7th Munich
Life Science Symposium
for Young Scientists

<interact> 2014

February 27, 2014 | Pre-event | Deutsches Museum
February 28, 2014 | Main Event | TU Munich downtown campus

Impressum

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Life.
Science.
Community.

Welcome to

<interact>2014

Dear Young Scientist,

It is a pleasure to welcome you to the 7th annual Munich Life Science Symposium, <interact> 2014.

Since the beginning of the <interact> in 2007, the major idea of the symposium stayed the same. That is not only to enable, but moreover to encourage the interaction between different life science research communities in Munich. Over the years, <interact> has grown and each year had the opportunity to improve and expand. We believe that it has fulfilled its role so far and we hope that it will do so this year as well.

This year we have more than 300 participants with over 90 scientific contributions. You will have the opportunity to enjoy lectures from distinguished scientists, get acquainted with methods, and hear about different career prospects. We are especially proud that this year the pre-event is taking place at the Deutsches Museum, a suitable venue for the start of the symposium.

We hope that you will learn something new, network with colleagues, and especially that you enjoy yourself and have a good time. Everything is in place for you to explore new ideas and be creative. Who knows, you might be on the path to discovering something big!

Your <interact> Organizing Team.

A little guide of the event

Content

Introductory Note by the President of the TUM, Wolfgang A. Herrmann

Ladies and Gentlemen,

It is my great pleasure to welcome you to the Interact symposium 2014! Our university, the Technische Universität München (TUM), is proud to host the main event of the 7th interdisciplinary PhD symposium organized by Munich-based PhD students for all young researchers in the life sciences.

Among the different scientific fields represented in Germany, the life sciences are a particularly thriving field and at the same time successfully covered in the greater Munich area. I am delighted to see Munich's life sciences doctoral students establishing contact between the city's various research institutions. By organizing an interdisciplinary conference like Interact you are contributing to assure Munich's position as a high-class place for scientific research and at the same time you take over an active part within the network.

Interact gives you the opportunity to exchange scientific tools and knowledge output by presenting your own research work in talks and poster sessions in an early stage of your career as researchers. Not only will you practice your communication skills, but also share and develop innovative ideas possibly merging into new collaborations and alliances in an interdisciplinary approach. The actions

that flow from this transcending of traditional boundaries are essential for progress in science, for our call to deliver cutting edge research, even more so looking at the great challenges modern societies are facing worldwide.

I hope that Interact 2014 will be a great success and wish all participants many fruitful discussions and exciting findings during the symposium and throughout their further career.

With my best personal regards

Wolfgang A. Herrmann

Introductory Note by the Mayor of Munich, Christian Ude

With the LMU and TUM, two of Germany's universities of excellence, as well as numerous academic institutions such as the MPI and the Helmholtz Zentrum, Munich enjoys an excellent international reputation as a unique research environment with top-laboratories in all fields of Life Sciences. Just as important to this reputation is the cluster of various biotech and pharmaceutical enterprises based in and around Munich, forming one of the "top biotechnology clusters" worldwide. The exemplary collaboration and exchange between academic and non-academic partners in this area has led to the scientific and economic success characterizing the still growing Munich Life Science research area.

This year the Munich <interact> symposium, held at the Deutsches Museum and TUM campus in downtown Munich, will again provide young scientists with a valuable platform for fostering new ideas, cooperation and friendships within an interdisciplinary atmosphere. Each year the conference continues to evolve and grow, but the basic aim still remains the same: to bring the young life science community closer together and facilitate networking between academic as well as non-academic partners.

It is a great pleasure for me to act again as the patron of this exceptional event and I wish the symposium a successful 7th meeting.

Christian Ude

Geleitwort

Mit dem *Zentrum Neue Technologien* haben wir im Jahr 2009 eine zukunftsweisende Ausstellung eröffnet, die unseren Besuchern einen breiten Überblick über das Gebiet der Nano- und Biotechnologie vermittelt. Neben den eigentlichen Ausstellungselementen, die durch zahlreiche Exponate die Entwicklungen und Fortschritte dieser beiden Schlüsseltechnologien widerspiegeln, ist es uns auch wichtig, im *Deutschen Museum* den Prozess der naturwissenschaftlichen Forschung aufzuzeigen. Wir tun dies mittels unserem „*Gläsernen Forscherlabor*", in welchem Wissenschaftler live an aktuellen Forschungsthemen aus der Nanotechnologie forschen und mit unserem „*DNA-Besucherlabor*", in dem Schüler einfache biologische Versuche durchführen und so den Prozess des Forschens vereinfacht nachvollziehen können. Das *Zentrum Neue Technologien* ist demnach nicht nur ein Ort der puren Informationsweitergabe, sondern viel mehr: ein Ort des Austauschs und der Diskussion, der Begegnung von Fachleuten und Laien sowie von jungen mit erfahrenen Wissenschaftlern. Schüler haben die Möglichkeit, ihr Wissen zu vertiefen und können außerdem wertvolle Impulse für die Berufswahl erhalten.

Dieser Auftrag des *Deutschen Museums*, die Wissenschaft an die Öffentlichkeit zu bringen und damit Wissenschaftskommunikation zu fördern, wird nun bereichert durch unsere Kooperation: Es ist uns eine besondere Freude, dass ein Teil der Konferenz Interact 2014 in unserem Hause stattfindet. Die Vernetzung der jungen Wissenschaftler und Wissenschaftlerinnen aus verschiedenen Instituten untereinander sowie mit erfahrenen, renommierten Forschern hat im *Zentrum Neue Technologien* nicht nur einen spannenden Ort, sondern auch einen inhaltlich passenden Rahmen gefunden.

Ich wünsche Ihnen eine spannende und erkenntnisreiche Konferenz!

Ihr Wolfgang M. Heckl

Generaldirektor Deutsches Museum und Oskar von Miller Lehrstuhl, TU München

Foreword

With the opening of the *Centre for New Technologies* in 2009, we have created a trendsetting exhibition to provide our visitors with a broad overview of the nano- and the biotechnology research fields. Besides the standard elements, which show the products of developments and advancements of the key technologies, it is also important to us to show in the *Deutsches Museum* the process of life science research. This is why we have established our *Open Research Laboratory*, where scientists carry out on-the-spot research on the latest nanotechnology related topics, and our *DNA Visitors' Laboratory*, where students can perform simple biological experiments and become acquainted with the research process. The *Centre for New Technologies* is not only a place for providing information, but much more: it is a place of exchange and discussion, with interaction between the experts and the non-professionals, as well as between the young and the experienced scientists.The students have the possibility to deepen their knowledge here as well as to receive valuable advice about the careers in science.

The mission of the *Deutsches Museum*, to make science more accessible to the general public and to promote scientific communication, will now be enriched through our cooperation. It is our pleasure that the pre-event of the **Munich Interact 2014** Conference will take place at the *Deutsches Museum*. The *Centre for New Technologies* will provide a suitable and an exciting place for the young scientists from different institutes to interact with each other and with the experienced, world-renowned researchers.

I hope that you have an interesting and a horizon-broadening conference!

Wolfgang M. Heckl

General director of Deutsches Museum and Oskar von Miller Chair, TU München

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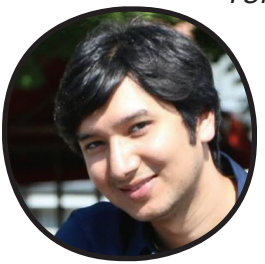

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Pre-event

As young researchers, we often question our present and future as scientists, and struggle to understand the impact of our work on society. During the pre-event, we will try to bridge the gap between "life" and "science" in the life sciences thanks to two successful scientists who also work to relay their experience to the "laymen". ***Prof. Hans Peter Peters*** *and* ***Dr. Adam Ruben*** *will share their inspiring perspectives on the personal and social impact of science.*

Why have you committed to spending the best years of your life without sunlight? How can you convincingly fudge data and feign progress? Which departmental events have the best unguarded free food? These and other questions will be satirically discussed by Dr. Adam Ruben.

Prof. Dr. Hans Peter Peters, will talk about scientists' involvement in public communication, motivating and regulating influences of scientific communities and research organizations. In addition, he will discuss the prospects and pitfalls of interactions with the media for (young) researchers.

Feb. 27th Deutsches Museum

5:00 - 5:45 p.m.	Registration & Admission
6:00 - 6:05 p.m.	Welcome words
6:05 - 6:20 p.m.	Opening remarks by Deutsches Museum
6:20 - 7:20 p.m.	Keynote lecture I **Prof. Hans Peter Peters**
7:25 - 7:40 p.m.	McKinsey talk
7:40 - 8:15 p.m.	Wine & cheese break
8:15 - 8:30 p.m.	Amgen talk
8:35 - 9:35 p.m.	Keynote lecture II **Dr. Adam Ruben**

Hans Peter Peters

pre-event keynote lecture I

A short biographical sketch

Hans Peter Peters is a senior researcher at the Institute of Neuroscience and Medicine, section Ethics in the Neurosciences, of Forschungszentrum Jülich, and Adjunct Professor of Science Journalism at the Free University of Berlin. His research deals with the formation of public opinion on science, technology, biomedicine and the environment under the conditions of a media society. In particular, he focuses on the interdependencies of science and journalism, the medialization of science and the role of mass media in science governance. He is a member of the Scientific Committee of the International Network on Public Communication of Science and Technology (PCST) and serves on the Editorial Board of Public Understanding of Science and the Editorial Advisory Board of Science Communication.

Abstract

Scientists as public communicators in a complex media world

In his address at the 1977 Annual Meeting of the German Research Foundation (DFG), Helmut Schmidt, then German Chancellor, appealed to scientists to increase their public visibility. Obviously, they have listened to him. Surveys show that most researchers nowadays talk to journalists and engage in other public communication activities. Some like it, some accept it as a necessity, but few outright reject it. Scientists' motives are diverse. They may enjoy sharing their knowledge and enthusiasm with the public, they may consider it a duty towards the taxpayer, they may want to increase the 'broader impact' of their research, or they may expect benefits for their personal career, their projects and for social support of science more generally. For a long time, talking to journalists from newspapers, magazines, radio and TV has been the major form of public science communication. But the Internet has created many opportunities for direct and dialogic communication between scientists and the public via websites, blogs and social networks. Furthermore, science festivals, science cafés, science slams and open days have added occasions for scientists to interact with the public face-to-face. Still, the journalistic media continue to be particularly important. Based on empirical data from international surveys of life scientists, my talk will analyze scientists' involvement in public science communication, motivating and regulating influences of scientific communities and research organizations, and repercussions of scientists' increased media orientation on scientific research. It will furthermore discuss the prospects and pitfalls of interactions with the media for (young) researchers.

Adam Ruben

pre-event keynote lecture II

A short biographical sketch

Dr. Adam Ruben obtained his PhD in molecular biology at Johns Hopkins University testing potential new malaria drugs. At the same time, he spent his nights performing as a stand-up comic, which he continues to do alongside his work at a biotech company called Sanaria Inc. This company is dedicated to the production of a malaria vaccine.

Learn more at *adamruben.net.*

Abstract

Which departmental events have the best unguarded free food? How can you convincingly fudge data and feign progress? And why have you committed to spending the best years of your life without sunlight? Adam Ruben (PhD!) answers all of these questions in his book, *Surviving Your Stupid, Stupid Decision to Go to Grad School*.

Like you, Adam Ruben once decided to enroll in grad school. He quickly realized the experience was not what he'd imagined it would be. Adam spent seven years in the Biology Department at Johns Hopkins University, working on malaria drugs that will never benefit humanity, publishing papers that no one will ever read, teaching classes no one remembers, and stealing bagels from seminars he didn't attend.

This talk will review the low points and, well, lower points of post-baccalaureate education. Adam will discuss his own grad school experience, read excerpts from the book, and field questions from the audience. If someone points out how much better PhD programs are in Europe, Adam may cry.

After the talk, Adam will try to sell you a book. Since you're such a nice person, you'll buy several.

