked substantially reduced activity in the nTS but essentially normal levels of activity in the DMSpV, suggesting that trigeminal detection of acids is intact. This correlates well with the preserved behavioral avoidance of acid solutions by P2X-dblKO mice despite the absence of taste transmission in this strain. We suggest this continuing avoidance is due to activation of

acid-responsive trigeminal or general mucosa nerve fibers rather than to transduction through taste buds. The molecular means by which the trigeminal fibers respond to acids remains to be elucidated.

Democritus: "Sour consists of atoms that are bulky, jagged, and many-angled, without curves"

S45
Olfactory training in patients with olfactory loss and in healthy people

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Patients with olfactory dysfunction benefit from repeated exposure to odors, so-called “olfactory training” (OT). This does not mean the occasional smelling, but the structured sniffing of a defined set of odors, twice daily, for a period of 4 months or longer.

Methods/Results: This effectivity of this treatment has been shown in several studies. In fact, patients with posttraumatic, postinfectious and idiopathic olfactory loss seem to benefit from this measure. In healthy subjects OT seems to work best in children and least well in older people, where it rather stabilizes olfactory function. In young, healthy participants OT appears to increase OB volume as assessed with MRI; it also seems to produce changes in responsiveness at the level of the olfactory epithelium based on electrophysiological recordings.

Conclusion: Short-term exposure to odors seems to modify olfactory function.

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S51
The role of human body odors in ingroup-outgroup relationships

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Why is interaction and integration across members of different so hard to achieve? We present the theoretical framework suggesting that humans have evolved a so-called “behavioral immune system”, a collection of mechanisms designed to early detect and avoid pathogens, including those associated with outgroup members. Here, we focus on the role played in such ingroup-outgroup relationships by chemosensory (body odor) messages. We present preliminary experimental evidence showing that odors from participants of Caucasian and African-American descent qualitatively differ on perceptual ratings (Study 1) and trigger distinctive decision-related strategies, based on the belief of the body odors being collected from the members of the ingroup or outgroup (Study 2).

S52

The impact of androstadienone on the multifaceted stress response in females and males

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Androstadienone (ANDR), a bodily secreted steroid compound, is a socially relevant chemosignal that modulates subjective and (neuro)physiological responses, predominantly in women. The impact of ANDR on stress responses in men and women has not been explored. Using fMRI we aimed to examine psychosocial stress reactions induced by mental arithmetic and social evaluation on behavioral, hormonal and neural levels in healthy women [15 naturally cycling women in their early follicular phase (EF), 15 females in their luteal phase (ML), 15 females on hormonal contraceptives (HC)] and 16 men in an ANDR/placebo treatment repeated-measures design. Additionally, 20 depressed patients have been measured with the same protocol.
In the talk, results regarding sex differences and impact of menstrual cycle phase will be presented. Taken together, our findings suggest that the male stress reaction was more strongly affected by ANDR than female stress reactions. More specifically, ANDR modulated stress-related processes including mood, anxiety, interoception and executive control particularly in men but also in mid-luteal females. Thus our data highlight its significance in communicating social threat that facilitates adaptive stress responses which are sex- and cycle-dependent. Moreover, a short outlook on the effect of ANDR on stress reactions in depressed patients will be given.

S53

Functional metabolomics using a microfluidic chemoreceptor cell assay

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Cell-based receptor assays have found many applications in compound discovery and research. These assays usually have multiwell plate formats and are optimal for high throughput screening. Cell based assays in a microfluidic format have advantages in other applications like screening for differences in receptor signalling dynamics and for applications involving a combination with other microfluidic techniques like liquid chromatography in metabolomics. We investigated the first step to combine LC-MS with a flow-through receptor assay. Next steps aim towards developing a functional metabolomics platform.
We developed a flow through biosensor consisting of human cells expressing both TRPV1, a calcium ion channel which responds to capsaicin, and the fluorescent calcium ion reporter protein, YC3.6. We analysed three contrasting *Capsicum* varieties. Two were selected with contrasting degrees of spiciness for characterization by HPLC coupled to high mass resolution MS. Subsequently, the biosensor was used to link individual pepper compounds to TRPV1 activity. Among the compounds in the crude pepper fruit extracts, we confirmed capsaicin and also identified both nordihydrocapsaicin and dihydrocapsaicin as true agonists of the TRPV1 receptor. Furthermore, the biosensor was able to detect receptor activity in extracts of both *Capsicum* fruits as well as a commercial product (Tabasco). Sensitivity of the biosensor to this commercial product was similar to the sensory threshold of a human sensory panel.
Our results demonstrate that the TRPV1 biosensor is suitable for sequential detection of bioactive metabolites. Novel opportunities for the development of a continuous functional assay, where similar biosensors are directly coupled to the LC-MS will be discussed.

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S54

A novel functional screening assay to monitor sweet taste receptor activation *in vitro*.

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Sweet taste transduction is mediated by the G protein-coupled heterodimeric receptor T1R2/T1R3. Currently, receptor activation responses are either monitored by intracellular [Ca^{2+}] mobilization using calcium-sensitive dyes or by quantifying cellular cAMP or IP3 levels. However, for many receptors it has been shown that, dependent of the ligand, different intracellular signalling pathways can be activated. With a full panel of methods to analyse these different intracellular signalling events a more relevant biological interpretation of responses will be possible. For these reasons, we developed an *in vitro* cell screening assay based on β-arrestin recruitment, a component involved in receptor desensitisation within several GPCR family members.
The screening assay is based on the PathHunter® GPCR platform of DiscoveRx BV. making use of the β-gal enzyme complementation technology in which the N-terminal deletion mutant of β-gal fused to β-arrestin (called enzyme acceptor or EA) is stably co-expressed with the small donor fragment ProLink™. Upon receptor activation by an agonist, β-arrestin-mediated endosomal receptor internalisation takes place forcing the two enzyme fragments to complement leading to increased β-gal enzyme activity which can be quantified using chemiluminescense. Experiments indicate that the sweet taste receptor T1R2/T1R3, activated by natural sugars, artificial sweeteners and enhancers or inhibitors induce β-gal enzyme activity indicating for the first time that β-arrestin recruitment is involved in the desensitisation mechanism of the sweet taste receptor.
The high-throughput 96-well plate screening format of the T1R2/T1R3 cell assay platform is just as sensitive (capable to distinguish 1% fractions between 4 and 9% sucrose) and follows similar S-shape activation curves as most used Ca^{2+} based GPCR read-outs but both assay configurations can also be combined making it possible to study different intracellular pathways at the same time. The assay is suitable to study agonist/antagonist modulator effects of sweet taste enhancers and inhibitors or the influence of sweeteners/sugar mixtures on receptor sensitivity. This *in vitro* screening assay for objective taste analyses is the first to make use of β-arrestin recruitment upon receptor activation and as such, the first within the class-C GPCR family.
We will discuss how such an assay platform provides opportunities studying the synergistic effect of sugar/sweetener mixtures, food-matrix effects, sweet taste modulator and the possibilities to extrapolate such platform to the other basic tastes.

S55

Biosensors based on immobilised Major Urinary Proteins from the mouse

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Major urinary proteins (MUPs) are ligand binding proteins excreted in mice urine. They belong to the lipocalin family, having a beta barrel structure with a highly hydrophobic binding pocket which retains organic molecules, allowing their transport in hydrophilic media. MUPs are involved in mouse chemical communication and they transport small odorant molecules which are slowly released into the environment. They are extremely resistant to environmental challenges like heat, dehydration and proteolysis, making them good candidates for hybrid biosensor implementation. A new bio-sensing platform based on MUPs as chemical recognition elements was developed. The transducers were surface acoustic devices coated with diamond nanoparticles as an intermediate layer enabling covalent attachment of the proteins. The resulting sensors detected 2,4-dinitrotoluene, 4-nitrotoluene and 2-isobutyl-3-methoxypyrazine at ppb levels. The best sensor showed a sensitivity of 24000 $Hz.ppm^{-1}$ to 2,4-dinitrotoluene when grafted with the protein isoform MUP20. Trends in sensitivity of the various volatile organic compound sensors were compared to the association constant values Ka of the proteins to target ligands measured by competitive assay in liquid phase. The system is able to detect analytes both in liquid as well as vapour phase and indicate that MUPs are robust bio-recognition elements that can be utilized in artificial olfaction applications.
Acknowledgement: Financial support from the European Commission Research Innovation - 7th Framework Program (FP7-SECURITY project n° 285203) is gratefully acknowledged.

S56
Multiple receptors contribute to the transduction of fat taste

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Fat is the most energy-dense macronutrient, yet there is incomplete knowledge on how fat is detected by the taste system. Two receptors GPR40, GPR120 and a transporter, CD36, contribute to detection of free fatty acids by the taste system. These three proteins are expressed in gustatory sensory cells, and knockout mice lacking either of those genes have diminished preference and taste nerve responses for fatty acids and oils. However the responses to oil and fatty acids are not totally abolished in any of these knockouts. Strikingly, very strong preference for intralipid, an emulsion of soybean oil, remains in all three lines of knockout animals. To determine if the residual response of GPR40 KO and of GPR120 KO mice to fat is caused by the activity of the other fatty acid-responsive GPCR, we analyzed the responses to fatty acids and intralipid of GPR40/GPR120 double KO mice by two bottle preference tests and compared them to the responses of single knockouts and wild type mice. We found that preference and/or intake were in the following order: WT > GPR120KO > GPR40KO = GPR40/GPR120 double knockouts. These results suggest that GPR40 plays a more important role than GPR120 in mediating preference for fat, and that in the absence of GPR40, GPR120 does not play a role.
We also investigated the response to fat of KO mice lacking Trpm5, a common signaling element downstream from G-protein-coupled taste receptors. We found that the preference for intralipid was more impaired in those mice, compared to that of GPR120 or GPR40 single and double KO mice or CD36 KO mice. Together the data show that in addition to GPR40, GPR120 and CD36, other receptors may contribute to the oral detection of fat.

S57
CD36 is involved in fatty acid detection in the murine olfactory system

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Olfactory signals influence food intake in a variety of species. To maximize the chances of finding a source of calories, an animal's preference for fatty foods and triglycerides already becomes apparent during olfactory food search behavior. However, the molecular identity of both receptors and ligands mediating olfactory-dependent fatty acid recognition are, so far, undescribed. We here describe that a subset of olfactory sensory neurons expresses the fatty acid receptor CD36 and demonstrate a receptor-like localization of CD36 in olfactory cilia by STED microscopy. CD36-positive olfactory neurons share olfaction-specific transduction elements and project to numerous glomeruli in the ventral olfactory bulb. In accordance with the described roles of CD36 as fatty acid receptor or co-receptor in other sensory systems, the number of olfactory neurons responding to oleic acid, a major milk component, in Ca^{2+} imaging experiments is drastically reduced in young CD36 knock-out mice. Strikingly, we also observe marked age-dependent changes in CD36 localization, which is prominently present in the ciliary compartment only during the suckling period. Our results support the involvement of CD36 in fatty acid detection by the mammalian olfactory system.

S58
Neural mechanisms underlying the ageing-associated decline in chemosensory perception

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Ageing is associated with anosmia - loss of the sense of smell - a fundamental feature of several neurodegenerative disorders such as idiopathic Parkinson's disease (IPD), Alzheimer's disease (AD) and Huntington's disease (HD). The decline in the capacity to perceive and discriminate odors in aged humans leads to a dramatic decline in the quality of life and weakened appetite with accompanying impacts on nutritional status. Although the gradual loss of olfaction is described in the context of ageing-associated neurodegenerative diseases, the paucity of information on the underlying cellular and molecular mechanisms in the aged olfactory system remain surprising. These mechanisms may include variations in neuronal populations, synaptic organization, and neural/synaptic functions. We have observed gradual regressions in the olfactory driven choice behavior of *Drosophila melanogaster* with natural ageing. Nonetheless, the vision and motility of aged flies were significantly intact in the behavioral analyses. Consistent with their reduced olfactory response, aged flies show declined olfactory neuronal activities. High throughput deep RNA sequencing revealed downregulation of 181 inheritable factor in the antenna of old flies. Then again, inhibition of superoxide dismutase2 (SOD2), which normally leads to premature aging, in olfactory projection neurons (PNs) instigated similar olfactory declines in young flies as their older counterparts. Remarkably, defunct olfactory perception in the older flies was fully rescued with overexpression of SOD2 in projection neurons. We anticipate that this comprehensive investigation will identify conserved and potentially causative mechanisms of ageing-associated olfactory functional decline.

S59

Oral and intestinal sweet taste T1R2/R3 receptors in mice; effect on consumption, over weight, blood glucose and insulin levels

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Stimulation of oral Type II taste receptor cells with T1R2/R3 receptors elicits sweet taste and invites to consumption. Intestinal Type II taste receptor cells with T1R2/R3 receptors facilitate glucose absorption by the glucose transporter type 2 (GLUT2). Type II taste receptor cells contain a calcium channel, CALHM1. Genetic deletion of the CALHM1 channel results in loss of ability to sense and perceive the sweet taste quality. Comparison between mice with CALHM1+/+ (WT) and without CALHM1-/- (KO), respectively, provides the means to examine T1R2/R3 receptors' effect on intake and intestinal absorption via measurements of body weight (BW), blood glucose (BG) and plasma insulin. In this study we confirm our findings that WT mice are heavier, eat more, and have higher mortality than KO mice. We report that higher BG and insulin levels accompany higher BW in both WT and KO mice, although KO mice with the same BW as their WT counterpart have lower BG and insulin level. Glucose gavage increased and prolonged BG and plasma insulin increases more consistently in WT than in KO mice. The effects o fructose gavage was small and did not differ between WT and KO mice. Gavage with the high potency artificial sweetener SC 45647 increased both BG and insulin levels in WT but less than with glucose. This increase was also larger in heavier WT mice than lighter. In KO mice the effects of gavage with water or SC 45647 on BG levels and insulin did not differ significantly. In WT mice there was a difference. These results suggest that inhibition of T1R2/R3 receptors lowers intake and intestinal uptake, which then decreases BG and insulin levels. These findings can be applied to weight control in humans.

S60

Rise of the urethral brush cells

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We recently identified a novel cholinergic chemosensory cell in the urethra of 14 mammalian species including humans (urethral brush cell = UBC). UBC utilize the canonical bitter and umami taste transduction signaling cascade to detect putative harmful compounds (e.g. bacterial bitter substances) and initiate reflex detrusor contractions by cholinergic transmission to sensory nerve fibers.
Here, we addressed their postnatal and their ontogenetic origin utilizing choline acetyltransferase (ChAT)-eGFP and Wnt1-tomato reporter mice, deep sequencing to identify potential novel markers for these cells, and a mouse model (MyD88-knockout) based on these sequencing data.
Postnatal appearance of UBC was assessed in urethral whole mounts of ChAT-eGFP reporter mice of both genders from day P0 to P100. UBC appear first at P4-P6 in male and at P13-P14 in female mice. After about 10 weeks, UBC numbers have reached the same level in both genders. MyD88 is a key factor for the genesis of UBC, since they were absent in urethrae of MyD88 knock out mice (N=5). Deep sequencing and immunolabeling of tissue sections from ChAT-eGFP mice revealed doublecortin like kinase 1 (DCLK1) as strong UBC marker. Antibodies against this marker were applied to urethral sections of Wnt1-cre tomato mice, a reporter strain for cells of neural crest origin. Neither DCLK1-positive UBC nor serotonin-positive endocrine cells expressed Wnt1-driven tomato, and, hence, are not derived from the neural crest. On the other hand, UBC were immunolabeled with antibodies against the epithelial marker cytokeratin 18 in ChAT-eGFP reporter mice. These data were also supported by deep sequencing.

Taken together, UBC represent cholinergic epithelial cells not derived from the neural crest which develop postnatally in a MyD88-dependent manner.

S61
Specificity versus Promiscuity: The Ligand-binding Pocket for Bacterial Signal Peptides in Formyl Peptide Receptors

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We recently discovered that bacterial signal peptides represent a novel pathogen-associated molecular pattern for formyl peptide receptors (FPRs) and showed that the vomeronasal receptor Fpr3 reacts to a specific set of signal peptides (Bufe et al., JBC 2015). By contrast the receptors FPR1 and FPR2 that are expressed in the mouse and human immune cells are broad detectors that can potentially detect more than 100,000 different peptides. A reliable and specific detection of bacterial signal peptides represents a molecular challenge because signal peptides are a vast group of molecules with a highly variable chemical structure. Direct binding studies with fluorescently labelled peptides, in combination with competition experiments and site-specific point mutations, enabled us to obtain the first insight into the underlying detection mechanism of bacterial signal peptides by FPRs. Competition studies using a chemical antagonist provide clear evidence that a single binding site in mouse and human FPR1 is sufficient for the detection of a vast array of different bacterial signal peptides. We identified three amino acid residues in the binding pocket of mouse and human FPR1 that are critical for bacterial signal peptide detection. These are highly conserved in the FPR1 sequence of different species and their mutation leads to a drastic loss in the functional response and the binding affinity of signal peptides. Sequence comparison of all mouse and human FPRs revealed a distinct pattern of conserved amino acid exchange between the immune and vomeronasal receptors at these positions. We propose that these alterations in the binding pocket contribute to the functional differences between immune and vomeronasal FPRs. Our experiments using receptor mutants in combination with peptide derivatives also provide clear evidence that ligand detection of FPRs does not primarily rely on specific interactions with conserved amino acid residues. Instead, these results suggest that FPRs favour a shape-oriented detection process that focuses on the ability of a peptide to assume a specific three-dimensional conformation. This novel concept of a primarily three-dimensional oriented ligand recognition may also lead to a better understanding of receptor-ligand interactions of other receptor families such as odorant or taste receptors that recognize broad ligand repertoires as well.