Main event

Feb. 28th TU Munich Downtown campus

Program at a glance

08:00 am - 08:45 am	Registration				
09:00 am - 09:10 am	Opening Remarks				
09:10 am - 10:20 am	Keynote lecture I - **Prof. Thilo Stehle**				
10:25 am - 11:25 am	Coffee break, Poster session I Company/Graduate School fair				
	parallel sessions	session I room 2750	session II room 2770	session III room 2760	session IV room 2100
11:30 am - 11:50 am		student talk 1	student talk 4	student talk 7	student talk 10
11:55 am - 12:15 pm		student talk 2	student talk 5	student talk 8	student talk 11
12:20 pm - 12:40 pm		student talk 3	student talk 6	student talk 9	student talk 12
12:45 pm - 02:10 pm	Lunch at the TU & Company fair				
02:15 pm - 03:00 pm	parallel sessions	Method session I	Method session II	Method session III	Method session IV
03:05 pm - 03:25 pm		student talk 13	student talk 15	student talk 17	student talk 19
03:30 pm - 03:50 pm		student talk 14	student talk 16	student talk 18	student talk 20
03:50 pm - 04:55 pm	Coffee break, Poster session II Company /Graduate School fair				
05:00 pm - 06:10 pm	Keynote lecture II - **Prof. Paul Frankland**				
06:15 pm - 06:30 pm	Closing Remarks				
06:30 pm - 08:00 pm	Dinner and Award ceremony at TUM				
	Party with open end at CADU				

Thilo Stehle

keynote lecture I

A short biographical sketch

Thilo Stehle is Professor and Head of the Biochemistry Institute at the University of Tübingen, furthermore Adjunct Professor in Pediatrics at Vanderbilt University School of Medicine.

Stehle started his scientific career as Chemistry student at the University of Freiburg, from which he also obtained his PhD in 1992 for the analysis of the structure and reaction mechanism of enzymes. He stayed in the field of structural biology for his Post-Doc and joined Stephen Harrison at Harvard University, where he elucidated the structure of complete virus particles. In 1997, Stehle established his own group at Harvard Medical School. This was the starting point for his still ongoing research about interactions between viruses and receptors on the molecular and structural level. In early 2005, Stehle moved to Tübingen and took up his current position.

Abstract

Attachment strategies of glycan-binding viruses

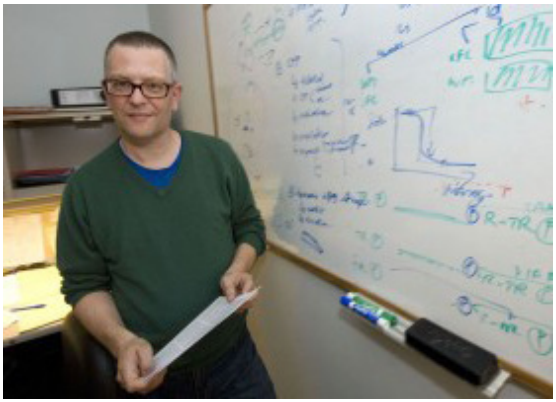
Virus attachment to cells initiates infection and is also a key determinant of host range, tissue tropism and pathogenesis. Carbohydrates such as sialic acid are prominently displayed on many cell surfaces, and they are frequently used by many viruses as their initial, and sometimes only, attachment receptors. Understanding how viruses engage sialic acid is essential for combating infection and designing improved therapeutic viral vectors. Recent advances in studies of virus-glycan interactions have made it possible to rapidly identify specific receptors using glycan array screening, define the atomic level structure of virus-glycan interactions using crystallography, and generate recombinant viruses or pseudoviruses to rationalize the effect of glycan binding in cell entry, tissue tropism, and disease pathogenesis.

I will report on the current state of our ongoing effort to define the receptor binding properties of human polyomaviruses, adenoviruses and coxsackieviruses. All three pathogens use sialylated glycan receptors for their cell attachment. In combination with mutagenesis experiments and functional studies, structural analyses have enabled us to understand the determinants of specificity in each case. Exploitation of these determinants provides an excellent platform for the development of antiviral agents. We are also able to show that receptor specificities can be switched through subtle changes in the binding pockets, demonstrating the dynamic aspects of virus interactions with receptors.

Paul Frankland

keynote lecture II

A short biographical sketch



Paul Frankland started out studying Psychology at the University of Sheffield in the north of England. In order to investigate what drives human behavior, he soon switched to the more experimentally driven neurosciences where in his final year as an undergraduate student he already published his first paper with Peter Redgrave about neuronal recordings in rats. He received his PhD in neuroscience from the University of Toronto where he worked in John Yeomans lab. There he focused on electrophysiological and behavioral methods in order to map the brain circuits for the startle reflex. Prof. Frankland gained postdoctoral research experience in the lab of Alcino Silva, who had pioneered the use of gene-targeting, at Cold Spring Harbor. During his postdoc he focused on the molecular basis of behavior.

Today Paul Frankland is an Associate Professor at the University of Toronto and his research focuses on how the human brain encodes, stores and maintains memories.

Abstract

Adult neurogenesis, forgetting and infantile amnesia

New neurons are continuously added to the subgranular zone of the hippocampus throughout the lifespan, but the functional consequences of hippocampal neurogenesis remain unclear. While the majority of previous studies have examined the impact of increasing or decreasing hippocampal neurogenesis on subsequent memory formation, few have examined the effects of similar manipulations on established, hippocampus-dependent memories. Computational models predict that addition of new neurons should lead to extensive remodeling of hippocampal circuits, and consequently degradation or forgetting of established memories. Consistent with this, lifespan changes in hippocampal neurogenesis are inversely correlated with memory persistence: During infancy, when hippocampal neurogenesis levels are high, freshly-generated memories tend to be rapidly forgotten. In contrast, during adulthood, when neurogenesis levels are lower, memories are typically much more persistent. We have conducted two types of experiments that suggest that neurogenesis and forgetting are causally related. First, in adult mice (P60), we find that increasing neurogenesis after memory formation is sufficient to induce forgetting. Second, in infant mice (P17), we find that decreasing neurogenesis after memory formation mitigates normal forgetting observed at this age. Our data suggest a causal relationship between neurogenesis and memory persistence, and provide a neurobiological account for infantile amnesia.

method session I

Statistics and Experimental Design in Contemporary Life Science Research

Dr. Tobias Straub

Biomedical research increasingly involves numerical interpretation of quantitative data. Decent knowledge of statistics is required to face the challenges of analysing own experiments and evaluating published results. Furthermore, experimental design can be improved based on statistical considerations allowing in return sustainable science. I will introduce a few key aspects of statistical analysis and experimental design which should serve as a guide for increasing competence in a crucial discipline of life science research.

method session II

Alternative Career Paths for Life Scientists

Thorsten Abs

Once you have your qualifications in place, it can be difficult to decide on what you want to do with them: should you pursue a classic university career, or would it be better to look beyond the lab? What other kinds of job are on offer in the academic world? What are the options for a scientist in industry? What does it take to make the leap into a commercial environment?

Many scientists are faced with this kind of dilemma. In this session the managing director of academics, Thorsten Abs will help you make an informed decision. He will talk about career opportunities outside the lab -- different professional roles, the qualifications you need for them, and the salary prospects they hold.

method session III

Visualization and Targeted Disruption of Protein Interactions

Prof. Heinrich Leonhardt

Protein–protein interactions are the basis of all processes in living cells, but most studies of these interactions rely on biochemical in vitro assays. I will review the most popular methods to study protein interactions and briefly discuss their technical challenges, advantages and shortcomings. I will then present a simple and versatile fluorescent-three-hybrid (F3H) strategy to visualize and target protein–protein interactions (*Herce et al., 2013, Nat Commun 4, 2660*). We used a high-affinity nanobody to anchor a GFP-fusion protein of interest at a defined cellular structure and measured the enrichment of red-labelled interacting proteins at these sites. With this approach, we visualized the p53–HDM2 interaction in living cells and directly monitored the disruption of this interaction by Nutlin 3, a drug developed to boost p53 activity in cancer therapy. We further used this approach to develop a cell-permeable vector that releases a highly specific peptide disrupting the p53 and HDM2 interaction. The availability of multiple anchor sites and the simple optical readout of this nanobody-based capture assay enable systematic and versatile analyses of protein–protein interactions in practically any cell type and species.

method session IV

Advanced Fluorescence Microscopy Techniques

Prof. Don Lamb

Development of the microscope is probably the single most important physical innovation for the life sciences. In the last couple of decades, new optical microscopy have been developed including the development of fluorescence fluctuation spectroscopies and super-resolution methods that have a broad range of applications in the physical, chemical and biological sciences.

In this talk, I will give a basic overview of some of these advanced methods and highlight their application with examples. I will give a brief introduction to fluorescence fluctuation spectroscopy (FFS) and how we can extract quantitative information out of fluctuations. FFS can be used to investigate molecular mobility and interactions.
In the second part of this talk, I will introduce the different types of super-resolution spectroscopy, structure illumination spectroscopy, stochastic optical reconstruction microscopy (STORM) (or photoactivation localization spectroscopy, PALM), and stimulated emission depletion (STED) microscopy. The advantages, limitations and accuracies of the different methods will be discussed. We will also discuss super-resolution optical fluctuation imaging (SOFI), which utilizes fluorescence fluctuation spectroscopy to create super-resolution images.

In the last couple of minutes, time permitting, I will introduce a novel orbital tracking microscope that we have developed that

can track single particles in real time with millisecond resolution and nanometer accuracy over centimeters and hours. We have used this microscope to track mitochondria transport in Zebra fish embryos.

Student Speakers

1
Session I
11:30 am

method development - room 2750

Highly efficient targeted mutagenesis in mice by using sequence specific nucleases

Sudeepta Kumar Panda[1], Benedikt Wefers, Oskar Ortiz, Wolfgang Wurst, Ralf Kühn

[1]Institute of Developmental Genetics, Helmholtz Zentrum München, German Research Center for Environmental Health, 85764 Munich, Germany

Genome engineered mice are instrumental for the analysis of gene function in health and disease. Conventional embryonic stem cell-mediated gene targeting is a time consuming and labor intensive process involving the 3 steps of targeting vector construction, chimera production, and germline transmission. Here, we developed an advanced approach for routine production of mouse disease models by microinjection of improved versions of transcription activator-like effector nucleases (TALEN) and clustered regularly interspaced short palindromic repeats (CRISPR)/Cas9 by tethering a polyadenylation tail (95 or 166A) and single stranded oligodeoxynucleotides into one-cell embryos. To knockout the C9ORF72 gene as a model for frontotemporal lobar degeneration, TALEN-95A mutagenesis induced sequence deletion in 41% of pups derived from microinjected embryos. Using TALENs together with mutagenic oligodeoxynucleotides, we introduced amyotrophic lateral sclerosis patient-derived missense mutations in the fused in sarcoma (Fus) gene at a rate of 6.8%. In response to oxidative stress, embryonic fibroblasts derived from these mutants assembled into perinuclear stress granules. Similarly to knockout of the Fus gene, single guide RNA (sgRNA)/Cas9-95A mutagenesis induced sequence deletion in 12% of pups derived from microinjected embryos. Using sgRNA/Cas9-95A together with mutagenic oligodeoxynucleotides, we introduced targeted codon replacement mutations in the Rab38 gene at a rate of 14%. By using two sgRNA/Cas9-166A targeted to introns 6 and 8 of Fus gene, we achieved one homozygous founder harboring a genomic deletion of 2.6 kb. Taken together, TALEN and CRISPR/Cas9 systems represent efficient and versatile genome editing tools that enable accelerated routine production of new disease models for studying genetic disease mechanisms.

method development - room 2750

2
Session I
11:55 am

Single-molecule force spectroscopy: Interrogating proteins on the rack

Fabian Ziegler

TU München, Physik-Department E22 - Lehrstuhl für Biophysik (Prof. M. Rief)

Single-molecule force spectroscopy has established itself as a versatile tool to gain insights into mechanisms of protein folding, enzyme kinetics and ligand binding at a single molecule level.Due to the difficulties that come along with a technique that gains its information by pulling at one single molecule and deals with sub-piconewton forces and sub-nanometer distances in solution and due to the low number of molecules one can examine in time (remember: you're watching only one single molecule) this method still kept its position as an exotic sub-genre in life science, although the obtained data provide an accuracy that cannot be reached with standard ensemble measurements.

In this talk, I would like to give a short overview about the possibilities, the challenges and the limitations of this method and present some recent developments that help us to bridge the gap between the abstract concept of energy landscapes and the real-time observation of protein folding on your computer screen.

3
Session I
12:20 pm

method development - room 2750

Developing optical tools for controlling transmembrane receptors with light

Matthias Schönberger[1], Dirk Trauner

[1]Ludwig-Maximilian-University Munich, Trauner Research Group, Chemical Synthesis & Neuroscience

Pain is a very unpopular feeling. Chemical research has brought forth small molecule drugs that inhibit or reduce pain, such as local anesthetics or opiates. Biochemistry has helped identifying the molecular targets of these drugs. Based on known anesthetics that act on voltage gated ion channels and opioid receptors, we have now developed novel photo-switchable analogs. These photo-switchable drugs can be used to control the function of their protein targets with light.

4
Session II
11:30 am

Pioneer neurons of the locust antenna: The development of a sensory system

E.E. Ehrhardt[1], George Boyan

[1]Graduate School of Systemic Neuroscience, Biocenter, Ludwig-Maximilians-Universität München

Pioneer neurons establish the first axonal pathways of the developing nervous system. Pioneers using similar navigational mechanisms have been described in many animal species, including both vertebrates and invertebrates.