S62
A calcium signaling 'fingerprint' in vomeronasal sensory neurons

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Recently, a growing number of chemosensory signals were discovered which are detected by the mammalian vomeronasal organ (VNO). These complex chemical cues regulate social behavior and carry information about sexual, social and reproductive status of both con- and heterospecific individuals. To date, however, little is known about both the sensory coding strategies implemented by the VNO and the stimulation-dependent activity patterns in single vomeronasal sensory neurons (VSNs).
In this study, we used Ca^{2+} imaging in acute mouse VNO slices to determine the activity of VSNs both on the population and the individual neuron level. Slices were loaded with the Ca^{2+}-sensitive dye Cal-520/AM in a custom-made circulating oxygenation chamber. This new loading approach significantly increased the yield of vital VSNs. Precise focal perfusion of gender-specific pooled urine samples allowed us to analyze the neural code of vomeronasal

information. In parallel, we analyzed the spatiotemporal response characteristics in single VSNs.
Together, we present an improved *in situ* Ca^{2+} imaging approach that will allow an effective VNO ligand screening, characterization of population response patterns, kinetics analysis of individual Ca^{2+} transients, as well as investigation of VSN adaptation and signaling robustness. Thus, on-going experiments aim to provide a quantitative perspective on vomeronasal coding at the VSN population level as well as a detailed analysis of Ca^{2+} signaling events in single neurons.

S63
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.

S64
Effect of aging in the activity of the posterior piriform cortex of rats during flavor recognition memory

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Deficits in chemosensory recognition memory are among the first signs of pathological aging. Brain areas involved in aging-related decay can be studied in rats using habituation of flavor neophobia as a model of recognition memory. The habituation of taste neophobia relies on the ability to assess the familiarity of a previously ingested flavor without negative consequences. This leads to increased intake over repeated exposures to the flavor. The habituation of taste neophobia seems to depend on a neural circuit that involves the insular cortex (IC) and its connections with the basolateral amygdala (BLA) and the perirhinal cortex (PRh) among others.

This type of memory has been studied using basic taste solutions (salty, sweet, acid and sour), and there are important differences in the olfactory component of the stimuli, ranging from odorless saccharin or quinine taste solutions to vinegar or lemon flavored solutions with a strong odorant component. Previous results in our lab using a flavored cider vinegar solution (3%) have shown that aged rats exhibit a peculiar pattern of brain activity that might be related to age-induced changes in flavor recognition memory. Research about the potential role of the piriform cortex (PirCx) in taste recognition memory is scarce. Given the

convergence between olfactory and gustatory circuits at the level of the posterior PirCx and the fact that this region is considered as a multisensory integration area of taste and olfaction, it seems particularly relevant to explore potential activity changes related to flavor novelty and familiarity. In order to explore this, 21 five-month-old (n=7 per group) and 24 twenty-four-month-old male Wistar rats were exposed to a novel cider vinegar solution (3%) for one, two or six consecutive days using 15-minute drinking sessions. Then, c-Fos immunohistochemistry was applied in the anterior and posterior Piriform Cortex as an index of neural activity. The behavioral results confirmed the habituation of flavor neophobia which progressed as the number of exposures to the vinegar solution increased in both groups of age, although it was delayed in time in the case of aged rats. The immunohistochemical analyses revealed an increased number of c-Fos positive cells in the rostral portion of the posterior PirCx in the group that had 6 exposures to vinegar compared to the groups that had one or two exposures to the flavor. No differences in c-Fos expression were found in the anterior PirCx between groups. This suggests a potential role of pPirCx in flavor recognition memory. Its selective involvement in the fully consolidated flavor memory is consistent with its anatomical connections since it maintains reciprocal connections with IC and it is considered a multisensory integration area.
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S65
Automated operant olfactory conditioning of group-housed mice

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Despite the staggering increase in specific as well as efficient techniques for generating transgenic mouse lines the behavioural analysis of these strains still relies heavily on manual characterisation of individual animals. Not only is this approach labour and cost-intensive but also highly prone to experimenter-induced errors and variations. Additionally, most tests are conducted on single-housed animals during the daytime – a suboptimal setting for natively social and nocturnal animals.
To circumvent these limitations, we used an automated operant olfactory conditioning setup that allows for group-housing of even large cohorts of animals (>18 subjects) while simultaneously training these animals on a go/no-go odour discrimination task.
Animals, identified via an implanted RFID-tag, could initiate trials themselves allowing for generation of unique training protocols specifically tailored to each animal. The animals reached >95% correct performance as quickly as during manual training and we were able to resolve discrimination time differences as described previously by us and others (Nixon et. al 2004, Shimshek et. al 2005, Nixon et. al 2010). Simultaneously we could monitor key additional parameters like the licking-patterns, air flow, air pressure, temperature and humidity with millisecond precision.
In summary, this setup enables automated training of socially housed mice while minimizing experimenter interaction with the animals. Apart from the odour discrimination tasks described here the setup can readily be expanded to encompass additional sensory cues or even serve as a pre-training phase to screen for high performing animals for use in further studies like awake 2P imaging or electrophysiological recordings.
This work was supported by funding from the Deutsche Forschungsgemeinschaft SPP 1392 “Integrative Analysis of Olfaction”.

S66
Odor threshold relates to sexual pleasure

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Introduction
The olfactory system contributes significantly to human social behaviour, playing a key role in mate choice and empathic functioning. In this context, previous research (Croy et al. 2012) discussed the importance of the sense of smell for intimate relationships. Following up on this, the present study examined the potential coherence between odor threshold in young and healthy participants and their sexual desire, sexual pleasure and sexual behaviour in a more detailed fashion.
Methods
In 80 healthy volunteers (29 male, 51 female; mean age 25±4 years SD, range from 18 to 36 years) odor threshold was assessed using the "Sniffin' Sticks". Further, participants responded to a battery of questions on sexual desire (SDI Sexual Desire Inventory, Kuhn et al. 2009), sexual pleasure (orgasm frequency, perceived pleasantness of sexual activities on a VAS) as well as sexual behaviour (stating the number of sexual partners, frequency of having sex, average duration of sexual intercourse).
Results
Correlational analysis revealed a coherence between odor threshold and variables for sexual pleasure: Participants that scored higher in the odor threshold test, reported a higher amount of perceived pleasantness of sexual activities. Further, women with a better sense of smell stated a higher frequency of orgasms during sexual intercourse. In women, a slightly, but not significantly higher value for the odor threshold could be determined compared to men. Our data did not show an association between the sense of smell and sexual desire or sexual behaviour.
Conclusion
The present results suggest a contribution of the sense of smell to the perception of sexual pleasure.

S67
The Effect of Contextual Odors on Emotional Evaluations of Facial Expressions

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Successful social interaction relies on accurate decoding of other peoples' emotions and intentions. Much of our social communication is non-verbal; faces are perceived in a context. Previous research has shown that emotions are recognized faster and more accurately in a congruent odor context (e.g. disgusting odor paired with a disgusted facial expression). Previous research has also shown that emotions may modulate the N170 ERP component. For the first time we can report how odors affect both behavioral measures and brain activity. We investigated how contextual odors affect the brain activity and the emotional valence and arousal when participants viewed facial stimuli. Facial expressions were rated as more emotionally valenced in an odor context compared to a no-odor context. Further, odors increased the rated arousal of the facial expressions. However, this arousal effect depended on the odor context for happy faces, in a pleasant-odor context the faces were rated as more arousing than in the disgusting odor context. In contrast the N170 component was stronger for disgusted faces in disgusting odor trials than in trials with a pleasant odor context. These results suggest that for evaluative responses positive information is more pronounced; whereas brain responses are more attuned to negative information in emotional odor contexts.

(Athens, 6th century BC)

POSTER PRESENTATIONS

P1
Highlighting the large variation in perceived properties of odors over time

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Individuals differ considerably when rating the perceived properties of odors, especially over time. A second glance at previously published data-sets from our laboratory revealed that the same invariant exposure often produced both floor and roof effects. An odor that at the end of the exposure session was regarded as non-existent by one participant, could border the ”absoulte maximum” rating category in another. We provide re-analyses from four exposure studies where we illustrate the perceptual variability over time, and outcomes associated with such ratings. We note that high, compared with low ratings of odor intensity over time is associated with ratings of unpleasantness and symptoms, but also with everyday distress, cognitive performance, autonomous nervous system activity and deviating responses in the so-called pain or saliency matrix of the brain. We bring an open question to ECRO regarding how this considerable variability should be interpreted, and what the consequenced are for research and for setting exposure limits.

P2
Effects of Androstadienone on dominance perception and gaze avoidance in low and highly socially anxious males

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In the animal kingdom, chemosensory information has been long known to convey signals of dominance, communicating information about the sender's social status and general fitness. Increasing evidence suggests that humans may also communicate both trait-dominance and state-dominance via chemical compounds. Androstadienone (androsta-4,16,-dien-3-one), a testosterone derivative chemosignal found in human sweat, seems to be a likely candidate for signaling dominance and aggression in humans.
A psychopathology involving concerns pertaining specifically to social dominance is social anxiety. Socially anxious individuals tend to see themselves as being low in the hierarchical rank and others as dominant competitors for social status. These individuals are hypersensitive to signs of dominance and they are prone to respond in maladaptive submissive behaviors. One such behavior is a symptom commonly reported in social anxiety - avoidance of eye contact. Socially anxious individuals tend to avert their gaze and avoid the eye region of their adversary as a sign of submission or in an attempt to prevent feared social catastrophes.
The current study aimed to investigate whether androstadienone serves as a chemosignal of dominance in low and high socially anxious (HSA) males. In the first experiment, we expose normosmic, heterosexual male participants to images of male targets depicting dominant, submissive and neutral facial poses. They are then asked to rate the target’s dominance level on a 9-point scale. In the second experiment, participants are exposed to a free-view task of male targets depicting similar dominant and neutral facial poses, while we examine their visual scanpath using an eye-tracker. Participants, divided to two groups according to their social anxiety level, complete these two tasks twice, once under exposure to androstadienone and once under exposure to a blank control solution. We hypothesize that compared to a blank control solution, when exposed to androstadienone, participants, and HSA individuals in particular, will rate the male protagonists as more dominant and show a lower number of fixations and shorter dwell time around the eyes region of the targets. Preliminary results from the first experiment show that HSA rated the dominance of protagonists higher during exposure to androstadienone compared to the control solution. Additional results of this ongoing effort will be presented.

P3
Odorant-binding proteins mutants having novel binding properties

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Odorant-binding proteins (OBP) are small soluble proteins present in the nasal mucus covering the olfactory epithelium. Vertebrate OBPs belong to the lipocalin superfamily, whose members share a common scaffold made of 8-stranded β-barrel. This folding pattern defines a central apolar cavity, named calyx, whose role is to bind hydrophobic molecules such as odorants. Although the physiological role of OBPs is not clearly established, they are supposed to carry odorants from the air to olfactory receptors through the aqueous nasal mucus. OBPs have been described in numerous species including pig, rat and human beings. OBPs are broadly tuned and bind a large spectrum of volatile molecules. Interestingly, it has been shown that the three rat OBP subtypes (rOBP1, rOBP2, rOBP3) have different and complementary ligand properties [1], suggesting that OBPs are involved in odorant discrimination. Protein sequence alignment of the three rat OBPs reveals the presence of an amino acid residue located in the binding pocket, which may be important for guiding binding specificity. Using site-directed mutagenesis, we generated variants of rOBP3, in which this amino acid residue has been substituted. Using isothermal titration calorimetry, we found that some substitutions decreased the affinity of rOBP3 towards some odorant molecules while others generated OBPs possessing novel binding properties. Our work gives new elements to understand the binding mechanisms of OBPs and opens the way towards technological applications based on OBP, as odorant biosensors.

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P4
Stability of Odorant Binding Protein Biosensors over Time

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Odorant Binding Proteins(OBPs) can be used as Biosensors to detect variety of volatile organic compounds(1,2,3). The previous work showed that OBPs immobilised on to quartz crystal microbalances (QMB) are capable of sensitive detection of different volatiles with selectivities dependent on the type of OBP and ligand affinity, but it was not known how stable these sensors are over time. An array of sensors comprising PigWT-OBP1, Agam-OBP4, Agam-OBP1-S82P, immobilised using a self-assembled monolayer on to QMBs were tested for their responses towards saturated vapours of Cocaine, Ephedrine, Trinitrotoluene and Tobacco continuously over a period of four months. The results indicated that although there were small daily variations in response the sensors were able to continue to sensitively detect the target analytes over the period tested. This indicates that biosensors comprising immobilised OBPs may be robust enough for long term use in industrial applications.
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P5

Sequence variation determining stereochemistry of a Δ11 desaturase active in moth sex pheromone biosynthesis

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A Δ11 desaturase from the oblique banded leaf roller moth Choristoneura rosaceana takes the saturated myristic acid and produces a mixture of (E)-11-tetradecenoate and (Z)-11-tetradecenoate with an excess of the Z isomer (35:65). A desaturase from the spotted fireworm moth Choristoneura parallela also operates on myristic acid substrate but produces almost pure (E)-11-tetradecenoate. The two desaturases share 92% amino acid identity and 97% amino acid similarity. There are 24 amino acids differing between these two desaturases. We constructed mutations at all of these positions to pinpoint the sites that determine the product stereochemistry. We demonstrated with a yeast functional assay that one amino acid at the cytosolic carboxyl terminus of the protein (258E) is critical for the Z activity of the C. rosaceana desaturase. Mutating the glutamic acid (E) into aspartic acid (D) transforms the C. rosaceana enzyme into a desaturase with C. parallela-like activity, whereas the reciprocal mutation of the C. parallela desaturase transformed it into an enzyme producing an intermediate 64:36 E/Z product ratio. We discuss the causal link between this amino acid change and the stereochemical properties of the desaturase and the role of desaturase mutations in pheromone evolution.

P6

***"Sensorial Chemistry"*: Functional characterization and structure-activity relationship understanding of OR5K1 and OR2AG1 allow to design and synthetize new selective compounds.**

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ChemCom has deorphanized more than 120 human ORs relying on (i) an efficient proprietary screening system and (ii) libraries of thousands of odorant compounds. Accordingly, ChemCom is currently identifying and characterizing new modulating molecules (enhancers or blockers) and novel odorant compounds for the whole range of hORs. An extensive comparative structure-activity relationship (SAR) data for two unrelated hORs, OR5K1 and OR2AG1, highly expressed in the whole olfactory mucosa, revealed firstly that OR5K1 presents a wider selectivity than OR2AG1, and secondly that none of the commercially available compounds tested so far is selective for OR2AG1. By making an in depth analysis on SARs established on both hORs, subtle differences on aza heterocycles were identified leading us to design ligands likely to display selectivity for OR2AG1 versus OR5K1. After chemical synthesis and *in vitro* characterization and human sensory assessment, that those compounds effectively present the expected selectivity showing, for the first time, that "Sensorial Chemistry" might allow to modulate the olfactory properties of volatile organic compounds and might be a valuable tool as medicinal chemistry is in the pharmaceutical area.

P7

Perceived stress, sensory irritation and levels of prostaglandin F2α in plasma after acrolein exposure - a pilot study

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Acrolein is a known sensory irritant present in cigarette smoke, smoke from fires, automobile exhaust and smog. Psychological and physiological stressors might work in synergy and potentiate each other by activating the same physiological pathways, such as inflammation, which may lead to an altered response to toxic agents such as acrolein. The objective of the study was to examine the relationship between perceived stress, sensory irritation and the concentration of four eicosanoids in plasma after exposure to acrolein.

Each participant (n=30) took part in two exposure conditions – one with the mild odorant heptane (the masking compound), and one with heptane and acrolein at a dose below previously reported sensory irritation thresholds. During the 60 minutes exposure, eye, nose and throat irritation was rated on Borg's CR-100 scale. Blood samples were collected before entering the exposure chamber, directly after exiting and 24 hours post-exposure. The four eicosanoids evaluated in this exploratory study were PGF2α, PGE_2, PGD_2 and TXB_2. Before exposure self-reported stress was assessed by the perceived stress questionnaire (PSQ).

Participants with moderate to high level of stress perceived the masked exposure to acrolein near the detection threshold as more irritating compared to the control condition and the participants with low stress. There was a significant correlation ($r=0.49$; $p<0.01$) between self-reported stress and increase of prostaglandin F2α immediately after the exposure and also 24 hours after ($r=0.39$; $p<0.05$). No correlation was found before the exposure or at any time before or after the control exposure. No correlations were found for the other three eicosanoids. The results suggest that perceived stress is associated with increased sensitivity to low-level exposures of acrolein via a prostaglandin F2α mediated response.

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P8

Molecular adaptation in olfactory functions in the fire ant social chromosome

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Insects use pheromones as a major mode of communication. Social insects need a complex language of signals to coordinate cooperation between thousands of individuals. The evolution of such chemical communication required genes for the synthesis of pheromones and for their olfactory perception. In the fire ant *Solenopsis invicta* chemical signals are involved in the determination of social organization. Single-queen (monogyne) or multi-queen (polygyne) social structure is determined by the "social chromosome": a non-recombining region consisting of approximately 527 genes with two distinct haplotypes: *SB* and *Sb*. Monogyne queens are always *SB*/*SB* and polygyne queens are always *SB*/*Sb*. Workers discriminate monogyne and polygyne queens based on olfactory cues, presumably non-volatile lipids on the queens' cuticle. We searched for candidate genes in the social chromosome that could be responsible for this discrimination.
We focused on olfactory receptors (ORs), because this gene family was dramatically expanded in the evolution of ants. We annotated 472 putative ORs in the *S. invicta* genome. The OR gene tree shows many *S. invicta*-specific expansions, and multiple branches show

evidence for positive selection. Notably, a cluster of 23 ORs resides in the social chromosome. Nine genes in this cluster are the result of recent duplications in the *S. invicta* lineage. We also identified significant differences in these genes between the *SB* and *Sb* haplotypes. The most dramatic difference is the complete deletion of two of these genes in *Sb*. Furthermore, projection of amino-acids changes along the *S. invicta* lineage on a structural model of ORs revealed significant changes in hydrophobicity in a putative ligand recognition site. This result is in line with the expected hydrophobic nature of queen cuticular pheromones. Therefore, these receptors are prime candidates for involvement in queen discrimination. These results suggest that the evolution of polygyne social organization involved adaptations in genes responsible for olfaction and opens the way for functional studies of the molecular mechanism of this phenomenon.