We are documenting the life history of the pioneers of the locust antenna, from their birth at approximately 1/3 of the way through embryogenesis, until their death at less than 2/3 of embryogenesis. The organization of the pioneers corresponds to the segmentation of the antenna; during early embryogenesis, each segment produces its own pioneers whose axons travel towards the brain.

We have set up a locust embryo culture system that will allow us to perform a variety of manipulative experiments on the developing antenna, including laser ablation and antibody block experiments. Molecular markers such as neuron-specific horseradish peroxidase and GPI-linked cell surface lipocalin Lazarillo allow us to visualize the neurons of the antenna, and thus to discover how the innervation of this sensory organ is constructed by pioneers, motor neurons and sensory neurons.

5
Session II
11:55 am

neuroscience - room 2770

Microtubule dynamics in developing and diseased axons

Tatjana Kleele[1], Petar Marinkovic, Monika Brill, Ronald Naumann, Emily Weigand, Derron Bishop, Martin Kerschensteiner, Leanne Godinho, Thomas Misgeld

[1]Institute of Neuroscience, Technische Universität München

Microtubules are major cytoskeletal components of all eukaryotic cells. In neurons, microtubules play key roles in polarization, organelle transport and neurite remodeling. Disturbances of microtubule organization can be detected early in neurodegenerative diseases, underscoring their importance in maintaining cellular structure and function and making them interesting structures to study in the context of health and disease. Microtubule organization is regulated by different microtubule associated proteins, including plus-end-tracking proteins (+TIPs), which accumulate at the growing plus-end of microtubules and indicate their dynamic remodeling. Microtubule behavior can be studied by fluorescently tagging +TIPs, a technique that has been applied in vitro and in invertebrates. To assay such remodeling by in vivo imaging in the mammalian nervous system, we generated transgenic mice that express the +TIP, EB3, fused to yellow fluorescent protein (YFP) controlled by the neuron-specific Thy1 promoter. Thy1:EB3-YFP mice allow essaying microtubular dynamics and polarization in different compartments of the nervous system as well as the status of the microtubular cytoskeleton in acute and chronic models of axonal injury and disease. We found that an increase in microtubule dynamics is an indicator of imminent axon degeneration. Injury-induced acute axonal degeneration can be reduced by titrating microtubule-stabilizing drugs to block microtubule destabilization. In addition to being an early indicator of axon degeneration, we found an increase in EB3-comet density during axon regeneration and developmental reorganization. This suggests that increased microtubule dynamics might be a general “plasticity tag” for axons that can be read out by our novel approach.

6
Session II
12:20 pm

Epigenetic mechanisms linking major depression and aging

Anthony Zannas[1], Torsten Klengel, Elisabeth Binder

[1]Research Fellow, Max Planck Institute of Psychiatry

Stressors and psychiatric disease have been linked with accelerated cellular aging, but the mechanisms underlying this relationship remain elusive. Our objective was to examine the role in this of DNA methylation changes in the gene encoding FK506 binding protein 51 (FKBP5), a co-chaperone and modulator of the glucocorticoid receptor complex. DNA methylation levels of 16 CPGs located in glucocorticoid response elements in the FKBP5 promoter, intron 2, and intron 7 were measured in peripheral blood via bisulfite pyrosequencing. Major depression was assessed using the Structured Clinical Interview for DSM-IV and the Beck Depression Inventory. Across all tested CpGs in FKBP5, we noted a demethylation with increasing age driven by effects on CpGs in intron 7. In the discovery sample and after controlling for sex and lifetime trauma exposure, we found that current depressive symptoms moderated age-related demethylation of CpGs that we had previously shown to be sensitive to trauma and glucocorticoid exposure and to be demethylated in aging human brain. Depressed subjects showed accelerated demethylation as compared with non-depressed subjects (reproduced in a replication sample). Similar moderation of age-related demethylation was observed with lifetime depression diagnosis. Demethylation of glucocorticoid-sensitive CpGs was further associated with increased levels of mRNA expression of pro-inflammatory genes in the peripheral blood. Overall, depressive phenotypes appear to accelerate age-related epigenetic modifications in FKBP5 glucocorticoid-sensitive CpGs. These effects may be linked to the increased risk for age-related disorders observed in depressed patients.

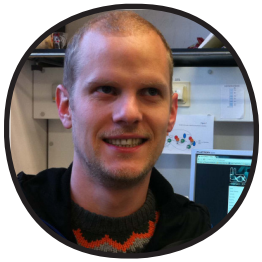
7
Session III
11:30 am

epigenetics - room 2760

A bivalent interaction between the chaperone NASP and histones suggests a molecular switch in early H3-H4 maturation

Andrew Bowman[1], Hari Singh, Gyula Timinszky, Andreas Ladurner

[1]Department of Physiological Chemistry, Adolf Butenandt Institute, LMU Munich



Factors that associate with soluble, non-chromatin bound histones play important roles in maintaining genomic stability, ensuring efficient replication and regulating gene expression. One such protein, sNASP, is conserved from yeast to man, and likely plays a key role in histone metabolism. sNASP contains three domains, a TPR repeat region, a predicted acidic loop and an unstructured N-terminal domain. Through in vitro and in vivo approaches, two distinct modes of interaction between sNASP and H3-H4 were uncovered. The TPR repeat domain of sNASP interacts with a peptide motif within H3. This overlaps with the binding site of the co-chaperone ASF1. sNASP and ASF1 binding are not mutually exclusive, with both being able to interact with the same H3-H4 molecule at the same time. This apparent discrepancy was resolved by the discovery of a second histone binding site within the acidic loop region of sNASP.

Our observations suggest a series of consecutive steps, whereby sNASP aids in generating deposition competent histones. sNASP sequesters the C-terminus of H3 after synthesis, before folding with H4. As folding of H3-H4 progresses, the acidic loop of sNASP contributes to a secondary interaction with the histone fold dimer. The C-terminus of H3 is relinquished to ASF1, as folding of H3-H4 changes the context in which sNASP interacts. sNASP remains associated with the complex, contributing to its solubility and preventing side interactions, until histones are passed on to the deposition machinery.

8
Session III
11:55 am

Function and regulation of the growth-promoting JAK/STAT signaling pathway in imaginal disc regeneration

Marco La Fortezza[1], Madlin Schenk, Alexander Pinduyrin, Bas van Steensel, Anne-Kathrin Classen

[1]LMU München Biozentrum Martinsried

The cellular signaling pathways that initiate and coordinate regenerative growth or tissue patterning after wounding are just beginning to be understood. We employ genetically induced cell ablation in Drosophila imaginal discs to investigate cellular responses to tissue damage and tissue stress. Using GFP-signaling reporter and real-time qPCR, we find that the JAK/STAT pathway and it's ligands, encoded by the unpaired gene family, are strongly upregulated during imaginal disc regeneration. Curiously, the extent of upregulation strongly correlates with regenerative capacity during development. We find that activation of this growth-promoting pathway is required for successful regeneration of disc-autonomously. In addition, we observe a disc non-autonomous requirement of JAK/STAT signaling in successfully inducing a developmental delay after tissue damage. We are currently analyzing the specific requirements for JAK/STAT in promoting compensatory proliferation and characterizing its contribution in mediating physiological delay response after cell ablation. Previous work suggests that unpaired genes are regulated by the Polycomb family of epigenetic silencers. In addition to the regulation of growth, Polycomb is best known for maintaining cellular identities by targeting transcription factors involved in differentiation. Our findings therefore provide us with a paradigm to study mechanisms of epigenetic plasticity during regeneration at dynamically expressed genes like unpaired. To visualize dynamic changes in epigenetic signatures, we are building DamID-based tools to map DNA-binding sites of chromatin-modifying complexes in very small cell populations, in vivo. We recently obtained a first genome-wide profile of Polycomb-binding in wild type wing imaginal discs, confirming that DamID is suitable for in vivo approaches.

9
Session III
12:20 pm

epigenetics - room 2760

The histone H3-K27 demethylase Utx regulates HOX gene expression in Drosophila in a temporally restricted manner

Omer Copur[1], Jürg Müller

[1]Chromatin-Biologie, Max-Planck-Institut für Biochemie

Formation of multi-cellular organisms is a dynamic process and genes have to be turned ON/OFF in a spatio-temporal manner to ensure proper development. Histone modifications regulate the ON/OFF state of genes. However, it is unclear how dynamic histone modifications are. Methylation of Histone H3 (H3K27) is correlated with gene repression and it was thought to be an irreversible reaction. However, discovery of demethylases that remove methyl mark from H3K27 has challenged this view. Unlike worm and mammals, Drosophila has a single ortholog of H3K27 demethylases, dUTX, making it an ideal system to study the function of this enzyme in vivo.

To this end, we have generated a knock-out allele of dUTX. Loss of maternal dUTX leads to early larval lethality and defects in body patterning. Interestingly, the expression pattern of homeotic genes is misregulated in subsets of cells in the maternal mutants. To analyze the function of dUTX in global gene regulation we performed immunoblots on larval extracts from mutant progenies. In contrast to previous studies, we haven't found any global change in levels of H3K27me3. Surprisingly, we found that global levels of H3K4me mark are reduced in mutants. These results indicate that dUTX acts as a maintenance element rather than a gene activator and it does not control global gene expression, but may control only a core network of developmental regulators. Current focus of the project is to understand, (1) whether dUTX antagonizes transcriptional repressors, (2) whether dUTX has catalytic-independent functions. I am currently performing genetic interaction experiments and using catalytically inactive mutants to address these issues.

10
Session IV
11:30 am

Human interactome of a thousand proteins in three quantitative dimensions

Marco Y Hein[1], Nina C Hubner, Ina Poser, Jürgen Cox, Nagarjuna Nagaraj, Anthony A Hyman, Matthias Mann

[1]Department of Proteomics and Signal Transduction, Max Planck Institute of Biochemistry

The protein interactome constitutes next level of complexity on top of the genome, transcriptome and proteome. Here we report the first large-scale study that characterizes the human interactome in multiple quantitative dimensions against the backdrop of the underlying proteome.

We generated a library of cell lines expressing 1,034 distinct GFP-tagged proteins under endogenous control. Using quantitative proteomics, we identified their interactors and estimated their binding stoichiometries. Our interactome data connect more than half of the expressed host proteome and span its entire dynamic range of protein abundances. We discovered a wealth of known and novel protein interactions and take a unique look at the interplay of proteome and interactome. The stoichiometry readout discriminates weak from strong interactions and classifies interactors into complexes, which explains phenotypes and genetic interactions. Furthermore, we show directly that a majority of weak interactions render the cellular interaction network as a 'hairball'. Our study adds several missing layers to the information previously offered by quantitative interactomics, providing a rich resource for exploration of the human proteome and interactome and demonstrating the interdependence of both systems.

11
Session IV
11:55 am

bioinformatics - room 2100

Identification of host interactions for phenotypic antimalarial hits

Andreas Spitzmüller[1], Jordi Mestres

[1]Post-Doc at Technische Universität München

Malaria is one of the most epidemic infectious diseases in the world affecting millions of patients and causing more than 500,000 deaths each year. Although there are several established antimalarial drugs in clinical use, there is an urgent need for new drugs due to rapid resistance development. In recent years, three independent screening campaigns disclosed more than 20,000 hits that are phenotypically active against *P. falciparum*, one of the major malaria causing agents. In order to make these hit libraries accessible to as many biological laboratories as possible, the Medicine for Malaria Venture (MMV) compiled and distributed the Open Access Malaria Box, a set of 400 chemically diverse active compounds. One important task is now to elucidate the mode of action of those compounds. However, besides targeting the parasite it is also necessary to identify potential host interactions in order to anticipate the risk of undesired side effects of those chemotypes at the earliest possible stage of development.

To this end, we applied a ligand-based virtual target profiling approach to predict possible interactions with human targets. Amongst others, GPCRs were identified as the most important target class. Subsequently, several hundred predicted interactions were selected for experimental testing. Results showed that a substantial part of the Malaria Box exhibits the potential of interacting with human GPCRs. To this extent, this was unexpected beforehand since the pathogenic agent does not contain any GPCRs. Particular attention was given to 5-Hydroxytryptamine receptor 2B (5-HT2B receptor) agonism, an effect associated with cardiac valvulopathy.

12
Session IV
12:20 pm

Inferring novel relationships between drugs and genes based on phenotypic features

Jeanette Prinz[1], Ingo Vogt, Monica Campillos

[1]IBIS Institute of Bioinformatics and Systems Biology, Helmholtz Zentrum Munich

The molecular mechanisms that translate chemical perturbations into phenotypic effects are largely unknown and consequently there is an urgent need for novel approaches to uncover these molecular relationships. Here, we exploited drug and gene phenotypic information to search for novel molecular associations of drugs. For that purpose, we annotated the side effects of 1346 drugs marketed around the world and 5384 genes from the MGI repository with the MedDRA ontology and measured the phenotypic linkage between drugs and genes by an extended semantic similarity approach. The analysis of the high scoring drug-gene associations shows that the genes in these pairs bear properties of drug targets i.e. they have a central function in protein-protein interaction networks, they tend to be specifically expressed across tissues and their expression profiles correlate with those of the associated known drug targets. Furthermore, we benchmarked the predicted relationships with drug targets from the STITCH database and observed a strong enrichment of physical as well as indirect gene-drug associations. Interestingly, we also predicted unknown associations of drugs and genes, suggesting that our method might reveal novel drug modes of action. We were able to prove this by experimental validation of a predicted interaction.

In summary, we demonstrate that our approach is able to detect direct as well as indirect drug-target associations giving new insights into the molecular mechanisms that translate chemical perturbations into phenotypic effects. Thus, it may help to find new therapeutic applications for drugs and may improve the rational use of medicines.

13
Session V
03:05 pm

cancer - room 2750

Mutant PIK3CA regulates ERK phosphorylation via MKP-1 and determines the response to AKT therapy

Anuja Sathe[1], Ferdinand Gürth, M Kronaeur, M Heck, M Thalgott, JE Gschwend, M Retz, Roman Nawroth

[1]Urologische Klinik und Poliklinik (TUM), Klinikum rechts der Isar, Ismaninger Str. 22, 81675 München

Urothelial carcinoma (UC) is the 6th most common cancer and patients with muscle invasive disease have a poor prognosis. No significant changes in treatment or prognosis have occured over the last 25 years. In our study we examined the PI3K/AKT signaling pathway, which is frequently altered in UC, as a target for novel therapeutic strategies.

We provide evidence that the therapeutic response to AKT inhibition requires mutations in a specific domain of the PIK3CA gene. The presence of these mutations leads to a decrease in ERK phosphorylation via the regulation of MKP-1, upon AKT inhibition. These findings provide a molecular rationale for the selective efficacy seen with AKT therapy and have important implications in targeting the PI3K/AKT signaling network in cancer. Moreover, our results can potentially be translated into the clinic as a personalized medicine strategy, by using the presence of PIK3CA mutations as a stratifying biomarker for AKT therapy.

Combinatorial treatment of lung cancer cell lines and their spheroids with multi-targeted small-molecule kinase inhibitors and salinomycin

Zhiguang Xiao[1], Bianca Sperl, Pjotr Knyazev, Axel Ullrich

[1]Department of Molecular Biology, Max-Planck-Institute of Biochemistry

Lung cancer is the leading cause of cancer-related death and accounts for the most common malignancy in the world. The development and widespread clinical application of inhibitors that target the epidermal growth factor receptor (EGFR) provide important insights for non-small-cell lung cancer (NSCLC) treatment, however, primary or acquired resistance limits the therapeutic success of these targeted therapies. The cancer stem cell (CSC) model states that a distinct subpopulation of tumor cells with stem cell-like properties is responsible for tumor heterogeneity and hierarchy and associated drug resistance, tumor metastasis and relapse. Targeting both differentiated and CSCs by standard chemotherapy and putative stem cell killer (salinomycin), respectively, could be an effective approach for lung cancer treatment. After chemical genomics based cell line selection and multi-rounds combinations screening, we dig out, that metformin, an antidiabetic drug with anticancer efficiency, modestly inhibited the growth of NSCLC cell lines, monolayer cells and spheroids (CSCs) in a dose-dependent manner, interacted synergistically with salinomycin. In addition, combination with metformin and salinomycin markedly inhibited EGFR signaling pathway via blocking Erk1/2 and Akt activation. Moreover, human phosphokinase array detected that combination exhibited upregulation of AMP-activated protein kinase and associated ribosomal protein S6 kinase inhibition as well as the change of expression of pro- and anti-apoptotic genes. Remarkably, significant reduction of tumor sphere formation was seen in all the treated NSCLC cell lines, regardless of their EGFR status. In conclusion, metformin and salinomycin could be a promising treatment option for NSCLC.

15
Session VI
03:05 pm

immunology- room 2770

How does the body prevent auto-immune diseases?

Julia Winnewisser[1], Maria Hinterberger, Lei Wang, Ludger Klein

[1]Medical Faculty – Institute for Immunology, Ludwig Maximilians University Munich

The development of T cells takes place in the thymus, where the generation of T cell receptors (TCR) is random and therefore also yields T cells with a reactivity to structures of the own body. But then, why do we not all suffer from autoimmune diseases? The thymus features the unique ability to express all different kinds of self-antigens of the organism to test the T cell pool for auto-reactivity. Upon antigen encounter, self-reactive T cells can either be eliminated or re-programmed to become a regulatory T cell (T_{reg}). The parameters that determine whether an autoreactive T cell is deleted or deviated into the T_{reg} lineage are not yet fully understood.

To address these questions of central tolerance induction, we generated a new TCR-transgenic mouse model having mainly T cells that display a specificity to the auto-antigen proteolipid protein (PLP) of the central nervous system. PLP is of particular interest as it is thought to be a candidate auto-antigen in Multiple Sclerosis. In the presence of the antigen PLP, PLP-specific T cells are largely deleted and a small fraction is deviated into the T_{reg} lineage. However, the deletion is not complete: there are also autoreactive T cells escaping the selection process and arriving in the periphery. Importantly, the mice do not display any signs of autoimmune disease. This possibly indicates a crucial role of T_{reg} cells in the periphery to keep the escaped autoreactive T cells in check.

TNF-β an up-coming therapeutic for Rheumatoid Arthritis

Constanze Buhrmann[1], Parviz Shayan, Bharat B Aggarwal, Mehdi Shakibaei

[1]Institute of Anatomy, Ludwig-Maximilian-University Munich, Musculoskeletal Research Group, Pettenkoferstrasse 11, D-80336 Munich, Germany

Anti-cytokine based therapies primarily with TNF- α are the main treatment option for Rheumatoid Arthritis (RA). However, patient treatment response is ambiguous and up to 50% become resistant. While TNF-α in RA is well studied, the role of TNF-β, also called Lymphotoxin-α (LT-α), is unclear. Therefore, here we investigated whether TNF-β and its receptor play a role in chondrocytes in inflammatory environment. TNF-β-mediated inflammatory signaling was studied in an *in vitro* model of primary human chondrocytes. In primary human chondrocytes TNF-β and TNF-β-receptor expression induced by cytokine mediated inflammation was accompanied by up-regulation of inflammatory (Cox-2), matrix degrading (MMP-9 and -13) and apoptotic (p53, cleaved caspase-3) signaling pathways. Cytokine-induced NF-kB activation and its translocation to the nucleus was markedly suppressed by anti-TNF-β, similar to the natural NF-kB inhibitor (curcumin, diferuloylmethane) or the knockdown of NF-kB by using antisense oligonucleotides (ASO), highlighting the crucial role of NF-kB in TNF-β-induced-inflammation in cartilage similar to that expected for TNF-α. Finally, TNF-β-induced inflammatory microenvironment significantly enhanced the adhesiveness between TNF-β-expressing T-lymphocytes and the responding chondrocytes in vitro demonstrating the overlapping, interactive role of TNF- β on the whole joint environment.

In this study we demonstrate for the first time that TNF-β is involved in microenvironment inflammation in chondrocytes during RA parallel to TNF-α through up-regulation of NF-kB signaling and activation of pro-inflammatory activity. These findings will provide vital pre-clinical evidence supporting future use of TNF-β in treatment of RA.

17
Session VII
03:05 pm

drug resistance- room 2760

Extension of specificity in the new β-lactamases: A combined theoretical and experimental study

Ewa Chudyk[1], Marc van der Kamp, Jim Spencer, Adrian Mulholland

[1]Centre for Computational Chemistry, School of Chemistry, University of Bristol, UK, current address: Fachgebiet Protein Modelling, Technische Universität München

Antibiotic resistance caused by Gram-negative bacteria has become a real treatment problem in medicine. It is mostly due to the activity of β-lactamases, enzymes responsible for cleavage of β-lactam rings in penicillins, cephalosporins and carbapenems, inactivating these drugs. In clinical use there are only a few inhibitors of β-lactamases, however, antibiotic resistance is still developing due to their clinical overuse. Therefore further investigation of the reaction mechanisms of β-lactamases followed by rational inhibitor design becomes a very challenging medical need.

As proven experimentally, carbapenems selectively escape most of β-lactamases due to slow deacylation of the acylenzyme intermediate. However, SFC-1 enzyme from *Serratia fonticola* hydrolyzes antibiotics, and so there is no effective treatment against those bacteria.

The acylenzyme intermediate from SFC-1 was studied using MD simulations and compared with other β-lactamases unable to deacylate, such as TEM-1, SHV-1 and BlaC. Some conformational differences within the active sites of these enzymes were found. Further comparison of the first step of deacylation reaction mechanism using QM/MM umbrella sampling simulations correctly revealed a significant difference in activation energy barriers for different β-lactamases. Analysis of the simulations indicates factors that may distinguish β-lactamases that can effectively hydrolyse carbapenems from those that cannot.

18
Session VII
03:30 pm

Studying of MRSA growth behavior and secretion of virulence factors in a three dimensional collagen gel

Stefanie Boellner[1], Tobias Veit, Jürgen Heesemann

[1]Max von Pettenkofer Institut, LMU Munich

S. aureus is one of the leading pathogens causing a variety of bacterial infections by expressing a large set of virulence-associated factors. Increasing resistance to antibiotics is an additional challenge during *S. aureus* infections. Since Methicillin Resistant *S. aureus* (MRSA) has been identified for the first time in 1962, its frequency has increased globally.

By using a three-dimensional collagen gel (3D-CoG) supplemented with fibrinogen (F), we obtained a suitable matrix with tissue-like characteristics to study bacterial growth. We studied the growth behavior of a set of eleven MRSA type strains isolated from patients in Europe. The production of extracellular matrix binding protein (Emp) and fibrinogen binding protein (Fib) of these strains was examined by confocal laser scanning microscopy (CLSM). We found that the eleven MRSA strains differ highly in colony size and colony structure when grown in 3D-CoG+F. Furthermore, we could show that most of the strains form a pseudocapsule, while only two strains exhibit a microcolony associated meshwork (MAM, consisting of fibrin) resembling that of the reference strain "*S. aureus* Newman". The production of the virulence factors Emp and Fib differed quantitatively within the strain collection and could be distinguish into three subgroups. These data show the high phenotypic diversity of MRSA patient isolates in regard of microcolony morphology and production of virulence factors such as Emp or Fib under tissue-like growth conditions. Whether these diverse phenotypes of MRSA observed in 3D-CoG+F correlate with the type of infection remains to be elucidated.

19
Session VIII
03:05 pm

advances in live imaging - room 2100

Directed evolution and screening of genetically encoded fluorescent biosensors

Julia Litzlbauer[1], Oliver Griesbeck

[1]Max Planck Institute of Neurobiology, Am Klopferspitz 18, 82152 Martinsried

The discovery of GFP more than 50 years ago revolutionized science, allowing us to make phenomena such as gene expression, protein localization and dynamics visible in living cells. By now, there are countless fluorescent biosensors available, which translate the binding of an analyte to a biological sensor region into a visible change in the fluorescent readout of the sensor. As we seek to explore events that are yet more complex, more subtle or happen faster, new biosensors would require more specialized indicators, which ideally should still remain flexible for broader application. Furthermore, the fact that fluorescent indicators are artificial proteins, consisting of a sensor domain fused to one or more fluorescent proteins, makes it difficult to predict their properties. Thus, it was deemed advantageous to utilize directed evolution and screening as tools to develop new biosensors, as they allow the experimenter to create and test many different sensors in a short amount of time.

Accordingly, our lab has developed a method to screen genetically encoded Ca^{2+} sensors on the basis of FRET (fluorescence resonance energy transfer) in bacteria, with measurements in vitro and in mammalian cells as secondary screening steps. This method assisted us in developing a series of improved Ca^{2+} FRET sensors, the Twitch series, which was successfully used to visualize complex dynamics including very fast action potential firing in neurons and high resolution functional tracking of T lymphocytes. The method was further adapted to develop a dynamic FRET reporter of gene expression.