P9

Olfaction versus audition: Perceptual, cognitive, and metacognitive functions in early blind, late blind, and sighted individuals

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Previous research has shown that blindness can lead to improved perceptual and cognitive abilities in the nonvisual senses. However, few studies have compared different sensory modalities using similar perceptual and cognitive tasks. In this study, olfactory and auditory absolute threshold, discrimination, identification, episodic recognition, and judgments of learning (JOL) were assessed in early blind (n = 15), late blind (n = 15), and sighted (n = 30) participants. The largest advantages for blind participants over the sighted were found for auditory discrimination and episodic recognition, advantages that were most clear for early blind participants. Although trends of group differences in the same direction were found for some of the olfactory tasks, these differences were not statistically significant. Moreover, early blind participants showed better metacognitive abilities in predicting olfactory and auditory memory than late blind and sighted participants. In conclusion, there were clear modality- and task-related group differences. The larger effects observed for the auditory sense might be related to increased attentional capacity and training, as this sense is crucial for everyday functioning among blind individuals.

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P10

Peripheral odour processing and configural perception in newborn rabbits

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Olfaction is involved in vital behaviours such as interactions between mother and young, searching for food, etc. Throughout life, these behaviours are displayed by animals (including humans) in a complex and changing olfactory environment composed by molecules (odorants) forming mixtures, perceived as odours after their processing by the olfactory system and brain. To deal with this complexity, organisms process mixtures in the elemental (perception of elements that compose the mixture) or weak/robust configural ways

(perception of a configuration in addition to/at the expense of the elements). These modes of perception, used alternatively or simultaneously, are shared between organisms across the animal kingdom. Sometimes, a given perception mode is shared between species for the same mixture. For instance, newborn rabbits perceive in the weak configural way a binary AB mixture of ethyl isobutyrate (A) and ethyl maltol (B) at the 30/70 v/v ratio, which is also perceived configurally, at the same ratio, in human adults. In addition, rabbits and humans perceive the binary AC mixture (C: guaïacol) in the elemental way. The two perceptual modes may already be in question at the peripheral level of the olfactory system: indeed, on the olfactory mucosa, receptor neurons are endowed by (few selective) olfactory receptors (ORs) which are in charge to bind odorants; facing mixtures, ORs are the site of odorant-odorant interactions which result in a first integration leading to a more or less important loss of the elemental information. Here, our work starts to explore, in newborn rabbits, whether the peripheral processing of the AB and AC mixtures could account, at least partly, for the perception and behavioural responsiveness to the mixture as a whole (AB) or to the elements composing the mixture (AC). It includes the recordings of electro-olfactograms (EOG) at olfactory turbinates and, in parallel, of sucking-related behavioural responses. The results point that the configural AB mixture is differently processed by ORs compared to the elemental AC mixture: AB evokes larger EOGs than its elements, while AC, A and C evoke EOGs which are similar in amplitude. This suggests synergistic vs. rather hypo-additive interactions between odorants in the case of configural vs. elemental mixtures and, additionally, supports that the two modes involved in odour mixture perception are engaged from the olfactory system periphery in rabbit neonates.

P11
Mother-child bonding is related to body odor perception

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Interpersonal communication is shaped by the – mostly unconscious– perception of body odors. Especially the odor of babies is described as a very pleasant and rewarding experience. Aim of the study was to examine the perception of the baby's odor in mothers with troubled and with normal bonding to their child.
In total 75 mothers and their children (0-12 months of age) were examined; 25 of those were recruited in a day hospital, specialized for mother-child bonding disorders. 50 age-matched healthy women and their children served as controls. All mothers were blindfolded to the odor of their own and stranger's babies; they rated pleasantness, intensity and "wanting" of those odors. Further, they were asked to identify the own baby. In addition, olfactory threshold and identification ability were tested and mothers rated the bonding to their child with a standardized questionnaire.
In result, healthy mothers showed a clear preference of the own compared to a stranger's baby odor, while mothers with troubled bonding did not. In addition, the degree of preference was correlated to the self-reported strength of bonding. Further, mothers with troubled relation could not identify their own baby, while mothers with normal relation could. General ability to smell however was similar in both groups.
We conclude, that dysfunctional bonding is related to specific olfactory abnormalities, which affect the processing of baby odors, but not general olfactory function. It can be assumed, that body odor perception plays an important role in human mother-child bonding.

P12

Bioaccumulation of heavy metals and inhibition of vesicular docking within osn in mudskipper [*Pseudapocryptes lanceolatus*]

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Olfactory neurons can respond to a wide range of chemical cues but are also vulnerable when exposed to various types of pollutants. The cytological consequences of bioaccumulation of heavy metals within olfactory sensory receptor neuron (OSN) has been studied under transmission electron microscope (TEM) attached with x-ray microanalyzer in *Pseudapocryptes lanceolatus* (Bloch and Schneider, 1801) [Anaesthetization: MS-222, dose: 100 – 200 mg./ lit.; Fixation: 2.5% glutaraldehyde and 1% osmium tetroxide in 0.1 (M) phosphate buffer (pH. 7.2 – 7.4) at 4°C for 1hour respectively]. The different morphs of vesicles *viz.,* small vesicles (10nm. – 20nm.), small dense core vesicles (30nm. – 40nm.), pleomorphic vesicles (40nm. – 60nm.), coated vesicles (60nm. – 70nm.), synaptic vesicles (70nm. – 90nm.), *etc.* are identified within the ciliated olfactory sensory receptor neuron (cOSN) [1]. These vesicles are characteristically associated with cytoskeletal structures *i.e.,* microtubules (25nm.) and neurofilaments (10nm.). Microtubules are geometrically arranged with the major subcellular organelles like rER and Golgi apparatus in perinuclear cytoplasm, centrioles at basal body of kinocilia, axial filaments of cilia, *etc.* The topology of neurofilaments is also indicating its position in between the microtubular elements forming cross bridge like cytoskeletal networks for bidirectional transport of vesicular cargo as well as docking at the terminal compartment of cOSN. Bioaccumulation of heavy metals (*i.e.,* copper, iron, nickel, cadmium, lead, *etc.*) causes disintegration of cytoskeletal structure in axoplasm of cOSN that inhibiting transport of vesicular cargo and lead to neural degeneration, *etc.* The detail cytology based x-ray microanalysis of heavy metals in cOSN under TEM may significant for exploring the events of metallobiology of neurodegenerative disorders in fish [2].

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P13

Human body odors and interpersonal relationships

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Social communication is one of the major functions of olfaction, but aspects of this function remain a mystery in humans. Body odors are undoubtedly important in human interpersonal relationships. Their importance shows for example through the serious consequences of olfactory loss on the quality of interactions with the beloved ones. Although the role of human body odor in mate choice and interpersonal relationships has gained scientific interest over recent decades, many aspects remain to be elucidated regarding body odor composition, relevant body odor compounds and effects on human physiology and behavior. A particular

family of body odor compounds, androgen steroids, has been in the focus of many studies, but the legitimacy of such attention is questionable. Other molecules might very well serve as chemosignals in humans. Many compounds undoubtedly remain to be chemically identified. Some others, which have been identified in human sweat, would be worth investigating as candidates, such as those found in sex-specific proportions. We investigated sex differences in the perception of (±)-3-hydroxy-3-methylhexanoic acid and in its effect on person evaluation through face. The precursor of this molecule seems to be present in higher proportions in men's than in women's sweat. Distribution of the studied population (*N*=40) for threshold detection abilities was bimodal, but men and women did not significantly differ on sensitivity, on other perceptual measures (pleasantness, familiarity, irritation, affective feelings) or on verbal descriptions. In spite of this, in another study (*N*=40), having participants smell this odor when evaluating faces with ambiguous gender modulated gender attribution (increased perceived masculinity) in male participants only. The possible role of (±)-3-hydroxy-3-methylhexanoic acid in intrasexual and intersexual relationships will be discussed, and preliminary results of an fMRI study involving this compound will be presented.

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P14
Salicin Preferences in Golden Hamsters (*Mesocricetus auratus*)

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Hamsters may have a T2R similar to the humanT2R-16 GPCR bitter-taste receptor for salicin because hamsters behaviorally generalize the taste of salicin to quinine [1]. Salicin and saccharin concentrations (1-100mM, in half log steps) from near rodent bitter taste thresholds were tested to compare preferences for saccharin [2] and salicin across concentration series. We hypothesized (I) salicin, bitter at high concentrations, will be preferred at low concentrations, but rejected at high concentrations compared to water; (II) saccharin, sweet at low concentrations but bitter at high concentrations, will be preferred at low but not high concentrations, compared to 10mM salicin. Water, 1mM quinine and 100mM sucrose were neutral-, bitter-, sweet-preference controls. 16 hamsters were tested, 8 in each study. They had continuous access to food and were provided the choice between two 12mL centrifuge tubes, switched between left and right side every 24 hours. A new trial with a new pair of substances was conducted after 48 hours. The total experiment ran for 36 days. Percent taste-preference, based on consumption, was calculated as: (mL stimulus ingested ÷ mL total fluid ingested) X 100. Scores of less than 50% indicate avoidance; scores greater than 50% indicate preference. Results showed (1) 73% control preference for water over quinine but 75% control preference for sucrose over water. (2) 1mM and 3 mM salicin were preferred to neutral water as well as to aversive 100mM salicin (t-tests ≤5% significance level). At higher salicin concentrations, beginning at 10mM, preference switched to water. (3) All saccharin concentrations were strongly preferred to 10mM salicin; preference for 100 mM saccharin was greater than for 1mM saccharin (t-test, $p = 0.006$). Conclusions: (1) At low salicin concentrations, 1mM and 3 mM, were preferred to water and may be sweet. (2) Saccharin, preferred over 10 mM salicin at all tested concentrations, was less preferred at the lowest than highest concentration (t-test, $p = 0.006$). (3) Testing saccharin concentrations vs. a constant concentration of purely bitter quinine (as aversive as 10 mM salicin) may further distinguish the 2 bitters.

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P15
Neuronal processing of malodor coverage

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Malodors and their efficient coverage are an important problem in our modern world. The aim of this experiment was to gain insights into the neuronal processing of malodor coverage. In this context, the neuronal correlates of a combination of an unpleasant and a pleasant odor and the mechanisms leading to certain physiological and behavioral responses to such mixtures were investigated using functional magnetic resonance imaging (fMRI). In particular, responses to caproic acid (CAP) as an unpleasant odorant, citral (CIT) as a pleasant odorant, and combinations thereof were evaluated. The odorant mixtures were a solution in which citral completely masked caproic acid (COMP) and a solution in which citral partially masked caproic acid (INCOMP). During the fMRI scan the task of the subject was to evaluate relative quality of the odors. We included 29 healthy and normosmic participants (age ranged from 20 to 36 years). Five participants were excluded from the fMRI analysis due to technical problems. On the behavioral level, we are able to show, that although relative quality ratings of CIT and COMP differed, perceived intensity and pleasantness did not and thus assume that CIT perceptually masked CAP in the complete mixture. On the neuronal activation level, CIT and COMP are both processed in the orbitofrontal cortex (OFC) and the precentral gyrus. Further, perception of CIT specifically activated the inferior occipital gyrus and perception of COMP specifically activated cingulate cortex. To conclude, while hedonic ratings of CIT and COMP did not differ and thus CIT can be used to perceptually mask CAP. Differences in the neuronal network involved in the processing of the odors are evident. Those differences are most evident in typical visual areas as well as areas responsible for attention processing.

P16
Patterns of olfactory receptor gene expression in the zebrafish olfactory epithelium arise from migration of specified olfactory sensory neurons.

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The functional architecture of the olfactory system shows a remarkable degree of similarity across phyla. For instance, individual olfactory sensory neurons (OSNs) express only a single olfactory receptor (OR) from a large genomic repertoire and axons of OSNs expressing the same OR converge onto defined glomeruli in the olfactory bulb. In addition, the pattern of OR expression appears to be non-random within the peripheral olfactory tissue in a variety of species.
In zebrafish, OSNs expressing a particular OR gene have been described to be arranged in partially overlapping concentric expression domains, closely resembling “zonal expression” in the rodent olfactory epithelium (Weth et al., 1996). Here we show that the apparent zonal organization of OR expression in zebrafish is a consequence of migration of functionally specified OSNs and is not an inherent property of molecularly defined zones laid out in the olfactory tissue.
OSNs in zebrafish are generated from two discontinuous proliferation zones located at the central and peripheral edge of the sensory epithelium. Using birthdating of OSNs by incorporation of the proliferation marker BrdU in combination with in situ hybridization for chemoreceptor and cell type-specific marker expression, we find that OSNs reach functional maturity in terms of OR expression as early as two days after they exit mitosis. Notably, OSNs express chemoreceptor genes while they are still in close proximity to their respective birth zone and subsequently invade the sensory tissue. The position of newborn OSNs shifts gradually over time and OSNs of similar age migrate as a coherent band of cells towards the center of the sensory tissue.

We propose a model by which the seemingly structured distribution of OSNs expressing the same chemoreceptor gene is generated by a combination of biased generation of different OSN subtypes, lateral migration across the sensory tissue, and limited lifetime of OSNs.

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P17

Role of the nucleus accumbens in flavor recognition memory: effect of aging and d1/d5 dopamine receptors blockade.

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The attenuation of neophobia to a novel flavor over repeated exposures is associated with changes in its hedonic value as the flavor becomes more palatable. Previous studies have suggested a role of the nucleus accumbens (Nacc) in processing taste palatability which is affected by aging. To explore the role of Nacc in flavor neophobia during aging, we applied c-Fos immunohistochemistry as an index of neural activity of the Nacc Core and Shell. Twenty one adult (5-month-old) and 24 aged (24-month-old) male Wistar rats were exposed to a 3% cider vinegar solution for 1, 2 or 6 consecutive days (*n*=7 adult and *n*=8 aged rats per group). Aged rats exhibited slower attenuation of flavor neophobia than adult rats. This was consistent with the Nacc pattern of c-Fos activity. Adult rats exhibited increased Nacc Shell activity on day 2 compared to days 1 and 6, while this increase was delayed to day 6 in aged rats. Adult rats did not show differences in the number of Nacc Core c-Fos positive cells. However, increased c-Fos expression was found on day 6 compared to days 1 and 2 in the case of aged rats. This suggests that changes in the activity of neural circuits during normal aging could underlie the slower attenuation of flavor neophobia in aged rats.

Due to the increased c-Fos activity found on day two in adult rats, we then explored the implication of D1/D5 dopamine receptors in the Nacc Shell during flavor recognition memory. Forty adult (five-month-old) male Wistar rats were exposed to either water (n=18) or a 3% cider vinegar solution (n=22) for six consecutive days using 15-minute drinking sessions. They received i.c. injections of either vehicle or the D1/D5 receptors antagonist SCH23390 (1µl per hemisphere, quantity: 5µg/µl) into the Nacc Shell 15 minutes prior to the drinking session on day two (coordinates from Bregma: AP=+2mm, ML=±1,1mm and DV=-7,8mm). The behavioral results indicated a neophobic response to the vinegar solution in all the groups. The control group receiving vehicle showed an increased consumption of the vinegar solution on day two, indicating attenuation of flavor neophobia. However, those rats that received SCH23390 did not increase consumption of vinegar on day two. In addition, the groups exposed to water did not differ in consumption regardless the i.c. injection received. These results are consistent with previous reports relating the area with the acquisition of learned taste aversions and support an additional role of the nucleus accumbens shell in flavor recognition memory mediated by D1/D5 dopamine receptors.

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P18

Sweet taste sensitivity relates to salivary leptin and food selection from a buffet meal in humans

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Macronutrient / food intake in humans has been related to variations on bitter and fatty acid taste sensitivities in humans. However, much less is known regarding how sweet taste sensitivity may affect food appetite. The current study aimed at assessing the impact of sweet taste sensitivity on hedonic and satiety responses to soup preloads and on food selection from a buffet meal in healthy human subjects. In a randomized crossover design whereby 30 young adult subjects (aged 24.2 ± 0.9 years, Body mass index: 21.7 ± 0.4; mean ± SEM) were offered one of three iso-energetic preloads consisting of a common soup base differing in taste quality: sweet, umami or no-taste control. A washout period of one week was established between any two test days. Individual sweet sensitivity was assessed using a 9mM sucrose one-solution method. The subjects were separated into two groups named as the sweet sensitive (SS, n=19) and non-sweet sensitive (NSS, n=11). At 60 min after the preload consumption, subjects were offered an ad libitum buffet which consisted of multiple food items varying in taste (sweet or savoury) and fat content (high or low). Subjective measures included appetite and satiety ratings (sensory specific satiety –SSS-). In addition, salivary leptin concentration was measured at arrival. Compared to NSS, the SS cohort had lower salivary leptin concentrations ($p < 0.001$), weaker umami SSS after consuming umami tasting soup and stronger sweet SSS after the sweet soup ($p < 0.05$). In addition, SS consumed more protein and fat, but less carbohydrate (expressed as percentage of total energy) from the buffet than NSS ($p < 0.01$), which contributed to the higher total energy intakes in the SS group ($p < 0.01$). In conclusion, SS subjects had lower salivary leptin concentrations and consumed more energy, as well as more fat and protein, but less carbohydrate from the buffet meal than the NSS subjects. This increase in fat and energy consumption over time could contribute to a positive energy balance and pose a risk of weight gain. The study was registered at anzctr.org.au as ACTRN12615001129572

P19

A generic microfluidic biosensor of G protein-coupled receptor activation

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During consumption of food and drinks by humans, signalling processes are triggered by a specific activation of G-protein coupled receptors (GPCR). This project aims to mimic on a chip the sensory capability of human smell and taste receptors and evaluate their activation by the wide range of ligands present in food products and plants. To reach this goal we functionally express taste and smell receptors in human cells. Cells expressing taste receptors are assembled into a flowcell and attached to a pump system which is mounted on a fluorescent microscope for analysis. This creates a microfluidic biosensor that enables direct detection of GPCR activation by monitoring dynamics in cytosolic calcium ion concentration using a genetically encoded calcium ion sensor. The biosensor platform is generic for all G-protein coupled receptors which use calcium as second messenger in their signalling pathway. Expressing taste and/or smell receptors generates a biosensor capable of determining taste aspects of food products or purified food/plant compounds. Finding novel ligands will improve our understanding of the complex sensory mechanism of taste and smell, and opens possibilities for new developments in food and agricultural sectors.