20
Session VIII
03:30 pm

Zebrafish imaging by means of volumetric optoacoustic microscopy

Moritz Kneipp[1], Hector Estrada, Antonella Lauri, Gil Westmeyer, Daniel Razansky

[1]Institute of Biological and Medical Imaging, Helmholtz Zentrum, Munich

Optical methods have long been key tools in performing biological and medical basic and clinical research. Their ability to accurately visualize intrinsic contrast and exogenous contrast agents has impacted all fields of life sciences. However, the established optical imaging methods are all handicapped by the strong scattering effect of biological tissues on light and the resulting degradation of spatial resolution with increasing tissue depth.

Optoacoustic imaging has the advantage of combining optical contrast with the undisturbed propagation properties of ultrasound in soft tissues. By exciting the samples with nanosecond laser pulses, acoustic pressure waves are created through the optoacoustic effect. These pressure waves are recorded using an ultrasound transducer. Ultrasound suffers very low scattering in soft tissues enabling high resolution imaging at depths far exceeding the established optical methods.

Herein we developed a fast scanning optoacoustic microscope system. By scanning a single focused transducer in two dimensions and recording time resolved optoacoustic signals, whole volumes are recorded on the fly, yielding a high resolution three-dimensional representation of the imaged sample. The system was subsequently used to image migratory melanocytes in zebrafish embryos, as well as the brains of highly scattering adult fish. We have shown that optoacoustic microscopy is capable of producing three-dimensional maps of single melanocytes in adult zebrafish non-invasively.

Posters

Poster Index session I: 10:25 am -11:25 am

Neuroscience

1. Pathway and biomarker discovery in a posttraumatic stress disorder mouse model
-Chi-Ya Kao

3. Angiogenesis promoting proteins in pituitary adenomas
-Ninelia Minaskan Karabid

5. Epigenomic Alterations in Glioblastoma
-Theo F. J. Kraus

7. The bone morphogenic protein 7 (Bmp7) plays a pro-tumorigenic role in pheochromocytoma
-Ines Leinhäuser

9. Early-life stress interacts with genetic predisposition in a mouse model of affective disorders
-Silja McIlwrick

11. How to make a photoreceptor neuroscience needs?
-Johannes Morstein

13. Measuring the human startle reflex with simultaneous EMG and fMRI
-Peterse YH

New methods/ Translational Biochemistry

15. MemConP: Accurate prediction of membrane protein contacts and helix interactions
-Peter Hönigschmid

17. Seeing is believing – How to visualize a new type of membrane scaffold
-Anna Kaufmann

19. Annotation of directed genomic states unveils variations in the Pol II transcription cycle
-Benedikt Zacher

21. Interaction of buckminsterfullerene (C60) with T. thermophila: Phenotypic changes due to nanomaterial
-Rajesh Rathore

23. Display glass of mobile phones for accident dosimetry using thermoluminescence measurements
-Michael Discher

25. Long-term effects of acute low-dose ionizing radiation on the neonatal mouse heart
-Mayur Bakshi

27. Anti-inflammatory acylphloroglucinol derivatives and involved biosynthesis enzyme in strawberry fruit
-Chuankui Song

29. Pyrazole ligandoside: Preparation, characterization, replication and more
-Meng Su

31. The nucleo-cytoplasmic partitioning of an ADP-ribosylation-recognising protein is regulated upon DNA damage
-Barbara Golia

33. Improving tendon healing by the use of human mesenchymal stem cells
-Chi-Fen Hsieh

35. Development of a dynamic model of the alveolar interface for the study of aerosol deposition
-Cei Daniele

37. Multispectral optoacoustic imaging - Molecular imaging engineering for translational medicine
-Subhamoy Mandal

39. Hepatitis C virus protein crystallization in outer space
-Hanaa Gaber

Immunology/Cancer/Epigenetics

41. T cell re-direction against Glypican-3 for immunotherapy of hepatocellular carcinoma (HCC)
-Christina Dargel

43. Ubiquitylation in the regulation of heterochromatic boundaries
-Marta Forn Bernaus

45. Structural studies on the Drosophila Polycomblike protein
-Jeongyoon Choi

47. FancA overexpression and radiation resistance in cell lines of head and neck squamous cell carcinoma
-Igor Gimenez-Aznar

49. α1-Antitrypsin regulates IL-8 release and CXCR2 expression in human neutrophils
-Nupur Aggarwal

51. Comparative global characterization of microRNA-expression in radiation-associated and sporadic breast carcinomas
-Christina Maria Wilke

53. Endocytosis drives *Kras*-dependent tumor initiation in the pancreas
-Clara Lubeseder-Martellato

Protein-Biochemistry/"-omics"

55. External regulators of the proteasome in human platelets
-Katharina Gründler

57. Making protein structures discoverable
-Maria Kalemanov

Pathways/Receptors/ Biochemistry

59. Agonist and mechanically induced receptor activations evoke distinct active receptor conformations
-Serap Erdogmus

61. Bayesian analysis of a series of FRAP experiments with mixed-effect priors
-Martina Feilke

63. Characterization of $G_{i/o}$-coupled receptors using a cAMP-sensor based FRET and a Kir channel based electrophysiological approach
-Julie Straub

65. Mesenchymal cells regulate growth of intestinal crypts by a Wnt independent mechanism in 3D culture system
-Agnieszka Pastula

67. Investigation of self-renewal and senescence of tenomodulin-deficient tendon stem/progenitor cells
-Sarah Dex

Poster Index session II: 03:50 pm - 04:55 pm

Neuroscience

2. How to stay smart with chronic stress
-Anja Kretzschmar

4. Long-term changes in hippocampal and cortical synaptogenesis after brain irradiation in young mice
-Stefan J. Kempf

6. Consequences of genetic ablation of proliferating NG2-glia in the adult mouse brain
-Sarah Schneider

8. New insights into the reaction of mouse oligodendrocyte progenitor cells after brain injury by live in vivo imaging
-Axel von Streitberg

10. A critical period for adaptive plasticity of neuronal circuits during postnatal mouse development
-Michaela S. Helmbrecht

12. Molecular mechanisms of circuitry organization and adaptive plasticity
-Maria Castiblanco

New methods/ Translational Biochemistry

14. Short time exposure to hydrogen peroxide induces a persistent glutathione export from neurons
-Michaela C. Hohnholt

16. Protein-monolayer interactions investigated by fluorescence microscopy and correlation spectroscopy
-Alena Khmelinskaia

18. Predicting contact distance probabilities using statistical modeling of amino acid pair couplings
-Susann Vorberg

20. Visualization of specific DNA sequences in living stem cells with a programmable fluorescent CRISPR/ Cas system
-Tobias Anton

22. Photosynthetic dermal scaffold for skin regeneration
-Myra-Noemi Chávez

24. Small-angle neutron scattering (SANS) of the segmentally deuterated human multi-domain splicing factor T-cell intracellular antigen-1 (TIA-1)
-Miriam Sonntag

26. Evaluation of scaffolds for the delivery of mesenchymal stem cells *in vivo*
-Elizabeth A. Wahl

28. Analysis of the molecular mechanism of chloroplast dimorphism in *Bienertia sinuspersici*
-Vinay Shekhar

30. Comparative study of different induction systems in amiR SNF4 plants showing conditional deregulation of SnRK1 proteins
-Raksha Ravikumar

32. Of cis, trans and feedback regulation: impact on genetic variation on nearby genes
-Daniel Bader

34. Measuring intermolecular FRET between talin-1 and (meta-)vinculin in living cells and sstablishing methods to measure intracellular FRET using organic dyes
-Anna-Lena Cost

36. Immediate prediabetes and diabetes in women after gestational diabetes is associated with obesity, disordered eating behavior and low physical fitness

-Marina Fugmann

38. Multispectral optoacoustic tomography: A highly promising optical imaging modality for clinical arthritis imaging

-Christian Lutzweiler

Immunology/Cancer/Epigenetics

40. The function of CD83 in the thymus

-Julia von Rohrscheidt

42. The MENX rats as an animal model for medullary thyroid carcinoma

-Nikolai Falk

44. Immune evasion of hepatitis delta from CD8+ T cell immune response

-Hadi Karimzadeh

46. YadA mediated virulence in Yersinia is dependent on its collagen adherence, but not its autoagglutination capacity

-Nicolas Jäger

48. DNA damage activates the chromatin remodeler ALC1 through ADP-ribosylation

-Hari Raj Singh

50. Clinical manifestations of complicated and uncomplicated malaria in Aligarh, India

-Umme Asma

52. Transcriptional and translational library of the naïve T cell surface - An advanced view on the early activation process

-Anke Gräßel

Protein-Biochemistry/"-omics"

54. SLC26A2 in primary aldosteronism

-Tarik Bozoglu

56. Structure and RNA-binding properties of the Not1–Not2–Not5 module of the yeast Ccr4–Not complex

-Varun Bhaskar

58. Identifying individual differences of fluoxetine response in juvenile rhesus monkeys by metabolite profiling

-Ying He

Pathways/Receptors/ Biochemistry

60. Investigating the chemistry behind red wine astringency - What is the role of protein/polyphenol interactions?

-Judith Delius

62. Phospholipase C-ε (PLCε) induced TRPC6 activation: A common but redundant mechanism in podocytes

-Jana Demleitner

64. Modified messenger RNA and its application in bone tissue engineering

-Mehrije Ferizi

66. The role of Cab45 in secretory cargo sorting at the Trans Golgi Network

-Birgit Blank

68. Transcriptome surveillance by selective termination of noncoding RNA synthesis

-Björn Schwalb

01 Neuroscience
Pathway and biomarker discovery in a posttraumatic stress disorder mouse model

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Posttraumatic stress disorder (PTSD) is caused by exposure to a traumatic event. Individuals diagnosed for PTSD not only experience significant functional impairments but also have higher rates of physical morbidity and mortality. The neurobiological pathways involved in the development of PTSD remain obscure. Using a PTSD mouse model, we have established a proteomic platform to study affected molecular mechanism. In addition, we have investigated the effects of chronic treatment with fluoxetine, an antidepressant used in PTSD therapy. The PTSD mouse model is generated by two electric foot shocks to emulate an aversive encounter. Shocked mice develop PTSD-like symptoms, including hyperarousal and conditioned fear following 28 days of incubation. PTSD-like symptoms in shocked mice were ameliorated after 4 weeks of fluoxetine treatment. Specific brain regions including prelimbic cortex (PrL), basolateral amygdala (BLA), central nucleus of amygdala (CeA), CA1 and nucleus accumbens (NAc), were punched from intact PTSD and control mouse brains. Punched tissue specimens were pooled and cytosolic and membrane associated proteins were subjected to mass spectrometry analysis using stable isotope metabolically labeled reference material to analyse protein expression level differences. Our preliminary results indicate that energy metabolism and cytoskeleton assembly are dysregulated in PTSD mice. In addition, pathway analyses indicate a significant down-regulation of the citrate cycle in the NAc of PTSD mice. Chronic fluoxetine treatment of PTSD mice reversed the downregulated expression of proteins that are part of these pathways. The proteomic data delineate PTSD dysfunctional pathways with the ultimate goal to improve diagnosis and treatment.

02 Neuroscience
How to stay smart with chronic stress

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Chronic stress is not particularly favorable: the release of cortisol causes a general state of arousal, suppression of the immune system, a reduction of hippocampal volume and dendritic arborization (de Kloet et al., 2005). However, there could be something good in response to chronic stress: the protein DRR1.
Being previously known as a potential tumor suppressor, DRR1 was recently characterized as a direct link between stress, actin dynamics, and behaviour. It was shown to localize to actin-rich cellular structures and, in neurons, primarily to synapses. Although it decreases neurite outgrowth and reduces LTP magnitude and spine density, mice with viral DRR1-overexpression show enhanced cognitive performance (Schmidt et al., 2011). These findings suggest a protective function of DRR1 during stress counteracting its adverse effects. Its relevance becomes evident as failing of stress coping imposes an increased risk for depression, anxiety or post-traumatic stress disorder.
Currently we are dissecting the molecular mechanism and synaptic function of this intriguing protein. It exerts a three-fold effect on actin dynamics by bundling filaments, inhibiting their elongation but also enhancing nucleation of new filaments. Assembly and reorganization of actin filaments is a key process for synaptic transmission and cell motility. However, up to now, a profound mechanistic understanding of the pathway from stress to neuronal reorganization and behaviour remains elusive.
Assuming that the mechanism of DRR1 is not only significant for coping with chronic stress but also during tumor development and progression, elucidating DRR1's mechanism of action could contribute to several physiologically relevant processes.