Reference: Roelse et al., A generic microfluidic biosensor of G protein-coupled receptor activation-monitoring cytoplasmic [Ca(2+)] changes in human HEK293 cells. Biosensors & Bioelectronics. 2013;47:436–444.

P20
How does Saliva Secretion and Composition Respond to Trigeminally Active Compounds?

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Identifying compounds that increase salivary flow and modify salivary composition in a desirable way is relevant for the development of treatments for chronic dry mouth (xerostomia). Flavour compounds in the oral cavity can interact with chemosensory receptors to cause reflex salivation, increase salivary secretion and potentially modify saliva composition. One group of flavour compounds can interact with Transient Receptor Potential (TRP) channels which are expressed in the oral cavity in a number of cell types. In the present study TRP agonist-TRP channel interactions in the oral cavity and the effects on salivation were investigated.

Healthy volunteers rinsed for 30 sec with 10 mL of different mouth rinses containing a TRP agonist - menthol, cinnamaldehyde or nonivamide. Either whole mouth saliva (WMS) or separate parotid gland and non-parotid gland salivas were collected following the mouth rinse and flow rates determined. The qualitative and quantitative protein composition of collected salivas were analysed using SDS-PAGE followed by protein staining and western blotting, and quantitative proteomics respectively. Intracellular calcium signaling in TRP channel expressing CHO cells and in salivary gland epithelial SMG-C6 cells in response to TRP agonists were assayed using Ca^{2+}-imaging.

One ppm nonivamide (TRPV1 agonist) and 500 ppm menthol (TRPM8 agonist) were demonstrated to increase salivary flow rate for 1 min after a 30s mouth rinse, whereas cinnamaldehyde (TRPA1 agonist) failed to elicit an increase even at high concentrations (300 ppm) (contradiction with FOP-abstract). Changes in salivary protein composition after the different mouth rinses were demonstrated. The tested compounds showed multiple interactions with the TRP channels investigated in this study.

It can be concluded that nonivamide and menthol but not cinnamaldehyde can illicit salivary secretion. The protein composition of saliva changes in response to nonivamide mouth rinse. The intracellular calcium assays showed multiple TRP agonist-TRP channel interactions.

P21
Motility of developing taste bud cells.

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We have recently shown (1) that developing taste bud cells including intermediate progenitors (expressing an fgf8a enhancer) and taste receptor cells (Type II) are displaced in a confined, random or directed mode relative to the 5-HT cell. All these types of displacement are observed during the period of organ assembly, in cells within the taste bud organ, cells within the epithelium that join a taste bud and cells that are displaced from one organ to another.

A distinct case is that of intermediate progenitors (expressing an fgf8a enhancer), located in the oropharyngeal epithelium which join taste bud organs in a directed mode (migration) and are maintained within the organ. In this case, *ascl1a* activity and the 5-HT cell

are required for the directed motility of intermediate progenitor cells and their maintenance within the taste bud, respectively.

We would like to understand the 'why and how' of the variety in cell motility during taste bud assembly. We are currently developing tools to address hypotheses on candidate molecules involved in displacement and the functional significance of diversity in motility of taste bud cells.

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P22

Analysis of Statistics and Semantic Relations of Odor-Describing Words in Written Olfactory Versus Non-Olfactory Contexts

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In comparison to the performance in visual object identification tasks, humans gravely underperform when it comes to naming odors. The poor ability in humans to identify olfactory stimuli has since long been established in psychophysical research; yet, the root cause of this peculiar shortcoming remains essentially unknown. Two primary explanations have been hypothesized: The first posits that poor odor naming is a consequence of neuroanatomical constraints limiting the sensory processing ability of cortical olfactory systems as well as their communication with cortical regions responsible for lexical and semantic representations. In contrast, the second hypothesis proposes that inability to name odors is caused by a mixture of social, cultural, and linguistic factors, whereby humans fail to learn strong and well-defined odor-word associations due to a lack of sufficiently odor-specific lexical labels combined with a negligence of accurate and consistent odor descriptions in everyday written and verbal communication. In this study, we attempt to disentangle and quantify the premise of the latter hypothesis. By applying computational linguistic techniques for semantic content analysis on a corpus of tens of millions of documents published online on a wide variety of topics, we quantify the semantic content, semantic similarity and usage frequency of a set of odor-descriptor words used in a previous psychophysical study to classify odors (Dravnieks, 1985). Crucially, we disambiguate between the semantic content in olfactory and non-olfactory contexts, allowing for an estimation of the semantic ambiguity (number of different meanings attributed to the word), olfactory ambiguity (number of types of smells related to the word), commonness (relative frequency in all contexts), and odor applicability (relative frequency in olfactory contexts) of the odor descriptors. These metrics are compared to the applicability values of the descriptors as reported in Dravnieks' dataset (1985).

P23

Thalamic-amygdaloid circuit and aging influence in gustatory memory.

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Habituation of taste neophobia is defined as a reduction of the initial neophobic response as a novel taste is presented in repeated exposures while it becomes familiar. This process evidences the formation of a safe taste memory. Previous data pointed to the implication of the gustatory thalamus and of the amygdala in the habituation of taste neophobia. In addition,

it has been proposed a potential role for the non-relay intralaminar thalamic nuclei in codifying salient events related with feeding behaviour. Among these nuclei the central medial thalamic nucleus (CM) has projections to different amygdala nuclei, including the central nucleus (CeA). In order to explore a potential role of the thalamic-amygdaloid connection in the habituation of taste neophobia, we examined the activity of the CM and the CeA in male adult Wistar rats while they were drinking a novel vinegar solution (3%) the first (Novel), second (Familiar I) or the sixth day (Familiar II). Given the fact that previous reports indicated that aging modifies the pattern of neural activity during taste memory, we also assessed aged (24-month-old) Wistar rats. Both age groups exhibited habituation of vinegar neophobia although it was delayed in old rats. In adult rats both CM and CeA exhibited an increase in Fos-like immunoreactivity after the consumption of the most familiar solution (Familiar II), indicating the same pattern of activation. These results support the involvement of both CM and CeA in the habituation of taste neophobia. In contrast, the CM-CeA taste familiarity increase was absent in aged rats. Also, aging was related with changes in the taste-induced regional CeA activity pattern. In all, the similar pattern of activation might indicate that a thalamic-amygdaloid circuit, which is modulated by aging, underlies the long-term maintenance of a safe taste memory.

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P24
Induced Pluripotent Stem Cell Derived Olfactory Receptor Neurons

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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.

P25
Russian Smell Identification Test

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Olfactory dysfunction can severely affect a person's quality of life, lead to poor nutritional choices and even poses a safety concern. Moreover, development of several serious neurodegenerative disorders, such as Parkinson's and Alzheimer disease, are hallmarked by a decline in individual's ability to smell and identify the odors at the early stages of the disease. Thus, evaluation of the olfactory function has been proposed as a method that may help to diagnose these disorders early, at the stages when the medical intervention is the most effective. However, up to this day no standardized test for evaluation of olfactory function in Russian population has been developed. To rectify this oversight, we conducted a study aimed at adapting The University of Pennsylvania Smell Identification Test (UPSIT) for the population of Central Russia. UPSIT is a widely used scratch and sniff odor identification test initially developed by Richard Doty and colleagues targeting US population. Subsequently the test was successfully modified to be used in several other populations, Italy and Brazil among others, adjusting for cultural differences in familiarity with certain odors and smells.
Our study sample included over two hundred people (17-83 years old, median age of 24.5 years) living either in a big city or in a small rural town in Central Russia. Median test score in the group of participants aged 17-59 years (n=172) was 34 out of 40 items (5^{th}percentile - 28). These numbers are in line with the original test criteria for inclusion of odor items. Less than 50% of the test subjects correctly identified five odors: "lime", "fruit punch", "lilac", "cheddar" and "grass". The most frequent alternate for "fruit punch" was "soap" (95% of respondents) which allowed us to rename the odorant to "soap" not excluding it from the test. To preserve the original number of test items, seven odor samples were suggested as potential substitutes by UPSIT manufacturer and were tested in 86 adult subjects from different age groups and places of residence. "Garlic", "grapefruit", "rubber tire" and coffee" odor samples were selected to replace unidentifiable odorants based on the threshold of 75% correct responses. Our data demonstrate that the modified version of the UPSIT could be successfully used for the assessment of olfactory function in Central Russia population. Further studies will be aimed at collecting normative data on the adapted version of the UPSIT and assessing its validity for early diagnostics of neurodegenerative disorders in Russian Population.
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P26
How do we use olfactory information effectively? -from behavioral experiment and brain imaging-

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Needless to say, the ability of olfactory perception will be improved by training. It is, however, still unclear which training method is better than others. In this study, we explored whether the performance of olfactory three-point identification test was changed by giving two types olfactory hints. And also, rCBF was measured by fMRI during these tasks.

We prepared three flavor components by bubbling soy source, kombu (kelp) soup stock and bonito soup stock. These flavors are frequently used in Japanese cuisine, therefore were familiar to all participants. We prepared two flavor samples A and B. A was composed 2L/min. soy source and 4L/min bonito soup stock bubbling. B was composed 1L/min. soy source, 2L/min bonito soup stock and 3L/min kombu soup stock bubbling.

Participants (n=46) were instructed to select one sample different from other two samples for seven trials as former session. And after this, they were presented hint flavor three times. Kombu stock flavor was presented as hint flavor into half participants (target focus condition), and soy source flavor was presented into another half participants (residual focus condition). After these presentations, we repeated seven trials as latter session. Before each trial, we presented hint flavor in latter session. EPI (TR=3s) were acquired during all these sessions, in MRI magnet.

Correct answer rate increased comparing former and latter session in 19 participants, no change in 10, and decreased in 17 participants. Participants under target focus condition were included in correct answer increasing group significantly, than decreasing one, which implied additional information (hint) will not always improve the olfactory performance.

EPI analysis performed comparing former session and latter one, using performance increasing group (In) and no change group (No). There was no difference in frontal lobe, between “In” and “No” group. There found, on the other hand, differential activation in occipital area (including visual cortices) in only “No” group, and found difference in posterior cingulate gyrus in only “In” group. These results showed that different strategy was performed between “In” and “No” group. Precise analysis will be performed from now on.

P27

Distinct stem / progenitor cell populations contribute to two different modes of neurogenesis in the adult zebrafish olfactory epithelium.

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Regeneration and replacement of lost neurons is a limited process in the post-developmental nervous system. The peripheral olfactory epithelium (OE) is a curious exception to this limitation as olfactory sensory neurons have a limited life span and are constantly replaced to maintain a sense of smell. In addition to maintenance of the neuronal population by perpetual neurogenesis, the OE is also capable of mounting efficient regenerative responses to acute injury. There is growing evidence that different stem / progenitor cell populations and molecular mechanisms contribute to neurogenesis under maintenance and repair conditions (e.g. Leung et al., 2007).

We use chemical ablation of the OE by nasal irrigation with Triton X-100 (Iqbal and Byrd-Jacobs, 2010) in zebrafish to examine differences and similarities between these two modes of neurogenesis. Using the proliferation marker BrdU we find that maintenance neurogenesis in unperturbed OE tissue is provided by proliferative fields at the center and the peripheral edge of the OE flanking the inner sensory tissue. In contrast, damage of sensory neurons by Triton X-100 treatment induces neurogenic proliferation throughout the sensory region of the OE. Using immunohistochemistry against the pan-neuronal marker HuC/D following Triton X-100-induced degeneration of the OE, we observe that the neuronal population declines to about 12 ± 9 % of its original value by 24h and that it recovers morphologically and quantitatively by 7 days following the insult. Double marker incorporation before and after chemical damage suggests that distinct stem / progenitor cell populations located at the periphery of and within the sensory aspect of the OE contribute to neurogenesis under maintenance and repair conditions, respectively. However, the nature and organization of these stem / progenitor cell populations in the zebrafish OE are elusive.

As a first approach to characterize non-neuronal cells in the zebrafish OE, including potential stem / progenitor cell populations, we employed immunohistochemistry and in situ hybridization against candidate stem cell markers, such as Sox2 and ΔNP63 that have been described in the rodent OE to selectively label globose and horizontal basal cells, respectively. Sox2- and ΔNP63-immunoreactive cells can be identified as continuous bands of cells throughout the basal OE. Most, if not all, Sox2-positive cells also label positive for the sustentacular cell marker cytokeratin II. Interestingly, Sox2-positive cells show proliferative activity at the central and peripheral edge of the OE in unperturbed tissue and throughout the sensory region of the OE upon Triton X-100- induced regeneration. This observation raises the hypothesis that a subpopulation of sustentacular glia cells in the zebrafish OE maintains stem / progenitor cell properties and actively contributes to neurogenesis.

To gain insight into the differential regulation of regenerative neurogenesis, we used RNA-seq transcriptome profiling over the time course of de- and regeneration to identify signaling pathways that may selectively regulate stem / progenitor cell proliferation following tissue injury. Among those, wnt-, egf-, and ΔNP63-regulated pathways appear to be significantly upregulated at time points that coincide with increased proliferative activity. We are currently verifying the contribution of the identified signaling pathways by functional assays using specific agonists and antagonists. As a preliminary result, induction of the wnt pathway using systemic or nasal administration of LiCl induces proliferative activity in the OE that closely resembles the pattern of proliferation observed upon tissue damage.

Acknowledgements
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P28

Cross-fostering of Mus musculus and M. spicilegus: Effect on response to conspecific odors

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Results of investigations concerned of preferences of con- and heterospecific odors of cross-fostering house mice are contradictory. According to some authors male and female mice, weaned by females of other species of rodents prefer odor of individuals of cross-fostered species in comparison with conspecific odor (Quadagno, Banks, 1970; Sokolov, Kotenkova, 1987). According to other authors response to the odor of con- and heterospecifics does not change (Kirchhof-Glazier, 1979; Wuensh, 1992). To determine the role of maternal environment in the development of adult olfactory preference, pups of two species of mice (Mus musculus and M. spicilegus) were reciprocally cross-fostered shortly after birth. At 30 days of age all pups were weaned and isolated in individual plastic cages. At 2 months of age all mice were tested for response to con- and heterospecific urine odors in two-choice tests (in their home cages by introducing two 35 mm Petri dishes as sources of odors). Mice of control (non-fostered) groups investigated urine odors of conspecifics significantly longer in all presented combinations. Cross-fostered M. musculus and M. spicilegus showed increased preference for heterospecific odor (M. musculus weaned at 6 days of age) or showed no preference (M. musculus weaned at 3-4 days of age and all M. spicilegus regardless of weaning age). These results suggest that adult species-specific odor preferences are learned during the neonatal period. We compare these results with those of other authors. In the future, we plan to explore the significance of different forms of learning modifying response to con- and heterospecific odors and neuronal activity of some regions of brain in control and cross-fostered individuals. Research were supported by the grant of the Russian Science Foundation (project №16-14-10269).

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P29

Divergence of the Olfactory Signals in Subspecies of the House Mouse Mus musculus

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The differences of olfactory signals in forms at early stages of divergence may be the firs step of the development of their reproductive isolation. In many rodent species, including house mice, olfactory signals play the main role in various types of behavior. Individuals of allopatric, parapatric, and sympatric species of house mice Mus musculus s.l. species group can distinguish odors of con- and heterospecifics of closely related taxa. (Kotenkova et al., 1989; Christophe and Baudoin, 1998; Kotenkova and Naidenko, 1999). The aim of our study was to evaluate the divergence in olfactory signals related to the mechanisms of precopulatory reproductive isolation in three subspecies of M. musculus (Mus musculus musculus, M. m. wagneri, M .m. gansuensis). In the first experiment males of three subspecies investigated urine odor of estrus and anestrus females in two-choice test. To confirm the data obtained with this method and to interpret the results, samples mouse urine were presented to humans, who were used as odor detectors. Our results (from experiments with both mice and humans) have shown that *wagneri* urine odor different from the urine odor of the other two subspecies. To compare neuronal activation in MOB and AOB MRI (magnetic resonance imaging) namely MEMRI (manganese-enhanced magnetic resonance imaging) method was used. Male mice S57B/6J (M.domesticus) were exposed to the odor of estrus and anestrus female S57B/6J, M.m.musculus and M.m.wagneri. The odor of urine of conspecific estrus or anestus female (M.domesticus) caused greater activation in AOB than the odor of closely related subspecies (M. musuclus). Differences of neuronal activation in the MOB were less pronounced. The least pronounced activation in the olfactory bulbs was observed in response to the odor of urine female M. m. wagneri. Our results confirm the divergence of olfactory cues in subspecies of M. musculus. In previous studies we demonstrated c-fos expression in AOB in two sympatric species *M. domesticus* and *M. spicilegus* males in response to stimulation with estrous female urine of conspecifics, but we did not observed c-fos expression in AOB in response to stimulation with estrous female urine from heterospecific females. The role of olfactory cues as isolating mechanisms is discussed.

Research were supported by the grant of the Russian Science Foundation (project №16-14-10269).

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P30
Identification of novel natural sweet taste modulating compounds

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Sweet taste is an undeniably attractive sensation for humans and signals foods that provide energy and essential nutrients. As a consequence, sugar is often added to foods to fulfill consumer's demands in taste and satisfaction. In recent times, however, added sugar has come under scrutiny as driver for obesity and diseases such as cardiovascular diseases and diabetes. To reduce the content of sugar in foods while retaining their appealing sweet taste without off-notes, addition of non-caloric and preferentially natural compounds that enhance the sweet perception of low amounts of sugar might prove an ideal solution.
Here, we report the identification of a class of plant-derived compounds exhibiting a significant and pleasant enhancement of the sweet taste of sucrose. Based on a combination of bioprofiling together with structural homology data analyses of our natural product library NatPure, a small targeted library of natural compounds was tested on our proprietary cellular Novel Sweet Taste Modifier Platform ("NSweeT"), which was genetically engineered to endogenously express the human sweet taste receptor. The natural compounds were assayed for activity as both agonists and/or enhancers of the human sweet taste receptor. In the course of this testing, we identified one compound, IMAX-005681, to specifically enhance the agonist activity of sugars and sweeteners on the sweet taste receptor while not activating the receptor on its own. Subsequently, several chemically related analogues and congeners were also tested on the assay with some found to be active as enhancers as well, thereby identifying a structure-activity relationship comprising a common structural motif. Furthermore, a preliminary taste assessment on IMAX-005681 involving a small human sensory panel was carried out, confirming the enhancement of the sweet taste of sucrose in human subjects.
These results thus emphasize the high potentials of our natural product collections and our novel NSweeT assay platform in the identification of novel natural sweet tasting or sweet taste enhancing compounds.

P31
Search for novel odorant receptor (OR) enhancers in the mouse genome using genetic, epigenetic, and evolutionary signatures

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Function of the mammalian olfactory system depends on specialized olfactory sensory neurons (OSNs) that each express only one allele ("monoallelic") of one odorant receptor (OR) gene ("monogenic"). The lysine-specific demethylase-1 (LSD1) protein removes activating H3K4 or silencing H3K9 methylation marks in a variety of developmental contexts, and is thought to be important for proper OR regulation. Most of the focus in the field has been on a potential "activating" function for LSD1; e.g., in the demethylation of H3K9 associated with the expressed OR allele. Here we show that depletion of LSD1 in an immortalized olfactory-placode-derived cell line (OP6) results in multigenic and multiallelic OR transcription per cell, while not seemingly disrupting the ability of these cells to activate new OR genes during clonal expansion. These results are consistent with LSD1 having a role in silencing additional OR alleles, as opposed to being required for the activation of OR alleles, within the OP6 cellular context.

P32

Lysine-specific demethylase-1 (LSD1) depletion disrupts monogenic and monoallelic odorant receptor (OR) expression in an olfactory neuronal cell line

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Previous work has demonstrated that the expression of a number of Olfactory Receptor genes ("ORs") is regulated by a series of DNA elements termed Locus Control Regions ("LCRs"). As opposed to conventional enhancer elements which up-regulate the expression of a single gene, LCRs are able to trigger the expression of multiple proximal genes in a single locus. Crucially, prior research has shown that knocking out an LCR sequence results in the total loss of expression of a set of proximal ORs *in vivo* (1). In turn, this suggests that the interaction between an LCR with their cognate promoters may be necessary for the establishment of monogenic OR expression from a repertoire of more than 1000 ORs in a given developing mouse neuron. In line with the canonical euchromatic epigenetic signature of enhancer elements, a recent study employed tissue-specific genome-wide H3K4me1 and H3K27ac ChIP-Seq data in conjunction with DNase Hypersensitivity-Seq enrichment to identify 33 candidate LCRs in OR gene clusters (2). In recognition of the vast size of the mouse OR gene family and data suggesting that LCRs regulate only on the order of 10 proximal ORs in *cis*, there are almost certainly more LCRs that can be computationally identified with a more nuanced analysis incorporating a broader range of criteria. An algorithm has been thus designed to combine the aforementioned genome-wide epigenetic data with H3K79me3 ChIP-Seq signal, sequence conservation, and predicted transcription factor binding motifs using a scoring system to best distinguish additional candidate LCRs from non-regulatory intergenic sequence in OR loci. Reassuringly, virtually all of these newly predicted regions preside in clusters that lack a previously determined LCR. Moreover, some of these predicted sequences are located proximal to sets of genes that have similar levels of expression in a clonal cell line population. The identification of new LCRs promises to help better understand the extent to which such *cis* regulatory mechanisms are necessary for the choice of an OR for monogenic expression.

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P33

Pheromones: a response to future sustainable farming
The example of the male effect

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In farm animals, as in the entire animal kingdom, exchange of chemical cues or pheromones play a major role in numerous reproductive behaviours. Indeed, olfactory cues have been shown to influence sexual attraction, estrus induction and indication, puberty acceleration, hormonal stimulation, and libido enhancement. In a changing and challenging agricultural world, the use of such molecules could participate to the development of new breeding management tools in line with a more sustainable farming and a more respectful animal welfare. Easy and fast, without any invasive procedure on the animal, pheromones respond to societal concerns appealing to both farmers and consumers. During the past decades,

efforts were concentrated on the identification of cattle pheromones. Several molecules identified in urine, faeces or saliva of cows have been characterized as sexual pheromones emitted specifically during oestrus. One combination of cow urinary molecules identified in our laboratory enhanced bull libido and semen production (Pherobull® ; Le Danvic *et al.*, 2015).

These researches have attracted the attention of two other livestock sectors, sheep and goat. In these two seasoned species, the introduction of a sexually active male in an anoestrus female herd induces a surge of LH leading to ovulation. This process conventionally used by breeders in their flock, known as "male effect", allows the control reproduction and appears to be also an interesting alternative to the use of hormone. Male effect depended predominantly on olfactory stimuli generated by the male (Okamura & Mori, 2005; Cohen-Tanoudji *et al.*, 1994). If some potentially involved molecules have previously been identified (Cohen-Tanoudji *et al.*, 1994 ; Murata *et al.*, 2014), the complete signal (those inducing ovulation) remains to be clearly characterized. Using an original approach based on the comparison of olfactory profiles of males during non-reproductive and reproductive periods, we undertook the complete “male effect” pheromones characterization in French breeds. First results clearly highlight variations of olfactory profiles during the season. Indeed specific ram and bulk olfactory profiles have been observed during the period of maximal sexual activity with appearance of numerous molecules (ethyl esters, ketones, … - chemical identification still on-going).

If potential chemical cues are identified, more work is needed to validate their biological activity and to find the best way to use them in a farm context. This task proves to be a new exciting challenge. But without doubt, male effect pheromones seem to be a promising response to the limitation of hormone used to synchronize cycle in sheep and goat, which would lead to a more sustainable livestock’s management.

P34

Aroma volatile odorous metabolites at olfactory mucosa level evidenced by *in vitro* and *in vivo* PTR-MS studies

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Olfactory mucosa can metabolize odorants through various enzymatic mechanisms of the Xenobiotic Metabolism Enzymes (XMEs) pathway, participating in odorants clearance and therefore in the termination of the olfactory signal [1]. *Ex-vivo* methodology using headspace-gas chromatography (HS-GC) was developed to measure odorant olfactory metabolism and revealed the formation of volatile metabolites when odorant molecules were injected above a fresh explant of rabbit olfactory mucosa (OM) [2]. Preliminary studies conducted with rat OM afforded the same results. However, this method did not allow accessing the data during the first five minutes of contact between the odorant and the mucosa, thus limiting the olfactory biological significance. Using a direct-injection mass spectrometry technique (Proton Transfer Reaction Mass Spectrometry, PTR-MS) we have been able for the first time to investigate the first moments of the enzymatic process of the metabolic capacity of *ex-vivo* rat OM in real time [3]. The protocol used a discontinuous sampling in headspace vials containing a fresh explant of rat OM in which a known concentration of the volatile ethyl acetate was injected in the gas phase [3].

The current study will present the results of a continuous monitoring obtained by implementing a two-way circuit connected to the PTR-MS instrument that allows direct odorant delivery either above the biological material or in the second branch serving as a control. Injection of ethyl acetate as model odorant above an *ex-vivo* rat OM resulted in immediate apparition of ethanol, the known main volatile metabolite produced by carboxylesterases. Using various food-grade odorous substrates pertaining to the dairy-fatty sensory context (the diketones pentane-2,3-dione, hexane-2,3-dione, hexane-3,4-dione, and 2-acetoxybutanone), we have been able to demonstrate that they can be metabolized by an *ex-vivo* olfactory mucosa within seconds, producing volatile metabolites that have been

identified. Significance for human olfaction has been investigated in an *in vivo* approach combining nasal odorant delivery and nosespace analysis with the PTR-MS instrument. Production of volatile metabolites, confirmed to be odorous in a HS-GC-Olfactometry experiment, will be discussed in terms of their potential impact on overall sensory perception.

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P35
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.

P36
Involvement of TRPA1 in the inhibition of adipogenesis, but not the maturation of 3T3-L1 adipocytes by pellitorine

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Adipose tissue is an important endocrine organ in the human body, however, pathological overgrowth is also associated with chronic illness such as coronary heart disease or type 2 diabetes mellitus. Therefore, regulation of adipogenesis and maturation of adipocytes via bioactive compounds in our daily diet has been in focus of research in the past years. For example, several trigeminally active amides such as piperlonguminine, capsaicin and nonivamide have been demonstrated to modulate lipid accumulation during differentiation and maturation of murine preadipocytes (3T3-L1). Here, we investigated the anti-adipogenic potential of the structurally related and tingling alkamide pellitorine present in *Piper nigrum* in 3T3-L1 cells as a model.
Addition of pellitorine suppressed lipid accumulation, which was assessed by oil red O staining, when applied during differentiation, but also during maturation of 3T3-L1 cells in a concentration range between 1 nM and 1 µM by up to 8.84±4.97% or 7.49±5.08% respectively. In addition, the transient receptor potential channel (TRP) V1 inhibitor *trans-tert*-butyl-cyclohexanol diminished the effects of pellitorine on adipogenesis and maturation of the adipocytes, while addition of AP-18 as inhibitor for TRP A1 only blocked the effect of pellitorine on adipogenesis, but not on maturation. These effects were further investigated on a mechanistic level, suggesting the following signaling pathway for the anti-adipogenic activity of pellitorine: Inhibition of adipogenesis is mediated by TRP V1 and TRP A1, which is associated with a decreased PPARg expression on gene and protein level. PPARg expression may be additionally regulated by increased expression of the micro-RNA mmu-let7b: In contrast, inhibition of lipid accumulation during maturation depends on TRP V1, but not on TRP A1, which was associated with decreased expression of the gene encoding for fatty acid synthase (*FAS*) as well. Down-regulation of *FAS* may be linked to increased expression of micro-RNA 103, as well as to a decreased fatty acid uptake on the functional level.
In summary, these data point to an involvement of TRP A1 in addition to the already identified TRP V1 cation channel in the regulation of adipogenesis by aroma compounds and provide a novel functional role for pellitorine. Since pellitorine does not directly activate TRP V1 and TRP A1, an indirect modulation of the channel activity is assumed and warrants further investigation.

P37
Odor memory game training improves olfaction and memory in older adults

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Ageing is associated with declines in both cognitive and olfactory functions. Moreover, olfactory impairments predict memory decline, subsequent onset of dementia, and morbidity. We investigated whether an odor memory game could improve both olfactory and memory functions in older adults, aged 65-80. Participants were randomly assigned to a memory training intervention, either odor memory or visual memory. Our preliminary results suggest that odor memory training resulted in improvements in odor naming and discrimination, as well as transfer of learning to the visual and verbal tasks. Olfactory-based interventions might be a promising means of improving memory in older adults at risk for memory decline and dementia.

P38
Olfactory attention

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Through their senses humans connect themselves to the environment in both spatial and temporal envelopes. Vision provides a very precise input related to both coordinates. In olfaction, we observe a lack of attention perceiving stimuli exactly in the spatial or temporal envelopes.
The present study aimed to investigate "olfactory attention" and its depending factors, like general attention (d2-R test), environmental hypersensitivity, nasal chemosensory function (tested by "Sniffin' Sticks"), the ability to lateralize odors due to trigeminal chemoreception and individual evaluation of the odorants regarding pleasantness and intensity.
83 healthy participants (aged 18 to 34, 50 women, 33 men) were presented to three different sequences of four odorants, including two pleasant and two unpleasant ones (peach odor in a low and a high concentration, fish odor in a low and a high concentration) in a pulsed mode using an olfactometer. The subjects were requested to press a button when perceiving a change of the odor regarding quality or intensity. All subjects were acoustically and visually shielded. For control purpose, participants were presented to similar sequences of pictures with the same instruction.
In result, change of visual stimuli were detected faster and far more precisely compared to change of olfactory stimuli. Considering the olfactory results, unpleasant odor quality and high odor concentrations facilitated change detection. However, only a few subjects were able to detect the change of odors above chance level. This subgroup showed a significantly better nasal chemosensory sensitivity, a higher individual significance of olfactory function and less environmental hypersensitivity.
In conclusion, human olfactory attention dependent on individual olfactory sensitivity.

P39
Bitter tasting compounds of red wine contribute to the regulation of gastric acid secretion, studied in HGT-1 cells

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In previous studies, we demonstrated that individual food compounds like organic acids in wine (*1*) stimulate gastric acid secretion (GAS). Moreover, red wines were shown to stimulate proton secretion in HGT-1 cells, a well established cell model for studying GAS *in vitro*, more pronounced than white wines (*2*). In this study, we investigated the effect of several bitter tasting compounds in red wine representative concentrations (~0.05 – 500 µM) on proton secretion in HGT-1 cells. Ethyl gallate, Ethyl vanillate, gallic acid and procyanidin B3 stimulated proton secretion, while protocatechuic acid, the hydroxycinnamic acids and epicatechin as well as pentagalloylglucose (PGG) had no effect. Since PGG has been described to be anti-secretory and an inhibitor of the gastric H^+-K^+-ATPase (*3*), it was tested in combination with histamine (1 mM), a natural stimulator of proton secretion. As a result, the bitter tasting PGG inhibited the proton-secretory effect of histamine indicating an involvement of bitter taste receptors (TAS2Rs). Since PGG is an activator of TAS2R4, 5 and 39 (*4*), mRNA expressions of these bitter taste receptors and of genes relevant for GAS were measured in HGT-1 cells using qPCR. After 15 min treatment of HGT-1 cells with the stimulating compounds gallic acid and procyanidin B3, mRNA expression of TAS2R4 was increased (fold change > 1.4) while mRNA expression of genes relevant for GAS was not. A 15 min treatment with PGG resulted in an increase of mRNA expression of TAS2R4, 5 and 39

(fold changes 1.3 – 1.4). Furthermore, co-incubation of HGT-1 cells with PGG and histamine inhibited the histamine-evoked decrease (fold change 0.6 ± 0.4) of TAS2R39 mRNA expression.
These results demonstrate that also bitter compounds, besides organic acids, in red wine regulate proton secretion in HGT-1 cells. Furthermore, an involvement of TAS2R4, 5 and 39 in the regulation of proton secretion is indicated.

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P40

Nicotine-induced Effects on Nicotinic Acetylcholine Receptors (nAChRs), Ca^{2+} and Brain Derived Neurotrophic Factor (BDNF) in Cultured Human Fungiform Taste Papillae (HBO) Cells

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We have previously shown that neural and behavioral responses to nicotine are modulated by nAChR agonists and antagonists (Oliveira-Maia et al. PNAS 106: 1596-1601, 2009; Ren et al. PLOS ONE 10:e0127936, 2015). Accordingly, we investigated the expression and localization of nAChRs in HBO cells using molecular techniques. Using specific human primers for α and β nAChR subunits and RT-PCR, the mRNAs for chrna3, chrna4, chrna5, chrna6, and chrnb4 were detected in HBO cells. In addition, we detected the mRNAs of α-ENaC subunit, β-ENaC subunit, γ-ENaC subunit, T1R1, T1R3, PLCβ2, TRPM5 and T2R38. Western blot experiments confirmed the presence of α4, α5, and β2 nAChR proteins in HBO cells. Immunofluorescence studies showed only a subset of HBO cells bind to α3 and α5 antibodies. In HBO cells loaded with Fura-2, acetylcholine (200 μM) or nicotine (200 μM) induced an increase in cell Ca^{2+}. Treating HBO cells with 0.25, 0.50 and 1.0 μM nicotine for 24h produced a dose-dependent increase in chrnb4 mRNA level without affecting the mRNA levels of chrna5 and chrna6. While treating HBO cells with 50 and 100 mM ethanol for 24h upregulated both chrna6 and chrnb4 mRNA levels in a dose dependent manner with no change in the chrna5 mRNA level. Treating HBO cells deprived of serum overnight with 0.25-1.0 μM nicotine for 30 min at 36°C increased the cellular content of brain-derived neurotrophic factor (BDNF) in a dose-dependent manner. We conclude that HBO cells express several nicotinic acetylcholine receptors and their expression is modulated by acute exposure to nicotine and ethanol. Nicotine at sub-micromolar concentrations increases the expression of BDNF in HBO cells. Thus, nAChRs are involved in the detection of the bitter taste of nicotine and ethanol and also in the synthesis and release of neurohumoral peptides. Supported by NIDCD grant DC-011569 (VL).

P41

Effect of Cyclic-AMP on Postnatal Development of Neural and Behavioral Responses to NaCl in Rats

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In rats, 7-50 days postnatal, NaCl chorda tympani (CT) responses and number of functional apical amiloride-sensitive epithelial Na^+ channels (ENaCs) in salt sensing fungiform (FF) taste receptor cells (TRCs) increase with age. Currently, the intracellular signaling effectors that regulate the postnatal development of mammalian TRC ENaC have not been identified. We investigated the role of cAMP in the postnatal development of NaCl CT responses and behavior in 9-23 day old rats. CT responses were monitored in the absence and presence of cetylpyridinium chloride (a blocker of the amiloride-insensitive NaCl CT response), before and after topical lingual application of 8-chlorophenylthio-cAMP (8-CPT-cAMP; 0-20 mM) for 10-15 min. CT responses were monitored under open-circuit conditions (zero voltage clamp) and under ±60 mV lingual voltage clamp. The data were fitted to an ENaC channel kinetic model. Behavioral responses were tested using 2 bottle 24h NaCl intake tests. Temporal relationship between [deamino-Cys[1], D-Arg[8]]-vasopressin (dDAVP) induced cAMP generation and ENaC subunit trafficking was investigated in cultured adult human fungiform taste cells (HBO cells). Our results show that in 19-23 day old rats, the ENaC-dependent maximum NaCl CT response (r_{asm}) was a saturating sigmoidal function of 8-CPT-cAMP concentration and increased by 2.18 fold. 8-CPT-cAMP also increased the voltage-sensitivity of the NaCl CT response. The response conductance (κ_{as}), defined as the slope of the CT response curve as a function of applied lingual clamp voltage, increased by a factor of 11.7. In HBO cells dDAVP increased intracellular cAMP, and cAMP in turn, increased trafficking of γ-rENaC from cytosolic compartment to the apical pole. Control 19-23 day old rats were indifferent to NaCl, but showed clear preference for appetitive NaCl concentrations after 8-CPT-cAMP treatment. We conclude that an incremental increase in cAMP mimics the postnatal age-dependent increase in TRC ENaC and thus the neural and behavioral responses to NaCl.