Neuroscience

Angiogenesis promoting proteins in pituitary adenomas

03

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Pituitary adenomas are frequent intracranial tumors that often associate with the hypersecretion of pituitary hormones or may be non-secreting (nonfunctioning pituitary adenomas, NFPA). Tumors resembling human NFPAs develop with complete penetrance in rats affected by the multiple endocrine neoplasia syndrome, MENX. This syndrome is caused by a germline loss of function mutation in p27Kip1.
Gene expression array analysis performed in our group identified a considerable number of genes deregulated in rat pituitary tumors compared to normal pituitary tissues. Some of the deregulated transcripts are associated with angiogenesis, including vascular endothelial growth factor (Vegf), angiopoietin-1 (Ang-1), -2 (Ang-2) and angiopoietin like-2 (Angptl2). VEGF, ANG-2 and ANGPTL2 were found to promote angiogenesis in several tumor types, while ANG-1 inhibits it and stabilizes mature vessels. We analyzed mRNA and protein expression changes of these 4 genes in the pituitary adenomas of MENX-affected rats and compared the results with similar analyses conducted on the corresponding human tumors. We could show that Ang-2 and Angptl2 mRNAs were highly expressed in almost all of the rat pituitary adenomas and were enhanced in less than the half of the human NFPAs. Vegf mRNA was up-regulated in MENX-rats but not in NFPAs. At the protein level, we have so far set up the immunohistochemical staining for Ang1 on both rat and human tumors. We could show that rat pituitary adenomas and human NFPAs show reduced cytoplasmic Ang-1 staining compared to adjacent non tumor cells. Ang-2 and Angptl2 immunohistochemical staining will follow.

Neuroscience

Long-term changes in hippocampal and cortical synaptogenesis after brain irradiation in young mice

04

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There is a great concern about the detrimental long-term effects of ionising radiation (IR) exposure. Focussing on the brain, children are in particular susceptible to ionising radiation as they have still an immature brain. Our aim is to enlighten these mechanisms on the behavioural and molecular level. Thus, mice were exposed to total body irradiation on postnatal day 10 with doses of 0, 0.02, 0.1, 0.5 and 1.0 Gy. Behavioural analysis showed alterations in cognitive function after 2 and 4 months post-irradiation. As cortex and hippocampus are centres for cognition and learning, we have studied IR-induced changes in these two compartments 7 months post-irradiation. Using mass-spectrometry, we observed changes in signalling of RhoGDI, RHO family GTPases and Ephrin B at 0.5 Gy and 1.0 Gy in both regions indicating aberrant synaptogenesis. Rac1 is a key RHO family GTPase and we demonstrated a significant decrease in the expression level at 0.5 and 1.0 Gy via immunoblotting. Further, we obtained via quantitative PCR a significant upregulation of miRNAs involved in synaptogenesis/ spine size regulation, namely miR-132 and miR-134. Interestingly, both miRNAs are linked to neurocognitive disorders and dendritic spine morphology as they are involved in the Rac1-Cofilin-Actin remodeling pathway.

05 Neuroscience
Epigenomic alterations in glioblastoma

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Glioblastoma (GBM) is the most frequent malignant brain tumor of adults. About 10%-15% of all intracranial tumors are glioblastoma. In Europe and North America, there are about 2-3 cases per 100,000 inhabitants. The World Health Organization (WHO) classifies glioblastoma as Grade IV, meaning that patients have a very poor prognosis: Most patients die within months. Despite intensive research, the mechanisms leading to this highly malignant brain tumor are still unknown.vBut there is increasing evidence that epigenetic alteration contribute to tumorigenesis. Till 2009, 5-methylcytosine (5mC) was the only known base modification of mammalian genomes. It is known that 5mC can lead to a functional inactivation of gene transcription. Then, in 2009 Kriaucionis et al. identified a new base modification: 5-hydroxymethylcytosine (5hmC). This poses numerous new questions: What is the biological meaning of 5hmC? What is its role in tumorigenesis? Is 5hmC the “missing link” in active demethylation? As an approach to understand the function of 5hmC we investigated both normal brain tissue as well as tissue derived from glioblastoma. We quantified the amount of 5hmC in normal brain and glioblastoma tissue. Additionally, we correlated expression data of proteins that are associated with methylation and possible active demethylation processes. We observed that there is a significantly lower amount of 5hmC in glioblastoma compared to normal brain tissue. Additionally we noticed that there are dysregulations in methylation and demethylation pathways in glioblastoma tissue compared with normal brain.

06 Neuroscience
Consequences of genetic ablation of proliferating NG2-glia in the adult mouse brain

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NG2-glia, also known as oligodendrocyte progenitor cells (OPCs) are the only proliferating cells outside the neurogenic niches in the adult brain and constitute a major fraction (5-10%) of the brain cells at this age. Despite their high number and substantial characterization, their actual function in the adult brain is vastly unknown. To address this question, we used conditional genetic deletion of the Esco2 protein, which triggers apoptosis of dividing cells during M-phase in the inducible Sox10-iCreER^{T2}xCAG-eGFPxEsco2^{fl} mouse line (Simon et al. 2012, Whelan et al. 2012), to specifically ablate proliferating NG2-glia in the adult brain.

Our results showed that deletion of the Esco2 in NG2-glia induced ongoing NG2-cell death that was partially compensated by the enhanced proliferation of non-recombined cells. Notably, specifically in the White matter (WM) of the cortex, we could observe a decrease in the number of newly generated oligodendrocytes (OL). Interestingly, these animals developed progressive motoric deficits, whereas control littermates maintained their abilities.

As the proliferation of NG2-glia is >15 fold increased at 3 days after stab wound injury (Simon et al. 2011), we reasoned this lesion could play an important role also in this context. Indeed we could observe a transient reduction of NG2-glia around the lesion site what seemingly influenced the reaction of other glial cell types. Our data suggest that NG2-glia are important for myelin maintenance in the physiological, and scar formation in the pathological brain, therefore giving first insights into their role in the adult brain.

Neuroscience

The bone morphogenic protein 7 (Bmp7) plays a pro-tumorigenic role in pheochromocytoma

07

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Despite the fact that a number of genes involved in phaeochromocytoma (PC) have been identified, the molecular pathways involved are not well understood. Rats carrying a germline loss-of-function mutation in p27 (MENX-syndrome) develop bilateral PC. Gene expression profiling of these tumors revealed up-regulation of the bone morphogenic protein Bmp7. Interestingly, BMP7 is overexpressed in 88% of sporadic and 69% of familial PCs. To investigate the effect of Bmp7 on the tumorigenic characteristics we performed in vitro assays including MTT, Boyden chamber and migration assay on the mouse PC cell lines MPC and MTT (high Bmp7), and on the rat PC cell line PC12 (low Bmp7). Up- or down- regulation of endogenous Bmp7 altered the tumorigenic phenotypes. Proliferation, migration and invasion were enhanced by the up-regulation of the protein in the PC12 cells, while down-regulation of BMP7 impairs these properties in MPC and MTT cells. Moreover, knock-down of the high endogenous Bmp7 levels in primary MENX-PC cells, reduced viability compared to control transduced cells. Since BMP7 stimulates the migration/invasion of PC cells, we elucidated the molecular mechanisms mediating these effects. The ectopic overexpression of Bmp7 enhanced both the expression of integrin β1 and phosphorylated-AKT (p-AKT) levels.
In conclusion, we observed that Bmp7 promotes the tumorigenic phenotype in PC cells by the activation of integrin β1 and p-AKT signaling. BMP7 represents a novel target for therapy of PC since the knock-down in vitro shows promising impairment of the tumorigenic phenotype.

Neuroscience

New insights into the reaction of mouse oligodendrocyte progenitor cells after brain injury by live in vivo imaging

08

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After acute injury in the adult brain, oligodendrocyte progenitor cells (OPCs), also referred to as NG2-glia, react by hypertrophy and proliferation. However, key aspects of their dynamic behavior, can only be monitored by live imaging. To elucidate these key aspects of OPC behavior after injury, we used repetitive in vivo two-photon laser scanning microscopy (2pLSM) to follow NG2-glia after smaller and larger stab wound injuries in the mouse somatosensory cortex. Live imaging revealed that the majority of OPCs reacts within 2 days after injury with hypertrophy, polarization towards the injury site, directed migration towards the injury site and proliferation. Only a small proportion of cells within ~500 µm of the injury did not react in any detectable manner. We noted that polarization and hypertrophy occur rather fast after inflicting the injury, while proliferation peaks 4 days after injury. Taken together, these observations support the concept of OPC heterogeneity and reveals new insights into the functional role of these cells after injury: the fast process orientation towards the injury site implies a contribution to wound closure and their substantial proliferation sometimes for several rounds amplifies the number of NG2-glia surrounding the injury site with implications for scar formation.

09 Neuroscience
Early-life stress interacts with genetic predisposition in a mouse model of affective disorders

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Exposure to stress during early life presents a risk factor for affective disorders. To mimic the clinical situation of a genetic predisposition interacting with environmental stress, we exposed animals of the stress-reactivity mouse model to a period of early-life stress (ELS) and measured the effects on relevant parameters later in life. The mouse model consists of three breeding lines, selectively bred for high (HR), intermediate (IR) or low (LR) HPA-axis reactivity in response to stressors.
Animals of the three lines were exposed to moderate ELS from postnatal day 2-9. The adult mice were then tested for effects of the ELS-treatment on physical condition, stress-coping behaviour and neuroendocrine function. The development of the pup's bodyweight confirmed that the ELS manipulation was effective in all mouse lines. Moreover, a significant interaction between genetic background and environment was apparent: Compared to unstressed HR mice, ELS HR mice showed a hyperactive stress-coping style, accompanied by strong even further increased HPA-axis reactivity. In this study, we demonstrate a clinically relevant gene-environment interaction of genetic vulnerability and early-life stress. Our model can be a powerful tool to gain further insight into the role of early-life adversity in affective disorders and the underlying molecular processes.

10 Neuroscience
A critical period for adaptive plasticity of neuronal circuits during postnatal mouse development

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The development of the central nervous system involves numerous molecular and physiological events that constitute key elements for the correct wiring of connections, which later will determine the functionality of the organism. Previous studies have revealed that absence of Sema3F-Neuropilin-2 signaling during embryonic development leads to specific axon guidance defects that alter the dorsal-ventral choice for motor axon outgrowth at the base of the limb, thereby impairing motor coordination. Therefore, Sema3F mutants provide a suitable model to analyze potential compensatory mechanisms of neuronal plasticity. In the grid walk test the number of slips as well as the time to cross the ladder were significantly increased, with improved performance during normal development. These improvements in motor coordination were further boosted by enriched environment housing starting at birth, but not starting at 4 weeks of age. These results suggest a critical period for postnatal adaptive plasticity in motor circuits. Additionally, neuroanatomical and functional analyses confirm the proposed plastic adaptions in Sema3F mutants. Interestingly, changes in excitatory synapses were found between the different environmental housing groups. Thus, we propose that perineuronal nets are formed in the spinal cord and close the critical period for adaptive plasticity by reducing the ability to form new synapses with affected motor neurons.

Neuroscience

How to make a photoreceptor neuroscience needs?

11

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In recent years “optogenetics” has been tremendously successful in providing new tools for a better grasp of how our brains function. While the field’s efforts first focused on the usage of photoreceptors from algae and their heterologous expression in mammalian neurons, more recent advances have also involved naturally occurring mammalian neuronal receptors. The two approaches used in optochemical genetics that involve chemically synthesized photoisomerizable molecules as a means of photoswitching the receptor, are based on (a) photochromic ligands (PCLs) or (b) photoswitchable tethered ligands (PTLs), with complementary fields of application such as photopharmacology for PCLs and circuitry mapping for PTLs. However, all efforts to come up with a tool that provides a means of naturally mimicking the depolarization of inhibitory neurons have failed so far. With this in mind, we started off reengineering the chimera α7/GlyR consisting of the ligand binding domain of an α7 acetylcholine receptor and the ion pore domain of a glycine receptor. The chimera α7/GlyR is a chloride channel that exhibits the postsynaptic depolarization of naturally occurring glycine receptors in inhibitory neurons. Thus, a couple of reprogrammed chloride channels could be engineered that are very promising as light-dependent hyperpolarizing tools, exceeding all known hyperpolarizing optogenetical tools.

Neuroscience

Molecular mechanisms of circuitry organization and adaptive plasticity

12

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The development of the central nervous system involves numerous molecular and physiological events that constitute key elements for the correct wiring of connections, which later will determine the functionality of the organism. In order to successfully establish communications between distant regions, different guidance molecules of either attractive or repulsive nature guide axons over long distances to create functional synapses with specific targets. Semaphorin 3F (Sema3F), together with its receptor Neuropilin-2, mediate a crucial repulsive interaction involved in axon guidance, neural differentiation and plasticity. Our group has previously shown that loss of Sema3F induces alterations in the organization of the motor pools in the spinal cord that correlate with impairments in motor coordination. However, not only motor neurons in the spinal cord, but also intraspinal and supraspinal circuits play a critical role in the coordination of movements. Therefore, the aim of the present study is to determine the effects caused by the lack of Sema3F on the organization of intraspinal circuits as well as the motor cortex and its projections by (1) morphological and anatomical analysis employing the rabies virus encoding eGFP as a retrograde neurotracer and (2) intracortical stimulation to determine the functional output of the respective cortical areas.