P42

Individuals with anosmia demonstrate enhanced performance in a multisensory binding task

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Auditory and visual sensory loss has repeatedly been shown to promote enhanced and compensatory abilities in the respective remaining sensory modality. In contrast, cross-modal compensation in anosmia (olfactory loss) has obtained very little attention. The sparse existing literature focuses mainly on the spared chemical senses, gustation and the trigeminal sense, with mixed results. Given the olfactory sense dependence on heterogeneous sensory cerebral areas, a fact that makes pronounced neural reprograming less plausible, it can be hypothesized that loss of olfactory functions might have *supra*-modal consequences. Specifically, we hypothesize that individuals with olfactory sensory loss, compared to healthy individuals, will demonstrate a more efficient information processing of multimodal stimuli.

To examine this, the perception of audio-visual temporal congruency was assessed in a group of individuals with anosmia and a group of matched, healthy controls. Participants followed a simple flash-beep task, in which the beep was presented either synchronously with the flash or asynchronously, ranging from -300 ms to +300 ms relative to the flash onset. After each trial participants judged whether auditory and visual stimuli were presented simultaneously or not.
Preliminary results indicate that individuals with anosmia compared to controls exhibit a narrowing of the temporal binding window, i.e., a slimmer time window between the onset of beep and flash for which the stimuli are perceived as simultaneous. Specifically, individuals with anosmia are better than healthy controls at detecting smaller asynchronous presentations as such, i.e., are less tolerant to temporal violations. This suggests that individuals with anosmia have developed an enhanced accuracy in judging cross-modal temporal congruency, possibly a multisensory compensational mechanism for the loss of olfactory function.

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P43
Construction of overexpression system of the human bitter taste receptor using a methanol-utilizing yeast *Pichia*.

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Taste plays an important role to identify the chemicals included in the food. A human bitter taste receptors (hTAS2Rs) have seven-transmembrane helixies and belong to the T2R family, which is one of the G-protein-coupled-receptor (GPCR).
To elucidate the bitter taste receptor mechanism, in this study, we tried to construct a overexpression system of the bitter taste receptor hTAS2R14.
For expression of human membrane protein in yeast, a DNA codon usage of the bitter taste receptor,TAS2R14 was optimized for yeast and chemically synthesized. The vector, pPICZαB, containing this TAS2R14 gene was transformed to a methanol-utilizing yeast Pichia and the antibiotics Zeocin resistant transformants were selected.
The transformants were inoculated fresh YPD media containing the appropriate concentration of Zeocin before methanol induction in BMMY media. We tried to confirm the protein expression by SDS-PAGE (coomassie-stained) and Western Blotting.

P44
INCREASE OF MEDIAL PREFRONTAL CORTEX ACTIVATION DURING THE HABITUATION OF FLAVOUR NEOPHOBIA IN ADULT AND AGED RATS

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While previous data has shown that the medial prefrontal cortex (mPFC) activation increases with the repeated presentation of an aversive auditory stimulus, there are no data on the effect of repeated exposure to neophobic flavours in this area. Immunohistochemistry of c-Fos as a marker of neural activity was applied in order to explore the activity of mPFC associated to flavour neophobia and its habituation. Additionally, the effect of aging was explored. Twenty one 5-month-old male Wistar adult rats (n=7 per group) and 24 aged (24-month-old) male Wistar rats (n=8 per group) were exposed to a solution of cider vinegar (3%) and sacrificed 90 minutes after drinking during the first (Novel), second (Familiar1) and sixth (Familiar2) day. The number of c- Fos-positive cells in mPFC was quantified dissociating the

prelimbic area (PRL) and infralimbic (IL) areas. The results showed attenuation of flavour neophobia in both age groups as the intake of the vinegar solution increased over exposure sessions. However, the process was slower in the older group. The overall statistical analysis showed greater number of c-Fos -positive cells in the group Familiar1 compared to the Novel group, regardless of age, in both PRL and IL. The results support a role for the mPFC in the formation of the familiar flavour trace during the habituation of flavour neophobia that is preserved at older ages.
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P45

A *Drosophila melanogaster* glutathione transferase overexpressed in the sensory organs after exposure to bitter molecules in food.

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Glutathione transferases (GSTs) are ubiquitous key detoxification enzymes that catalyze the conjugation of glutathione to a large variety of xenobiotic chemical including odorants and sapid molecules. A previous study in *Drosophila melanogaster* demonstrated the existence of a GST specifically expressed in the antenna and involved in signal termination by catalyzing biotransformation of odorant molecules. Genomic analysis revealed the presence of 40 genes coding GSTs. However, their biochemical properties are poorly documented. Herein, we report that among the GSTs, the isoform dmelGSTD2-2 is strongly and preferentially overexpressed in the sensory organs (antennae, proboscis, legs, and wings) after exposure to bitter molecules. To further understand its physiological role, dmelGSTD2-2 was heterologously expressed then purified using bacteria. Its enzymatic characterization was performed revealing that this enzyme is able to interact and bioconvert bitter compounds. Moreover, we solved (1.5 Å resolution) the crystal structure of dmelGST D2-2 dimer. This is the first resolution of this enzyme subclass. The biochemical function in *Drosophila melanogaster* of this enzyme toward bitter compounds is discussed in the light of the *in vivo* study supplemented by *in silico* binding analysis and enzymatic assay with a panel of bitter molecules.

P46

Differential and sex-dependent manner of the *Mup* genes expression in the reciprocal crosses from CBA/LacSto and C57BL/6J laboratory mice strains

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Major urinary proteins (MUPs) of the house mouse form a large group of highly polymorphic acidic isoforms with molecular masses of 18-20 kDa. MUPs are encoded by the *Mup* gene cluster, which consists of about 35 genes and pseudogenes and is mapped to chromosome 4. Since 1992 and up to now MUPs are considered as a key component of the mouse olfactory signature (fingerprint) which can provide all essential information about individuality of donors (genotype, sex, age, hierarchical status) (e.g. *Bacchini et al., 1992; Böcskei et al., 1992; Churakov et al., 1992; Stopková et al., 2007; Novikov et al., 2009; Kwak et al., 2012; Enk et al., 2016; Sheehan et al., 2016*).

This study was performed on laboratory mice from the reciprocal crosses of C57BL/6 and CBA strains which represent two different mice lineages (*Cheetham et al., 2009*). Using one-dimensional gel electrophoresis we evaluate eight different bands (A-H) which form genotype- and sex-dependent MUPs patterns in parental strains (*Novikov et al., 2009*). There are distinct intersexual differences in the volume of *total urinary proteins* in F1 progeny from reciprocal crosses CBAB6F1 and B6CBAF1. This volume in CBAB6F1 males is 2.3 times higher than in CBAB6F1 females (6.67±0.24 mg/ml vs. 2.85±0.34 mg/ml) and 2.8 times higher in males than in females from the B6CBAF1 hybrids (7.63±0.14 mg/ml vs. 2.69±0.26 mg/ml). However, there are no any significant differences in *total MUPs* content within the same sex between reciprocal hybrids: it is 5.09±0.33 mg/ml and 5.59±0.25 mg/ml for CBAB6F1 and B6CBAF1 male mice, 2.14±0.33 mg/ml and 1.92±0.25 mg/ml for CBAB6F1 and B6CBAF1 female mice, respectively. Moreover, F1 progeny have highly similar MUPs profiles within the same sex: Spearman's rank-order correlation between hybrids of the same sex from reciprocal crosses reveals that for both sexes similarity of MUPs patterns is highly significant: r_s=0.91 in males and r_s=0.98 in females (P<0.01). Concentrations of individual MUP fractions practically do not differ between CBAB6F1 and B6CBAF1 animals of the same sex, except fractions *A* and *D*: their concentrations in B6CBAF1 males are significantly higher than those in CBAB6F1 males. These data suggest that genes which control MUP expression are not linked to the Y chromosome, the trait in overall is not influenced by the mother's organism and exhibits an additive inheritance in F1.

Bearing in mind that C57BL/6 laboratory mice line has predominantly *Mus musculus domesticus'* genome (*Piálek et al., 2007*), the obtained results can provide valuable insight into olfactory mechanisms of sexual recognition and mate choice in feral mice in order to pinpoint concrete *Mup* genes as a testable cause of mate preferences in hybrid zone between two European subspecies – *Mus musculus musculus* and *Mus musculus domesticus* (*Stopková et al., 2007; Mucignat-Caretta et al., 2010; Smadja et al., 2015*).

Key words: CBA and C57BL/6 laboratory mice strains, F1 reciprocal crosses, major urinary proteins (MUPs), MUPs ratio, combinatorial *Mup* gene expression, olfactory signatures

P47

Feeding stimulants in an omnivorous species, crucian carp *Carassius carassius* (Linnaeus 1758)

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Many fish are during feeding dependent on both an olfactory and gustatory sense. Olfaction that acts as the distance sense induces arousal, food search behaviour and attraction to the source, followed by examination of food items by the gustatory sense. During buccal handling the fish decide if the feed will be rejected or swallowed. Amino acids are often stimulatory to the gustatory sense and can act as feeding stimulants. There are, however, inter-species differences concerning what kinds of amino acids act as feeding stimulants or deterrents. The species differences are probably dependent on the natural food choice. As feeding stimulating molecules increase feeding and growth, but deterrents have the reverse effect, it is important to know what kind of molecules have either effect. In the present study we record mouth handling time in the omnivorous crucian carp, *Carassius carassius*, of agar pellets containing water extracts of meal consisting of ordinary food pellets, blue mussels or a commercial carp attractant. These tests were followed by testing with agar pellets with synthetic amino acids, based on the content of the water extracts of the food pellets that was the only feeding stimulant. Neither extracts of mussel meal or of commercial carp attractants had a stimulating effect, i.e. no significant difference in handling time compared to agar pellets with only water. A mixture of five of the major amino acids in the food pellet extract (40 mM alanine, 20 mM glycine, 20 mM arginine, 8 mM serine, 8 mM leucin) gave a significant longer handling time compared to agar pellets with only water. The handling time was also

longer for the three amino acids that had the highest concentrations (40 mM Ala, 20 mM Gly, 20 mM Arg) and finally with only alanine (128 mM). Agar pellets with only Ala gave, however, a significant shorter handling time compared to agar pellets with food pellet extract. The mussel meal extract had the same content of free amino acids and their ranking order was the same as in extracts of food pellets, but at much higher concentrations. Based on the free amino acid content, the mussel extract should have stimulated feeding. This indicates that the mussel extract contained compounds that acted as feeding deterrents in omnivorous crucian carp that do not feed on blue mussels in their natural environment. Previous studies have shown that blue mussel extracts act as feeding stimulants in several bottom feeding carnivorous fish. We finally tested betaine (100 mM) but the molecule had no significant stimulating effect that have been observed in some other fish species.

P48
Sex odour preference in guppy (*Poecilia wingei*) males are influenced by the social environment

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The social environment of animals, particularly in the early stages of life, can have great impact on species-specific and sex-specific behaviours. These changes can be irreversible and continue during the entire life. In the present study we asked the question whether the social environment of male Endler's guppies, *Poecilia wingei*, housed in an all-male community could affect their preference response to female or male odour cues in a flow through Y-maze. After 30 days in an all-male group males were tested for their preference-avoidance responses to conspecific odours. The males were attracted to male-scented water but not to water scented by females. In simultaneous choice between male and female odours they demonstrated no significant preference. The males were attracted to male-scented water after they were kept for 48 hours or 12 days with females. After the Y-maze tests the males´ were placed with two females and their courting behaviour were recorded. The males showed low frequencies of reproductive behaviours. In the all-male group the males had been courting each other. The results show that the social environment influence sexual odour preference and courting behaviour in guppy males.

P49
Multisensory detection of sickness

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Converging evidence suggests that humans have a behavioral repertoire that assists the immune system in the defense against infectious disease. Behavioral detection of subtle and early sickness cues in others, and subsequent avoidance of the infected conspecific, would indeed be a cost-efficient way of coping with an environment fraught with pathogens. That humans can detect early and subtle cues of sickness by way of both olfaction and vision was recently demonstrated. The current study targeted how sickness cues affect social perception

and how these visual and olfactory cues, alone and in unison, activate the brain.

Healthy sample donors were injected with placebo or with endotoxin (lipopolysaccharide, 2.0ng / kg body weight), which induces a transient inflammation-driven sickness response that vanishes within hours. Body odor and facial features were sampled 5 and 2 hrs, respectively, after injection. By presenting facial photographs and body odors of these donors when sick and when healthy (saline injection) to a separate group of participants during fMRI scanning, we could determine the effects of sickness cues on rated "liking" of the presented donors and the underlying neural mechanisms mediating sickness perception.

Results demonstrated that sick faces received lower liking ratings than healthy ones, and that faces, in general, paired with sick body odor tended to be less liked than faces paired with healthy body odor. Moreover, olfactory and visual sickness cues rendered a unique neural processing profile, characterized by activation of the odor perception network and mediodorsal thalamus; as well as activation of face processing areas in superior and middle frontal areas; both in line with threat signal discrimination in the respective modality. Interestingly, multisensory sickness cues elicited superadditive activity, indicative of multisensory integration, in the intraparietal sulcus (IPS), motor cortex, and visual areas. Activity in the IPS to sickness signals where functionally linked to changes in ratings of liking.
We argue that olfaction and vision may be part of a behavioral defense, shaping receiver's neural and behavioral responses to health-threatening interactions. Such early detection of subtle sickness cues, leading up to altered social perception, may have a great significance in limiting the toll of pathogenic disease through avoidance behavior.
Keywords:
Body odor, lipopolysaccharide, endotoxin, smell of sickness, disease detection

P50
Chemosingals of women's fertility affect emotion recognition in single men

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An abundance of studies suggest that there are several detectable changes that increase the attractiveness of women during the ovulation. One significant change that occurs across the menstrual cycle is the change in women's body odor, which is perceived as more attractive by men, when taken from periods of higher fertility. Motivation for reproduction, which rises with exposure to women, specifically at more fertile periods of the menstrual cycle, influences men's behavior in various ways. Thus, it is possible that odors of ovulating women have a role in shaping mating behaviors among men.
Here we investigated whether chemical signals associated with women fertility can influence the ability to accurately identify emotional facial expressions. We first recruited 23 healthy women, not using hormonal contraceptives, who donated two samples of body odor, one from the time of ovulation and one from the luteal phase of the menstrual cycle. Women's samples were mixed together, creating two sets of body odor pools, each for a different experimental condition; low and high fertility odor condition. The mixed pools were presented to 19 single and 21 pair-bond males, while performing the Face Morphing Task, that assess identification of emotional facial expressions of men and women. We found a significant three-way interaction between the odor condition, the relationship status and the gender of the figure in the task. When presented with body odor of the high fertility condition, single men identified more accurately women's emotional facial expressions. This effect was not found among subjects who were in a romantic relationship.
We conclude that while the exposure to women's body odor of high fertility may affect the behavior of single male, it has no effect on pair-bond males. A possible mechanism for these changes is the oxytocinergic system, which is known to regulate pair bonding as well as the perception of emotion in facial expressions. From an evolutionary standpoint, it is possible that increased sensitivity to female's emotional expressions may contribute to detecting and choosing potential mating partners in non-pair-bond males.

P51

A Bacterial Signal Peptide Increases Mucociliary Clearance in Explanted Mouse Trachea

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Bacterial signal peptides are known to trigger innate immunity responses by activation of formyl peptide receptors (FPRs) present in immune cells, e.g. leukocytes. Members of the FPR-family are also found in the murine vomeronasal organ where they are candidates for chemosensory recognition of bacterial pathogens. Here, we investigated the effects of bacterial signal peptides on mucociliary clearance in the murine trachea.

Methods: The trachea of C57Bl6, TRPM5-deficient (transient receptor potential cation channel subfamily M member 5; a crucial component of the canonical bitter and umami taste transduction) and FVB/NCrl mice was explanted and particle transport speed (PTS) was visualized by tracking directed transport of dynabeads at the surface. The transcriptome of single tracheal ciliated and brush cells, a chemosensory epithelial cell type, was analyzed by single cell deep sequencing.

Results: Deep sequencing showed FPR expression in both ciliated and brush cells. The N-formylated bacterial signal peptide FL185 increased PTS from 43.48±5.05 to 75.96±3.56 µm/s (N=8; $p<0.0001$) at 10 µM which addresses FPR1-3. Specific FPR1 and FPR2 inhibitors [cyclosporine H (1 µM) and t-BOC2 (10 µM)] did not reduce the effect. The effect was conserved in FVB/NCrl mice which are lacking a functional FPR3. In contrast, FL185 was ineffective in increasing PTS in TRPM5-deficient mice. Four other tested bacterial signal peptides did not increase PTS.

Conclusion: A bacterial signal peptide stimulates cilia-driven mucociliary clearance, that could represent a novel defense mechanism against invasive bacteria in the trachea. This effect involves elements of the classical taste transduction cascade.

P52

Odor Object-related Activity in PPC Increases with the Number of Stimulated Senses

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Recognizing and categorizing odor objects is a fundamental task in olfactory perception. Recent data suggests that the posterior piriform cortex (PPC), one of the core olfactory areas, plays an important role in the formation of odor objects. It is further known that concurrent multisensory information facilitates and improves the perception of odor objects on a behavioral level. However, whether this beneficial multisensory influence modulates neural processing in core olfactory areas and whether this influence is linear with the number of stimulated sensory modalities is not yet known. In the present study, we investigated the effect of multisensory stimulation on information processing in olfactory areas: the anterior

piriform cortex (APC), the PPC, and the orbitofrontal cortex (OFC). We hypothesized that brain regions encoding odor objects (i.e. PPC) would show a linear activation increase with the number of modalities providing congruent object information. In an event-related fMRI paradigm, participants (N=16) were exposed to uni-, bi-, and trimodal combinations of short video sequences (2s) of familiar objects with congruent sounds and/or odors. We extracted subject-wise activation in regions of interest (APC, PPC, OFC) to investigate linear activation change, driven by the number of involved modalities. We observed that activation in the PPC increased with the number of stimulated sensory modalities while activation in APC and OFC remained constant, independent of the number of modalities. This result suggests that the amount of sensory information provided about one object influences the benefit for odor object formation. Taken together, these results suggest that multisensory signals influence odor object processing in the PPC in a quantity-dependent manner.

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P53
Mosquito olfactory receptors as essential biomimetic odorant sensor for human volatile

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The detection and identification of human odorants is highly demanded in the fields of life rescue and medical diagnosis. Mosquitoes exhibit exquisite sensitivity and selectivity to human odorous stimuli due to their blood-feeding behaviour, they are able to distinguish between different chemical moieties found in human bodies. Mosquito olfactory receptor (ORs) proteins are considered the most important molecular in the processes of olfactory events, and the olfactory system is much simpler than mammalian's due to its independent of G-protein signaling pathway. Therefore, considerable effort has been expended to develop an artificial nose based on the mosquito OR proteins. The present work analyzed the interaction between ORs, human odours, and environmental components in mosquitoes' olfactory system. Furthermore, a biomimetic odorant sensor which expresses these OR genes in living cells is being constructed, in order to detect specific types of human skin volatile. This study provides an alternative way for the construction of odorant sensors, and would promote the development and application of biomimetic odorant sensors in a range of applications such as life rescue and disease diagnosis.

P54
Generation of an iPSC- based Olfactory Receptor Neuron (ORN) culture *in vitro*

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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.

P55
Taste in Parkinson's disease: a four-year prospective evaluation

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It is well-known that Parkinson's disease is characterized by a variety of non-motor symptoms. A gustatory deficit is hypothesized to be one of them although few and only cross-sectional studies are available. The aim of our study was to prospectively investigate the taste function in Parkinson's disease patients after some years from the first evaluation (mean follow-up: 4.35 ± 4.9 years; time range: 3.5-5.6 years). A group of 26 patients was re-examined (M:F=16:10; age range: 54-88 years; mean age: 70.9 ± 8.4 years). Taste function was assessed in one session, by means of the Whole Mouth Test (WMT, Burghart Company, Germany) and Taste Strips Test (TST, Burghart Company, Germany). Olfaction was also evaluated with the Sniffin' Sticks Identification Test (SST, Burghart Company, Germany). All patients were able to understand and complete the procedure.
Although scores decreased over time, no significant difference was found between global taste scores of first and second evaluation (WMT: p=0.234, Mann-Whitney *U*-Test; TST: p=0.747, Mann-Whitney *U*-Test) confirming a persistent slight taste impairment. Our results point to a quite steady condition of this impairment across time. A further statistical analysis regarding every single taste quality is currently ongoing. Considering that the patients' pool re-examined, after a mean disease duration of 9.7 ± 4.9 years, had similar motor evaluation (H&Y=1-3) and a good cognitive status with no impact on daily life activities (MMSE>24), a deep taste impairment could probably appear in the more advanced disease's phase. Future studies on a much larger sample of patients are certainly required.

P56

Odorant-odorant metabolic competitions: *ex vivo* inhibition of the mammary pheromone catabolism

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Lactating female rabbits emit in their milk an odorant, the mammary pheromone (MP; 2-methylbut-2-enal), which elicits orocephalic "searching-grasping" movements in newborns and helps them to find the nipples and suck. Rapid responsiveness to the MP is crucial for the pups since the mother is nursing briefly (5 min) and only once per day. It is then essential for newborns to remain sensitive to the MP during the whole duration of a nursing session in order to optimize milk intake. Therefore, the MP must be efficiently processed by the olfactory system.

In the olfactory perireceptor environment, Odorant Metabolizing Enzymes (OMEs) have the dual function of volatile compounds detoxification and active clearance of odorants from the perireceptor environment to respectively maintain the integrity of the tissues and the sensitivity of the detection. Thus**,** OMEs could modify the bioavailability of odor molecules and influence the olfactory perception**.** The enzymes involved in such modifications are similar to those of the general xenobiotic metabolism for which a competition between the substrates is well known (drug-drug interaction for example). Although emphasized by recent studies, this enzymatic olfactory mechanism is poorly documented in mammals. Here, we (i) used an innovative *ex-vivo* Mass Spectrometry technique to study the kinetics of the MP olfactory metabolism, (ii) explored potential competition between the MP and other odorants toward OME, and (iii) determined if this competition influences newborn rabbits' perception and behavioral responsiveness to the MP. For this purpose, we used Proton Transfer Reaction Mass Spectrometry (PTR-MS), a very sensitive technique for real time online monitoring of volatile compounds, and tested the orocephalic behavioral response usually displayed by newborn rabbits to suck.

The results showed that the MP is metabolized *quasi* instantaneously and that its olfactory metabolism is modified in presence of another odorant competing metabolically in a dose dependent manner. This modification of the MP olfactory metabolism by a competitor has consequences on behavioral responsiveness of pups to the MP. Thus, this odorant-odorant interaction can lead to changes in odorant bioavailability and *in fine* in olfactory perception. OMEs appear as good candidates to interfere with peripheral processing of odorants, inducing such changes in their bioavailability and thus modifying their detection, perception and resulting behaviors. We are currently exploring these mechanisms also in humans.

P57

Variation in TAS2R38 bitter gene: a possible association with feeding behaviour of infants at weaning

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Variation in *TAS3R38* taste receptor gene influences bitter taste sensitivity of children and adults, accounting for individual differences in taste preferences and consequently food selection.

In this study we analyzed the association between three polymorphisms in TAS2R38 gene

(rs1726866, rs713598, rs10246939) and the feeding behaviour of infants at weaning.
The Infant Feeding Questionnaire (IGQ) was collected in 131 new-borns, as well as information on weaning, breastfeeding and anthropometric measures at birth and at six months of age. TAS2R38 polymorphisms were genotyped and infants were classified in three groups: PAV/PAV, PAV/AVI and AVI/AVI. In the statistical analyses AVI/AVI infants were considered "bitter insensitive", while PAV/PAV and PAV/AVI infants "bitter-sensitive".
For the first time a significant association was found between TAS2R38 genotypes and the time required for eat a standard volume of meal (150 ml) (p-value=0.02). While 31% of "bitter-insensitive" infants achieve this aim at the first day, only 13% of "bitter-sensitive" eat all the meal at the first day.
The association was statistically significant also after correction for confounding variables such as gender, type of food supplied at the weaning, age at weaning and breastfeeding.
Therefore, our study indicates that variations in the TAS2R38 gene may influence feeding behaviour of infants at weaning, resulting a much more effective weaning in "bitter-insensitive" infants compared to the others.
Understanding the factors able to modulate the weaning, included the genetic ones, may be of particular interest since food habits in weaning period may mark out food preferences development across the life course.

P58

Genetic variation associated with individual differences in human salty taste perception

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Genetic variations in taste receptors may contribute to individual variability in the perception of different tastes such as bitter, sweet and umami, while little is known on the genetic bases of human salt perception.
In this study salt taste intensity perception was measured on ~ 900 healthy Italian individuals with the labelled magnitude scale (LMS) using a concentration of NaCl 1M. All of the subjects have been genotyped with the 370k Illumina Chip and then imputed using the 1000G SNP set.
The present work report the first Genome Wide Association Study (GWAS) conducted using the log10 of the NaCl intensity ratings and including sex and age as covariates.
Our study confirmed the association between *rs3765964* SNP in *CA6* gene and NaCl perception (p-value=0.018), while no association was found with polymorphisms in SCNN1A, SCNN1B and TRPV1 genes, previously reported in other studies.
Moreover, GWAS detected an association between salt perception and *rs547916* polymorphism, closely located to the KCNA5 gene (p-value=5.6×10^{-08}).
KCNA5 belongs to the delayed rectifier K+ (DRK) channels, already involved in the modulation by a variety of taste stimuli, including acids, sweeteners, bitter stimuli and fatty acids. Moreover, a study has also shown that KCNA5 is the main functional DRK channel expressed in rat taste buds.
Further studies are needed to clarify the biological role of *KCNA5* in salt perception and liking and possible implications for human health, such as the potential involvement in the genesis of hypertension and in the related cardiovascular diseases.

P59

Human diversity and complexity in TAS2R-mediated bitter taste response: from individuals to populations.

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Many inherited traits are obvious and observable within or between populations, such as a blue, green, or brown eye colour. Other traits, although having a direct impact in our daily life, are less apparent. Bitter taste perception drives innate aversive behaviour and is therefore considered to prevent ingestion of numerous bitter-tasting toxic compounds. Yet, individual differences in bitter taste perception exist and arise from genetic functional polymorphisms in *TAS2R* genes, e.g. the long-known genetic predisposition to taste phenylthiocarbamide is driven by the *TAS2R38* gene. Since bitter taste is mediated by approximatively 25 bitter taste receptors (TAS2Rs), characterized by complex molecular receptive ranges with most TAS2Rs recognising multiple bitter compounds and many compounds activating several TAS2Rs, such simple associations of bitterness perception with a single gene are rare.
Therefore, we first investigated systematically the genetic differences in bitter taste response by examining the worldwide genetic diversity in *TAS2R* genes. To this end, we analysed genetic data sets of 2504 individuals from 26 worldwide populations retrieved from the International 1000 Genomes Project (1000G) Phase 3. We identified most *TAS2R* haplotypes, and we functionally characterized allelic variants. Our data reveal the extent of diversity of TAS2R-mediated bitter taste responses and provide evidence for a population-specific genetic structure among the African, Ad Mixed American, European, East Asian, and South Asian populations.
Secondly, to decipher how *TAS2R* genes shape variation in bitter taste perception, we applied, in a sample of the Caucasian population, an integrative approach, by sequencing all *TAS2R* loci, inferring long-range haplotypes, mapping their effects on perception, and characterizing functionally causal allelic variants. Our analyses revealed the complexity of genetically driven perceptual differences in bitter taste, arising from genetic functional polymorphisms and genomic structure.

Keywords: Gustatory perception; bitter taste receptor (TAS2R); genetic polymorphism; genomic structure; genetic population structure.

P60

Some behavioural responses to chemosensory stimuli of wild type and TMEM16A conditional knockout mice

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The TMEM16A Ca^{2+}-activated Cl^- channel is expressed in vomeronasal sensory neurons and is involved in the transduction current in response to stimuli. Conditional TMEM16A knock out (cKO) mice were generated using loxP-Cre system with *Cre recombinase* driven by OMP promoter and were tested for a variety of behavioural responses. Adult mice (n=44) were maintained in single sex/mixed genotype cages and tested over three consecutive days. The

time to descend from a pole was faster in both males and females cKO mice, compared to the WT. When mice had to search for hidden food using their nose, males were faster than females. Moreover, cKO females were slower when food was visible. Mice were then exposed to two pairs of chemosensory stimuli. Adult male urine was used as a stimulus activating the vomeronasal and olfactory epithelium, paired to linalool as a main olfactory stimulus: females were faster than males to approach the stimuli, and gave more sniffs. Interestingly, cKO mice gave more sniffs to both stimuli compared to WT. Moreover, female cKO mice were faster to approach linalool. In the second test, adult male urine was paired to perilla essential oil, that activates the trigeminal chemosensation. All mice approached faster perilla oil than adult male urine. In conclusion, cKO mice appear less frightened in new challenging situations, like in the pole test. They reacted to complex chemosensory stimuli similarly to their WT counterparts, however cKO mice appeared more interested in olfactory/trigeminal stimuli, compared to stimuli that activate also the vomeronasal pathway.

P61
Disease detection: Volatile biomarkers in acute inflammation

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Through history, infectious bacteria and viruses have posed a threat to humanity. Being able to detect and avoid pathogens is, therefore, of crucial importance. It has been shown that body odor samples, such as urine, from immune-activated animals contain sickness cues and detection of which, results in avoidance behavior in conspecifics. Perceivable changes in body odor samples have also, recently, been shown in immune-activated human participants. The main aim of this study was to identify potential volatile biomarkers of the acute inflammatory response. Healthy volunteers were injected twice in a crossover design, once with the bacterial endotoxin lipopolysaccharide (LPS, 2ng/kg bw) and once with placebo (saline). LPS caused a transient systemic inflammatory response as shown by pro-inflammatory cytocines, tympanic temperature and subjective sickness ratings (significant interactions between condition and time with all *p*s<.001, and all $\eta\rho^2$>.663). Axillary sweat and urine were collected both before and 2-4 hours after injection. Headspace from these samples were analyzed using gas chromatography-mass spectrometry (GC-MS). GC-MS data analyses assessed the differences in the profile of volatile compounds of urine and sweat from LPS and placebo donors. Results regarding possible differences between volatile biomarkers in LPS and placebo condition will be presented and discussed.

P62
Describing odorspace

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In the pursuit of connecting odorant physical properties to odorant perceptual properties a fundamental question is how to parametrize the perception of odors. Our work has focused on an unambiguous measure that must be a part of any full model of odor perception: the perceived similarity of odorants. In order to model realistic odor percepts we compared odor mixtures of varying number of components. In earlier work we successfully modeled the

similarity between mixtures of equally intense components. Here we first apply our previously published algorithm to an extensive new data set of odorant similarity tests recently published by Keller, Vosshall and colleagues. We find that our algorithm effectively predicts odorant similarity in this independently collected data set ($r = 0.65$, $p < 0.001$). A limitation of our algorithm is that it assumes odorant mixtures whose components were first diluted such that they were equal in perceived intensity. Such mixtures exist in lab but not in nature. To overcome this limitation and extend our model we used a new set of 45 molecules that were not included in our previous work and this time were not equated for intensity. We used 30 test subjects who rated similarity in 100 comparisons of mixtures. We then extended our similarity model by adding a concentration constant to account for differing component intensities. This modified model effectively predicted perceptual mixture similarities in mixtures made of components with unequal intensity (r=-0.79, p=e-22). It should be noted that the extended model is identical to our previous model when the intensities of the components are equated. We will present these recent results and discuss the related question of modeling odorant intensity itself from molecular structure.

P63

Amniotic liquid of sows is sensitive to dietary supplementation of eugenol, anethol and cinnamaldehyde during late gestation

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Some flavours intrinsically related with the common feed ingredients are known to be transferred in small amounts from the maternal diet to the amniotic fluid and milk, promoting mother rewarding effects on the newborn and weaned pig, a phenomenon known as a pre-natal or perinatal conditioning. It has been reported that perinatal conditioning is determinant to reduce the stress and improve the feed intake in weanling pigs. Therefore, the use of volatile compound not commonly present in sow gestating diets could be a useful strategy for the swine industry to manage and reinforce the perinatal conditioning effect. However, the transfer of some volatiles like anethol, eugenol or cinnamaldehyde rom the maternal diet to the amniotic fluid and milk has not been reported in swine by direct analysis with solid-phase micro-extraction, gas chromatography and mass spectrometry (SPME-GC-MS). The presence of volatile compounds in the amniotic fluid and milk was determined in 10 sows fed a late gestating diet (last month of gestation) differing by flavour inclusion (four sows from the control (C) diet and six from the flavour supplemented diet (F). Fluidarom 1003® (> 25% of anethol and cinnamaldehyde and > 10% of eugenol) a commercially flavored feed additives from Norel S.A., supplemented at 375 ppm was used as source of the main volatiles under study. Farrowing was induced, two days before the expected date of farrowing, by 2 cm^3 of D-cloprostenol (PGF2α) followed 24h later by 2 cm^3 of oxytocin. After approximately 1 hour, farrowing began and the amniotic fluid was collected. Milk samples were collected on d 10 of lactation, after intramuscular oxytocin injection. All of the samples were kept frozen until lab analysis. Amongst other compounds, anethol, cinnamaldehyde and eugenol were detected in amniotic fluid within the most influent compounds differing F vs. C sows but, only traces were observed in milk. There was an increase ($P < 0.01$) in anethol abundance of sows fed the flavor diet compared with sows fed the control diet (7579 vs. 1601 mass number/charge number (m/z)). There was also an increase ($P < 0.001$) for sows fed the flavor diet compared with the control diet for cinnamaldehyde (2376 vs. 1346 m/z) and eugenol (12606 vs. 1209 m/z). The Principal Component (PC) analysis of the volatile compounds showed clear differentiation between C sows and F Sows and also the presence of anethol, Cinnamaldehyde and eugenol was only in the amniotic fluid of sows fed the flavor diet. Therefore the two first PC already explained a 83% of the total variability which indicates that those compounds present in the amniotic fluid clearly differentiate the sows supplemented than those fed the common diet. Therefore it can be concluded that the amniotic fluid of sows is sensitive to dietary anethol, eugenol and cinnamaldehyde inclusion and that could be a useful strategy to manipulate piglet feeding behaviour before and after weaning due to the positive maternal reward.

P64
Chemosensory cholinergic signaling network in the thymic medullary epithelium

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Objective: A subset of medullary epithelial cells in the thymus (mTECs) was previously shown to be cholinergic and to express components of the bitter taste transduction cascade. In this study we set out to further characterize these cells and elucidate their function.

Methods: Immunohistochemistry, real-time RT-PCR and intracellular calcium measurements were conducted on thymi from ChAT- (choline acetyltransferase) and Chrna3-eGFP (nicotinic receptor subunit alpha3) reporter mice, mice expressing diphtheria toxin A driven by TRPM5 promoter (TRPM5: channel in taste transduction signaling), and wild-type mice with streptococcal pneumonia. Newborn human thymi were subjected to immunohistochemistry.