13 Neuroscience
Measuring the human startle reflex with simultaneous EMG and fMRI

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The startle reflex is a bodily response to protect vital body parts from immediate threat. One component of this is the contraction of the muscles around the eyes, which can be measured by placing electrodes near these muscles (electromyography; EMG), and which is commonly used as a readout for fear and anxiety responses in both humans and animal models. Behavioural fear learning studies have demonstrated an increased startle magnitude in response to aversive stimuli in healthy subjects, as well as an increased magnitude in anxiety patients. Additionally, functional neuroimaging (fMRI) studies using fear learning paradigms have found altered patterns of activity in brain structures related to the fear network. By simultaneous measurement of EMG and fMRI, it is possible to correlate the startle magnitude to the changes in brain activity after presentation of a threat-related stimulus. This way, the severity of anxiety can be related to patterns of brain activity, which is interesting to study in anxiety patients and in fear learning paradigms. We present here a method to perform the technically very challenging combined EMG/fMRI recording, which has only been shown in a few studies. In addition, we show the first results of the correlation of anxiety severity to brain activity.

14 New methods/Translational Biochemistry
Short time exposure to hydrogen peroxide induces a persistent glutathione export from neurons

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Hydrogen peroxide (H_2O_2) is a normal byproduct of the cellular metabolism and can in excess cause oxidative stress and cell damage. We investigated the consequences of an exposure of cultured cerebellar granule neurons to H_2O_2. Exposure to up to 60 μM H_2O_2 did not affect cell viability after 4 h. In the absence of H_2O_2, neurons released only low amounts of their cellular glutathione, while exposure of the cells to H2O2 caused a time- and concentration-dependent glutathione export. Within 4 h of incubation with H_2O_2, the extracellular glutathione content was elevated up to 4-fold compared to control cells. This was accompanied by a matching decrease of the cellular glutathione contents. The glutathione export was significantly elevated for H_2O_2 concentrations above 30 μM and became maximal after application of 100 μM H_2O_2. The stimulated glutathione export remained elevated after removal of H_2O_2 by catalase after 5 min exposure to H_2O_2, demonstrating that a persistent presence of H_2O_2 was not required for maintenance of stimulated glutathione export. In summary, a short time exposure of viable neurons to micromolar concentrations of H_2O_2 induces a prolonged accelerated export of cellular glutathione which may enhance the sensitivity of neurons towards oxidative stress.

New methods/Translational Biochemistry

MemConP: Accurate prediction of membrane protein contacts and helix interactions

15

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Membrane proteins, which form about 25-30% of the proteome, are important molecules in many biological processes like signal transduction or molecule recognition. Despite their abundant occurrence in the proteome, the databases contain only about 1% solved membrane protein structures, making computational prediction of their structural features an important task.

MemConP is a machine learning based tool for prediction of residue-residue contacts and helix interactions in α-helical membrane proteins. As there were significant improvements in calculating correlated mutations in the last years, we combined one of the best performing methods, current membrane protein structure data and a modern database search algorithm to create a superior prediction tool. The applications of the method's results can be manifold, ranging from using the predicted topology for membrane protein classification tasks to creating constraints for de novo structure prediction.

Using the common contact definition of a distance less than 5.5Å and the machine learning device's default threshold, our tool reaches precision/recall values of 51.0%/24.4% for residue-residue contacts and 86.2%/67.5% for helix interactions. Performance values for the commonly used L/5 threshold are 64.0%/12.1% and 96.2%/43.2% respectively.

New methods/Translational Biochemistry

Protein-monolayer interactions investigated by fluorescence microscopy and correlation spectroscopy

16

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Lipid bilayers are composed of a wide variety of lipids and proteins which can lead to spatial heterogeneity and formation of domains, so-called 'lipid rafts', which play an important role in cell signaling. Model membrane systems have become invaluable tools to investigate these specific features of cellular membranes. Although a variety of experimental assays exist, many of them are rather complicated in their preparation and difficult in their practical realization. Here, we use a new simple miniaturized monolayer assay combined with confocal fluorescence microscopy and fluorescence correlation spectroscopy (FCS). This approach allowed us to investigate morphology and lipid fluidity of the monolayer and to correlate these factors with protein binding. Particularly, we studied the influence of phase separation on Cholera toxin β and Streptavidin binding to lipid monolayer. Our results show that the affinities of Cholera toxin β and Streptavidin to the monolayer depend on lipid surface density. Moreover, FCS measurements indicate a correlation between higher protein binding and increased lipid diffusion.

17 New methods/Translational Biochemistry
Seeing is believing – How to visualize a new type of membrane scaffold

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Autophagy is a catabolic pathway that delivers cytoplasmic components to the lysosome for degradation. During macroautophagy, a de novo generated membrane sack progressively encloses cytoplasm, giving rise to a double-membrane autophagosome after sealing. Numerous proteins orchestrate the formation, closure, and fusion of autophagosomes with lysosomes. Two Ubiquitin-like conjugation systems are involved in these processes: one involves the conjugation of ubiquitin-like Atg12 to Atg5, which forms a constitutive complex with the coiled coil protein Atg16. The second conjugation system covalently attaches Atg8 to the lipid phosphatidylethanolamine (Atg8-PE) in autophagic membranes. Previous studies in our lab using Atg-proteins from S. cerevisiae indicated that upon association of Atg12–Atg5-Atg16 with Atg8-PE at membranes, immobile multimeric complexes are formed, which might represent a new kind of membrane scaffold. We thus set out to challenge this hypothesis by trying to visualize the structure. We reconstituted Atg8-conjugation and scaffold-formation on supported lipid bilayers, which were characterized by Atomic Force Microscopy (AFM). Our investigations demonstrated that Atg8-PE represents a membrane anchor, which is cross-linked by the Atg12–Atg5-Atg16 complex, forming a two dimensional membrane scaffold with meshwork-like architecture. A quantitative analysis of our AFM-data revealed important insights into the molecular structure of the scaffold.

18 New methods/Translational Biochemistry
Predicting contact distance probabilities using statistical modeling of amino acid pair couplings

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Methods to accurately predict protein structures are becoming indispensable as experimental procedures are not capable of closing the constantly widening sequence-structure gap. Precise determination of contacting residues is of great benefit for structure prediction in various ways, e.g. for ranking of homology models or as distance constraints for structure prediction tools like 'Modeler'. Contact prediction is based on the concept of correlated mutations: an existing functional or structural constraint that is violated by a mutated residue can be reconciled by a compensating mutation of a nearby residue. Using statistical models these correlated mutations can be detected from large Multiple Sequence Alignments (MSAs) to retrieve evidence for evolutionary conserved contacts. As of now, the full potential of information encoded in MSAs has not been exploited by available methods. We enhance our predictions by explicitly modeling amino acid pair specific couplings derived from a global statistical model of the MSA. Furthermore we overcome the commonly used 8 Å C-beta distance definition for a contact. This rigid threshold is not biologically profound, as interacting residues operate at much broader distance ranges. We refine the prediction output turning a binary contact definition into a probabilistic continuous distance measure.

New methods/Translational Biochemistry

Annotation of directed genomic states unveils variations in the Pol II transcription cycle

19

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DNA replication, transcription and repair involve the formation of protein complexes that undergo transitions in their composition as they progress along the genome. Average occupancy profiles of DNA-bound proteins over genes have been instrumental to understand these processes. However, averaging requires predefined gene sets, hides gene-specific variations, and obscures transitions that do not occur at fixed distances from the aligned gene feature. To overcome these limitations, we introduce bidirectional HMMs which infer directed, genomic states from occupancy profiles de novo. Application to RNA polymerase II-associated factors in yeast identifies 32 new transcribed loci and uncovers gene-specific variation of the transcription cycle. The analysis indicates a regulated initiation-elongation transition, the absence of elongation factors Ctk1 and Paf1 from a class of genes, a distinct transcription mechanism for highly expressed genes, and novel DNA sequence motifs associated with transcription termination. We anticipate bidirectional HMMs to significantly improve analysis of genome-associated directed processes.

New methods/Translational Biochemistry

Visualization of specific DNA sequences in living stem cells with a programmable fluorescent CRISPR/Cas system

20

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DNA contains, beside genetic, also epigenetic information in form of complex modifications and higher order structures with sequence-dependent functional properties. Labeling and tracing of specific sequences in living cells has been a major challenge in studying spatiotemporal dynamics of native chromatin. Here we repurposed the prokaryotic CRISPR/Cas adaptive immunity system to specifically detect endogenous genomic loci in vivo. We constructed catalytically inactive version of the Cas9 endonuclease, fused it with eGFP (dCas9-eGFP) and co-expressed small guide RNAs (gRNAs) to target specific DNA sequences without endonucleolytic cleavage. To validate this approach we designed gRNAs to label pericentric, centric and telomeric repeats, which are enriched in distinct nuclear structures. With major satellite specific gRNAs we obtained a characteristic chromocenter pattern, while gRNAs targeting minor satellites and telomeres highlighted smaller foci coinciding with Centromere Protein-B and Telomeric Repeat-Binding factor-2, respectively. DNA sequence specific labeling by gRNA/dCas9-eGFP complexes was directly shown with 3D-Fluorescent in Situ Hybridization. Structured Illumination Microscopy of gRNA/dCas9-eGFP expressing cells revealed chromatin ultrastructures and demonstrated the potential of this approach for chromatin conformation studies by super resolution microscopy. The programmable dCas9 labeling system opens new perspectives to study the spatiotemporal dynamics of endogenous DNA sequences during cell cycle progression and differentiation.

21 New methods/Translational Biochemistry
Interaction of buckminsterfullerene (C60) with *T. thermophila*: Phenotypic changes due to nanomaterial

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Nanomaterials are the new hot cake in the application science because of their incredible properties and multiple applications in human life. Buckyminsterfullerene (Fullerenes) and its derivatives (Fullerenol) belong to carbon-based nanomaterial and their current use is ahead of their toxicological profile. They are used for various purposes ranging from drug delivery to solar cell to coating of surfaces. And there is raising concern because of their ability to induce interfering oxidative and cytotoxic responses in cells- posing a threat both for health and environment. The Tetrahymena genus is fresh water ciliate, typified by an ovoid body shape and T. thermophila are usually about 50×20cm in size. In this investigation, we observed (video and picture) the behavior (swimming) of the protozoa toward the fullerene and fullerenol nanoparticles at different time points-3, 6, 12, and 24 h. Tetrahymena thermophila SB210 shows increased swim speed to eat the more nanoparticles-food or perhaps to neutralize the negative effect of particles. This behavior changes with time. We tried to connect cell's behavior with its molecular response by testing membrane integrity, oxidative assays and dopamine release. We further explore, if Tetrahymena can be a model organism for Parkinson's disease? And to what extend?

22 New methods/Translational Biochemistry
Photosynthetic Dermal Scaffold For Skin Regeneration

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Tissue engineering has opened a new therapeutic avenue that promises a revolution in regenerative medicine. However, to date, the regenerative potential of engineered tissues is limited and the clinical results are disappointing. This is mainly attributed to the unsolved problem of poor vascularization and the resulting hypoxia of engineered tissues. Here we propose an alternative source of oxygen to blood vessel-perfusion by using photosynthetic scaffolds. We have demonstrated that the unicellular photosynthetic algae *Chlamydomonas reinhardtii* can be cultured in scaffolds for dermal regeneration showing high biocompatibility and photosynthetic activity. Moreover, *C. reinhardtii* can be co-cultured with fibroblasts, decreasing the hypoxic response to low oxygen culture conditions. Upon engraftment of the photosynthetic scaffolds in a full skin defect in nude mice, symbiotic tissues composed of algae and murine cells were generated. The results obtained here represent the first step towards engineering human autotrophic tissues.

New methods/Translational Biochemistry

Display glass of mobile phones for accident dosimetry using thermoluminescence measurements

23

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In case of radiation exposure by radiological events a technique is needed to determine the absorbed dose of individuals, if no personal dosimeter is available. Previous studies have demonstrated that certain components of a mobile phone (electronic components of the circuit board, filler material of the SIM card chip, glass samples extracted from the display screen) are sensitive to ionizing radiation and suitable as accident dosimeters using optical stimulated luminescence (OSL) and thermoluminescence (TL) methods. For glass samples dosimetric properties were investigated in detail with regard to signal stability, signal bleaching, dose response and optimization of the minimum detectable dose. Dose recovery tests were successfully carried out using irradiation trials under realistic conditions. The reconstruction of the absorbed dose using glass display is a competitive alternative to dose assessment techniques using other electronic components of the mobile phone and provides a useful option for retrospective accident dosimetry.