Results: Analysis of murine thymi at different age stages revealed that expression of ChAT and chemosensory components in the mTECs starts at birth but not before. The ChAT-positive cells in the thymus are in proximity to terminally differentiated mTECs (Hassall-like bodies) carrying Chrna3. In human newborn thymus, these cells closely surround or are integrated in the outer layer of the Hassall's corpuscles. Hassall-like bodies were not observed in TRPM5-DTA mice lacking chemosensory cells. These cholinergic cells respond to the bitter substance denatonium with an increase in intracellular calcium concentration. Thymic mRNA expression of TRPM5 and alpha-gustducin was up-regulated in murine model of streptococcal pneumonia.

Conclusion: We hypothesize that the novel chemosensory cholinergic cell type in the thymic medulla senses bacterial products, presumably coming from the bloodstream and responses by release of acetylcholine which in turn stimulates Hassall's corpuscles.

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P65
Functional Overexpression of Vomeronasal Receptors Using a Herpes Simplex Virus type 1 (HSV-1)-Derived Amplicon

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The accessory olfactory system is specialized in processing social interactions such as those conveyed by pheromones, kairomones, and other socially relevant chemosignals. These molecular cues are detected in the vomeronasal organ (VNO) by vomeronasal sensory neurons (VSNs) expressing one of three major families of G protein-coupled receptors (GPCRs), vomeronasal receptors (*VRs*) type 1 and 2 (*V1Rs* and *V2Rs*), and formyl peptide

receptors (*Fprs*). These receptors are implicated to recognize a highly diverse collection of ligands. Matching socially relevant molecules with their specific receptors provides important knowledge about their function and biological relevance. Thus far, only few receptors out of nearly 400 receptor genes, have been matched with specific ligands, in part due to the lack of suitable heterologous expression tools. To overcome this limitation, we developed a herpes virus-based amplicon delivery system to overexpress three types of murine vomeronasal receptor genes in native VSNs and characterized cell responses to their proposed ligands. Through Ca^{2+} imaging in infected cells we show that virus-induced overexpression of *V1rj2, V2r1b* or *Fpr3* caused a pronounced increase of responsivity to sulfated steroids, MHC-binding peptide or the synthetic hexapeptide W-peptide, respectively. Other related ligands were not recognized by infected individual neurons, indicating a high degree of selectivity by the overexpressed receptor. Removal of G-protein signaling eliminates Ca^{2+} responses, indicating that the endogenous second messenger system is essential for observing receptor activation. In summary, our novel expression system for vomeronasal receptors provides a new tool to deorphanize the molecular logic of VNO ligand detection contributing to decipher chemosignal-based mammalian communication.

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P66

Glomeruli of the OR37 subsystem possess a more stable interneuronal network than others

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The olfactory subsystem OR37 is targeted to the paraventricular nucleus (PVN) of the hypothalamus and seems to be involved in social buffering effects under moderately stressful conditions. Due to the pheromone-like character of the system we hypothesized that its neuronal network in the olfactory bulb may be more stable than those for general odorants. Therefore we have analyzed adult-generated interneurons around transgenically labeled glomeruli of mOR37 members in comparison to glomeruli of receptors responding to general odors.

Results of BrdU labelling experiments indicated that at glomeruli of the OR37 subfamily the proportion of adult born interneurons was significantly lower than at glomeruli of mOR256-17 or mOR18-2. This observation was confirmed by immunohistochemical stainings for Doublecortin, a marker of migrating neuroblasts in the olfactory bulb; it was found that also the number of immigrating neuroblasts was lower around OR37 glomeruli compared to other glomeruli. Thus, the proposed role of OR37 odorant receptors in social communication coincides with a more persistent, “hard-wired” functional glomerular domain, whereas the glomerular domains for non-social odorants are characterized by a more fluctuating setting of interneurons indicating a more flexible neuronal network.

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P67

Effects of different isocaloric oral nutrient solutions on olfactory functioning, hunger and food craving.

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Food intake influences human olfaction, hunger and food craving. However, little research has been done in this field to elucidate the effects of different nutrients. Thus, the goal of our study was to investigate the effects of oral ingestions of different nutrient solutions on olfactory functioning, hunger and food craving. 20 healthy men (mean age: 23.8 years, SD: 3.22, BMI: 22.79 Kg/m², SD: 1.44) participated in our study employing a double-blind, cross-over, repeated measurement design. Participants were tested on 4 different study days. Each day participants received one of three isocaloric (carbohydrate or fat or protein, 600 kcal, 1500 ml) or a placebo (NaCl) solution in randomized order. Olfactory and cognitive tests were conducted three times, i.e. 100 min before the beginning of the nutrient intake, following oral ingestion of the nutrient solution and 60, and 240 min after. Craving and metabolic function tests were performed 7 times each examination day (observation period: -100 min, 0=nutrient intake, +60, +120, +180, +240, +340 min).

We tested olfactory functioning by means of the ”Sniffin´ Sticks” test battery comprising a threshold (n-butanol), a discrimination and an identification test. Participants rated odor intensity and hedonics for the odorants of the identification test by means of analog rating scales. We registered ‘hunger’, and ‘food craving’ by means of VAS (visual analogue scales, ranging from -10, to +10, including 0 as a neutral point). Craving was registered following the presentation of five pictures (craving order of pictures: 1. fat-rich food, 2. protein-rich food, 3. carbohydrate-rich food, 4. sweets, 5. vegetables). In addition we monitored cognitive and metabolic functioning using the Tests for Attentional Performance 2.2 (Vera Fimm, Germany) and analyzed specific metabolic parameters.

We found significantly different TDI-scores for the four oral solutions used with lowest olfactory functioning following the solution of fat. Ratings of hunger significantly differed over the observation period with lowest ratings following the solution of protein. Ratings of craving significantly differed over the observation period with lowest craving for fat, protein, carbohydrates, sweet and for vegetables following the solution of proteins. Highest ratings of craving were found following solution of placebo as expected. Estimates of olfactory hedonics and intensity did not differ significantly following nutrient solutions. Our study revealed that the type of nutrients possess a significant influence on olfactory functioning, hunger and food craving.

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P68
Female mouse tears inhibit aggression in males

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The lacrimal fluid of prepupertal and adult male mice is a powerful pheromonal source, eliciting effects on the copulatory behavior of females and modulating male mating behavior and aggressiveness.
Here, we report that the lacrimal fluid of the female mouse, when rubbed onto a male intruder during an intermale aggression test, inhibits aggressiveness and induces mounting behavior in a resident male. Resident male aggressiveness is retained when the rubbed lacrimal fluid was collected from females subjected to ovariectomy or extraorbital gland removal.

P69

Infraslow Intrinsic Rhythmogenesis in a Subset of Mitral Cells Entrains Oscillatory Microcircuits in the Accessory Olfactory Bulb of Mice

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The accessory olfactory system (AOS) is a key component in rodent conspecific chemical communication. Despite its fundamental function, however, sensory coding in the accessory olfactory bulb (AOB), the first stage of information processing in the AOS, is poorly understood. Here, mitral cells (MCs) receive sensory input from peripheral vomeronasal neurons and relay this information to the vomeronasal amygdala and the hypothalamus. Recently, we demonstrated that a subpopulation of mouse MCs is intrinsically rhythmogenic and exhibits slow stereotypical oscillatory discharge triggered by cyclic activation of three interdependent ionic conductances: subthreshold persistent Na^+ current, R-type Ca^{2+} current, and Ca^{2+}-activated big conductance K^+ current. Here, we identify an excitatory circuit within the AOB network that entrains oscillatory activity in a second MC subpopulation. Using a battery of physiological techniques in acute AOB tissue slices we show that entrained MCs display periodically increased excitatory synaptic input that correlates with their respective rhythmic discharge patterns. Several such MCs are often organized into synchronized microcircuits. Ongoing experiments aim to identify the detailed mechanisms of oscillatory entrainment and synchronization, and the role of slow rhythmic activity in AOB information processing.

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P70

Positive allosteric modulation of mosquito olfactory receptor function

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Allostery is an important element of regulation of protein function. It modulates structure-function relationships in complex oligomeric assemblies, such as GPCRs and ion channels, and has significant consequences for drug-related applications. Insect odorant receptors (ORs) constitute a family of ligand-gated ion channels (LGICs) that are not related to other membrane-anchored receptor families including mammalian olfactory receptors, which are members of the G-protein coupled receptor (GPCR) superfamily. They are heteromeric complexes of a variable (ORx) and a conserved (ORco) subunit (ORx/ORco) of as yet unknown stoichiometries. Their study has received much attention both in the context of basic biology and evolution and that of olfaction-based potential applications for insect pest control.
In this presentation, we describe our findings on the positive allosteric modulation exerted on odorant-gated mosquito ORs by ORco agonists. This is demonstrated by heterologously expressing four different *Anopheles gambiae* ORx subunits, along with ORco and the photoprotein Photina, in insect cells and by using a toolbox of specific odorants (full and partial agonists) and ORco agonists. We find that low concentrations of ORco agonists potentiate significantly specific ORx ligand-induced responses, affecting both the efficacy and potency of the ligands. These findings provide new insights into the basic biology of the receptors and may also have practical implications for the development of new reagents for enhancement of insect OR responses.

P71
Comparison of research methods for functional characterization of insect olfactory receptors

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Insect olfactory receptors (ORs) in the peripheral olfactory system play an important role detecting elements of information from the environment. The identification of the OR repertoire is the first step towards understanding how the insect can integrate and process the huge diversity of volatile compounds present in their environment, including signals of food, enemies and mates. At present, various approaches are being adopted for deorphanization of ORs in insect. In this study, we compared methods for functional analysis of ORs *in vitro* and *in vivo* taking the candidate pheromone receptor OR13 of *Helicoverpa assulta* (HassOR13) as the object of our experiments. We found that the natural system was more sensitive than those utilizing transgenic *Drosophila.* The two-electrode voltage-clamp recording is more suitable for functional screening of large numbers of ORs, while the *in vivo* transgenic *Drosophila* system could prove more accurate to further validate the function of a specific OR. We also found that, among the different solvents used to dissolve pheromones and odorants, hexane offered good reproducibility and high sensitivity. Finally, the function of ORs was indirectly confirmed in transgenic *Drosophila*, showing that odor-activation of ORs-expressing olfactory receptor neurons (ORNs) can mediate behavioral choices. In summary, our results compare advantages and drawbacks of different approaches, thus helping in the choice of the method most suitable, in each specific situation, for deorphanizing insect ORs.

P72
Effects of Sex and Sexual Preference on Olfactory Awareness

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The Olfactory Awareness Survey (OAS; Smeets et al., 2008) measures the relevance of olfactory cues in daily life. Although the sex differences observed in both olfactory sensation (e.g. Navarrete-Palacios et al., 2003) and in the value of olfactory information in mate selection (Herz & Inzlicht, 2002; White & Cunningham, 2015) might lead to the suspicion that the sexes differ in odor awareness, findings from the OAS vary considerably in this matter (e.g., Dematté et al, 2011; Sodavari et al., 2015, Nováková et al., 2014). The present study explored the possibility that sexual orientation may have acted as an intervening variable that contributed to the diversity of findings by presenting the OAS to 453 individuals: 142 heterosexual women, 161 heterosexual men, and 150 gay men. A one-way ANOVA conducted on the positive items of the OAS showed a difference between the groups [$F(2, 450) = 3.31$, $p = .04$] that reflected differences between heterosexual and gay men, $t(289) = -2.50$, $p = .02$. Analysis of the negative items of the OAS with a one-way ANOVA also indicated a difference between the groups, $F(2, 450) = 3.49$, $p = .03$. Follow-up tests indicated that heterosexual men differed on the negative items from both heterosexual women [$t(299.6) = -2.40$, $p = .01$] and gay men [$t(289.96) = -2.15$, $p = .03$]. These findings indicate that heterosexual men are less aware of the negative aspects of olfactory stimuli than are gay men and heterosexual women; heterosexual men are also less aware of the positive aspects of odorants in their environment than gay men. This is in keeping with previous findings that heterosexual men value odors in their environment less than other groups. *Funding for this project was provided by the Stipend Funds of the Department of Psychology at Le Moyne College.*

P73
Are there ERP correlates of olfactory and visual mental imagery?

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The ability to create mental images has been studied in various modalities (visual, auditory, olfactory, tactile etc.) and using different techniques such as subjective vividness measures, functional magnetic resonance and positron emission tomography. Sensory areas involved in perception of stimuli have been shown to be involved in mental imagery for both visual and olfactory imagery (McNorgan, 2012). We were interested in studying the temporal dynamics of mental imagery. We used electroencephalography to measure the activity connected to mental imagery of smells and objects. Additionally, we manipulated participants' ability to image odors by requesting them to wear a nose clip during half of the experiment. Our study corroborates previous findings showing that odors are more difficult to imaging than visual objects, especially if the nose is blocked (Arshamian et al.,2008). Interesting, our results indicate a different relationship between the amplitude of event related potentials and vividness of imagined smells vs imagined objects.

P74
The surrounding host plant volatiles can enhance the sex pheromones recognition of green plant bug, Apolygus lucorum

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At present, it is an intractable problem that the synthetic sex pheromones lure of green plant bug *Apolygus lucorum* with high biological attraction in the field does not work indoors We assumed that there were interactions between the identification of sex pheromones in *A. lucorum* and their perception of the surrounding host plant volatiles. The initial behavior assays showed that the synthetic sex pheromones could not attract the virgin male bugs, even the sex pheromones plus cotton volatiles still had no attraction to virgin male *A.lucorum*. After feeding on cotton leaves, the virgin males showed significant taxis selection to sex pheromone plus cotton volatiles, while the lure without cotton volatile did not work. In the following parallel tests, we found that virgin males which were fed on favorite host plants had strong tendency to sex pheromone plus cotton volatile, whereas after feeding on green beans and maize, male bugs showed no obvious taxis response to sex pheromone plus cotton volatiles. The results suggested that the host plant volatiles may play a key messenger role in the course of *A.lucorum* males searching for spouse. In the long process of adaptation to natural environment, perceiving the surrounding host plants become the precondition and foundation for *A. lucorum* to mate and breed population.

P75
Olfactory dysfunction in athletes following moderate and severe head injury: a possible cut-off from normality to pathology.

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Traumatic brain injury (TBI) is one of the most common causes (approximately 20%) of olfactory impairment. Suggested underlying mechanism accounting for complete or partial loss of olfactory function include olfactory nerve fiber injury, nasal bones and/or skull-base

fractures and hemorrhage-related damage in olfactory brain areas. Sports players are at an increased risk of such injuries. Here we report on olfactory function (identification and detection threshold) in 9 sports players (3 case studies and 6 matched controls) who suffered moderate (2) and severe (1) concussion during motocross, rugby and football practice. We investigated the relationship between severity of head trauma and olfactory loss and whether the stage in between moderate and severe head trauma can be identified as a cut-off from olfactory normality to pathology. Results supported this notion showing that only the participant with a previous severe TBI performed significantly worse than controls and moderate TBI subjects.
Our outcomes stress the importance of an olfactory assessment, along with a general neuropsychological evaluation, in case of sport injuries.

P76

Spontaneous Firing Pattern in Mouse Vomeronasal Sensory Neurons lacking TMEM16A Channel.

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The Vomeronasal Sensory Neurons (VSNs) use a PLC mediated cascade to transduce the chemosensory signals leading to increase of intracellular Ca^{2+} mainly through the activation o of TRPC2 channels. Moreover, it has been shown that VSNs express two members of the calcium-activated chloride channels TMEM16A and TMEM16B. Previous study performed by our group found that the specific deletion of TMEM16A channel in VSNs abolished the calcium activated chloride currents without affecting the expression of other components of the transduction machinery. Here, we investigate the basic electrophysiological properties and the spontaneous activity of the VSNs from TMEM16A conditional knockout (cKO) mice compared with WT mice.
Whole-cell recordings both in voltage and current-clamp mode showed that the voltage-activated inward and outward currents, the input resistance of the membrane, resting membrane potential and excitability of the membrane after current steps application were not statistically significant different in cKO animal. Then we measured the spontaneous activity of VNSs by loose-patch recordings. The mean frequency of spontaneous activity was not different between WT and TMEM16A cKO mice. However, calculating the Interspike Interval distribution (ISI) we found that VSN from WT mice showed a narrow distribution of firing between 50 and 200 ms (between 5 and 20 Hz), with around 78% of spikes presents in that interval. In contrast in VSNs from TMEM16A cKO mice the percentage of spikes that felt in this range was reduced to about 50%. These data suggested that TMEM16A channels do not play a critical role in resting membrane conditions in VSNs. However, it was shown that the specific deletion of TMEM16A channels from VSNs altered the spontaneous firing pattern leading to a broader distribution of the firing without affecting the mean frequency revealing a role of calcium-activated chloride channels in the regulation of firing pattern in VSNs.

P77

Elimination of a ligand gating site generates a supersensitive olfactory receptor

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Olfaction poses one of the most complex ligand-receptor matching problems in biology due to the unparalleled multitude of odor molecules facing a smaller but still impressively large number of cognate olfactory receptors. Sometimes a single functional group of the ligand determines the specificity of the ligand-receptor interaction, in other cases an ensemble of molecular features is recognized. We have recently deorphanized an olfactory receptor of the trace amine-associated receptor family, TAAR13c, as a specific and sensitive receptor for a bifunctional compound, the death-associated odor cadaverine.

Here we have modeled the cadaverine/TAAR13c interaction. Several predicted binding residues were exchanged by site-directed mutagenesis, and after heterologous expression the functionality and pharmacological properties of the resulting receptors were compared with wildtype TAAR13c. We observed a binding site for cadaverine at the external surface of the receptor, in addition to an internal binding site, whose mutation resulted in complete loss of activity. Unexpectedly, site-directed mutagenesis of the external binding site resulted in supersensitive receptors. Modeling suggests this site to act as a gate, limiting access of the ligand to the internal binding site and thereby downregulating the affinity of the native receptor. This constitutes a novel mechanism to fine-tune physiological sensitivity to socially relevant odors.

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