New methods/Translational Biochemistry

Small-angle neutron scattering (SANS) of the segmentally deuterated human multi-domain splicing factor T-cell intracellular antigen-1 (TIA-1)

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Alternative splicing generates protein variants with different, sometimes antagonistic functional properties. T-cell intracellular antigen-1 (TIA-1) is an apoptosis promoting factor that modulates alternative splicing of various pre-mRNAs including the human Fas receptor. TIA-1 is a multi-domain protein which contains three RNA-recognition motifs (RRMs) and a C-terminal glutamine-rich domain. Central RRM2 and RRM3 domains associate with cognate sites in fas pre-mRNA introns, whereas the role of RRM1 still remains elusive. To characterize RNA binding of TIA-1, we are investigating structures of different TIA-1 constructs in the presence of RNA. Information about the multi-domain arrangements and dynamics is essential to understand biological activity of these proteins. Therefore, we apply complementary methods like solution state NMR and small angle X-ray/neutron scattering. However, NMR studies of multi-domain proteins are often challenging due to signal overlap, line-broadening and spectral complexity. One approach to overcome these technical difficulties is to exclusively isotope-label one segment of the studied multi-domain protein. Additionally, deuterating individual domains within a multi-domain complex can provide information on the relative domain positions by SANS. The deuterated domain can be located within the envelope using contrast variation methods by adjusting the ratio of H_2O to D_2O in the buffer. We present segmental labeling strategy for selectively isotope-labeling only one domain within the TIA-1 RRM2-RRM3 construct. We applied an enzyme-based approach to record NMR and SANS data of this tandem construct in the absence and presence of RNA.

25 New methods/Translational Biochemistry
Long-term effects of acute low-dose ionizing radiation on the neonatal mouse heart

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Epidemiological studies establish that children and young adults are especially susceptible to radiation-induced cardiovascular disease (CVD). The biological mechanisms behind the elevated CVD risk following exposure at young age remain unknown. The present study aims to elucidate the long-term effects of ionizing radiation by studying the murine cardiac proteome after exposure to low and moderate radiation doses.

NMRI mice received single doses of total body ^{60}Co gamma-irradiation on postnatal day 10 and were sacrificed 7 months later. Changes in cardiac protein expression were quantified using Isotope Coded Protein Label (ICPL) and tandem mass spectrometry (LC-MS/MS).

We identified 32, 31, 66, and 34 significantly deregulated proteins after doses of 0.02, 0.1, 0.5 and 1.0 Gy respectively. The four doses shared 9 deregulated proteins. Bioinformatics analysis showed that most of the deregulated proteins belonged to a limited set of biological categories, including metabolic processes, inflammatory response, and cytoskeletal structure. The transcription factor PPAR-alpha was predicted as a common upstream regulator of several deregulated proteins.

This study indicates that both adaptive and maladaptive responses to the initial radiation damage persist well into adulthood. It will contribute to the understanding of the long-term consequences of radiation-induced injury and developmental alterations in the neonatal heart.

26 New methods/Translational Biochemistry
Evaluation of scaffolds for the delivery of mesenchymal stem cells in vivo

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Mesenchymal stem cells (MSCs) have been shown to improve tissue regeneration in several pre- and clinical trials. These cells have been used in combination with three-dimensional scaffolds as a promising approach in the field of regenerative medicine. In this work, we compare the behavior of human adipose-derived MSCs seeded on four different biomaterials that are commonly used in clinical settings to determine the most suitable one for delivering the cells to wounds. MSCs were isolated, characterized, and seeded onto scaffolds constructed from bovine collagen, fibrin, chitosan, and decellularized porcine dermis. Chick chorioallantoic membranes (CAM) provide an in vivo analysis of the angiogenic potential of factors released from the cell seeded biomaterials. Results showed that the composition of the scaffolds strongly influence key parameters of the cells such as, seeding efficiency, cellular distribution, attachment, survival, metabolic activity, and paracrine release. This work provides in vitro and in vivo information for clinical translation and optimization for the use of MSCs in FDA approved biomaterials for tissue regeneration.

New methods/Translational Biochemistry

Anti-inflammatory acylphloroglucinol derivatives and involved biosynthesis enzyme in strawberry fruit

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Strawberries are one of the most important fruits in daily life of human and may have beneficial effects against oxidative stress mediated diseases. Berries contain multiple phenolic compounds, which are thought to contribute to their biological properties.
The aim of this study was to isolate and elucidate the structure of interesting bioactive metabolites in strawberry fruits by HPHC-MS/MS, NMR and to investigate the putative biosynthesis enzymes that are involved in the biosynthesis of the new metabolites.
Besides, I succeeded in the cloning and characterization of genes that are involved in the biosynthesis of these new metabolites. Three chalcone synthases (FvCHS) were cloned from Fragaria vesca, and the enzymatic properties of those FvCHS were investigated after heterologous expression in BL21(DE3)pLysS cells. The enzymes catalyze the transformation of isovaleryl-CoA or isobutyryl-CoA with three molecules of malonyl-CoA to form phlorisovalerophenone or phlorisobutyrophenone, respectively that are intermediates in the biosynthesis of the acylphloroglucinol derivatives. Besides, FvCHS enzymes also catalyzed the formation of naringenin-chalcone from malonyl-CoA and p-coumaroyl-CoA. Substrate specificity of the FvCHS enzymes was also studied.
Downregulation of the chalcone synthase gene in strawberry plants resulted in a decrease of the acylphloroglucinol derivatives.

New methods/Translational Biochemistry

Analysis of the molecular mechanism of chloroplast dimorphism in *Bienertia sinuspersici*

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Bienertia sinuspersici is one of four currently known Single-cell C4 species. Surprisingly for a C4 plant it lacks two different cell types forming Kranz anatomy, which was considered as a characteristic feature of C4 plants. Dimorphic chloroplasts, separated spatially in individual chlorenchyma cells, enable these plants to perform C4 photosynthesis. Previous studies have shown differential accumulation of C4 photosynthetic enzymes in the two different chloroplast types. This is unusual because *B. sinuspersici* chlorenchyma cells have one nucleus but two chloroplast types. Thus, all chloroplast targeted proteins are under transcriptional control of one nucleus. The mechanism that targets chloroplast protein into two different chloroplasts is currently unknown. To understand the mechanism, in vivo localization studies with selected *B. sinuspersici* proteins were conducted. Therefore cDNA sequences of five *B. sinuspersici* genes were amplified from isolated *B. sinuspersici* RNA and fused to GFP in a binary vector. After Agrobacterium mediated transformation in *B. sinuspersici* and Nicotiana tabacum leaves, cells were isolated and observed under a scanning laser confocal microscope for subcellular localization of the GFP fusion proteins. Initial results show equal presence of GFP fusion proteins in both chloroplast types or in the cytoplasm indicating that they have not been targeted differentially.

29 New methods/Translational Biochemistry
Pyrazole ligandoside: Preparation, characterization, replication and more

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For decades, nucleic acid mimics have aroused continued interest of chemists and biologists, shown impressive applications in molecular biology and pharmaceutical sciences. Design, synthesis and construct of novel nucleic acid analogs for multifunctional complex and for expanding the genetic alphabet are of tremendous interest. Previously, our group has reported the profile of metal-salen base pair. Here, we present a novel pyrazole ligandoside. The ligandoside is based on 2-(1H-pyrazol-1-yl)phenol (Pz). After standard solid-phase synthesis, biophysical properties are tested by UV and CD spectroscopy. The melting temperature of the duplex depends on pH and transition metal-ion species. The neutral Pz–Cu^{2+}–Pz base pair is slightly more stable than a canonical G:C base pair and the insertion preserves the duplex in B-form DNA. Furthermore, Pz-self-base pairs are able to complex five and ten metal ions in a row in DNA-like structure. When Pz is mixed with salen ligandoside, copper ion shows a preference for Pz pair. To expand the genetic code, Pz monomer is used in a replication system. The ligandoside shows a good performance in extending the primer to full length with Therminator polymerase. The crystal structure of Pz pair with polymerase is solved.

30 New methods/Translational Biochemistry
Comparative study of different induction systems in amiR SNF4 plants showing conditional deregulation of SnRK1 proteins

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Sucrose-nonfermenting1-related protein kinase 1 (SnRK1) is a protein kinase of the evolutionarily conserved AMPK family (AMP activated protein kinase). While AMPKs are found in animals, SnRK1s are present in plants and are involved in physiological adaptation of plants to stress by regulating gene expression in response to energy depletion. SnRK1s are heterotrimeric enzymes consisting of a catalytic α (KIN10/11), a substrate targeting β (KINβ1/2/3), and an activating γ subunit (KINβγ/SNF4). Studies have shown that SnRK1 plays a very important role in the overall plant growth and development. Its activity is tightly regulated by dynamic phosphorylation of its catalytic subunit, but the exact molecular mechanism of its regulation, in particular, the post-translational regulation of SnRK1 proteins is still not clear. An artificial micro RNA inducible (amiR) system was developed for conditional silencing of SNF4 in the model plant Arabidopsis thaliana. Upon incorporation, deregulation of the SNF4 subunit of the SnRK1 proteins is caused. In this study, various types of induction systems and their impact on both the wild type and the amiR SNF4 induced plants were determined. Three types of induction systems were employed and their impact on sugar levels, starch levels, nitrate reductase activity and transcript levels were determined.

New methods/Translational Biochemistry

The nucleo-cytoplasmic partitioning of an ADP-ribosylation-recognising protein is regulated upon DNA damage

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DNA damage is a threatening phenomenon for cellular life, with several pathways having been evolved to cope with it. A number of mechanisms regulate the DNA damage response, including specific post-translational modifications and regulated protein compartmentalization. ADP-ribosylation is a common post-translation modification that has been implicated in a number of repair pathways. We have discovered a protein able to recognise ADP-ribosylation that, upon DNA damage, is both recruited to the site of DNA damage, and exported from the nucleus. We focus on the regulation of such nuclear export, addressing it through the use of live cell imaging, complemented with biochemical and proteomic approaches. Using these tools we have found that nuclear export is dependent on PI3K-like kinases, a family of kinases known to be master regulators of the DNA damage response. In particular, inhibition of Ataxia-Telangiectasia-Mutated (ATM) is sufficient for the complete abolishment of the nuclear export. Consistent with this, a number of SQ motifs, the preferred consensus of PI3K-like kinases, are present within the protein, with their mutation being sufficient for the failure of partitioning. These findings are an important starting point for the understanding of the upstream events of this particular DNA damage-induced nucleo-cytoplasmic partitioning of proteins.

New methods/Translational Biochemistry

Of cis, trans and feedback regulation: impact on genetic variation on nearby genes

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The vast majority of regulatory genetic variants are found in close vicinity of the regulated gene. The distinction between cis- and trans-acting variants is a fundamental starting point to understand the mechanisms underlying these regulatory variants. Typical cis-regulatory variants affect transcription factor binding sites or RNA stability. Local trans-regulations have been less studied and include feedbacks, an essential regulatory feature of biological systems. To understand the contribution of cis- and trans-regulation and their potential interplay, we devised a novel experimental design in which allele-specific expression in a hybrid cross of two distant yeast strains is compared to allele-specific expression in a pool of segregants of the same cross. Our statistical model of the allele-specific RNA count data is based on generalized linear models for RNA-seq counts. Moreover, we control for allele selection in the pool population by robustly estimating allele frequency from genomic DNA sequence of the pool. We found significant differences of cis-effects among major gene categories (essential, non-essential, and non-coding). Furthermore, our results shed light on the effects of feedback in buffering or enhancing the impact of genetic variation on gene expression.

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New methods/Translational Biochemistry

Improving tendon healing by the use of human mesenchymal stem cells

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In this study, we investigated the effect of human mesenchymal stem cells (hMSC) on Achilles tendon repair. HMSC have been suggested as a promising cell type for tissue engineering of mesenchyme-derived tissues, such as cartilage, bone and tendons. We implicated a defect model in Rattus norvegicus, consisting of a surgical removal of three millimetres tissue from the Achilles tendon proper. The defect was then reconnected by suture and was filled by a 3-dimenssional cell pellet. An empty defect group (ED, without cell implantation) was prepared in parallel as control. The Achilles tendons were harvested for histological and molecular biological evaluation at 16 weeks after surgery. Our analysis of periodic acid schiff staining revealed that the mucopolysaccharide content in both, ED and hMSC groups, suggests a primary stage of remodelling in these animals. Next, based on safranin O and alcian blue stainings (spesific for proteoglycans), we found residual cartilaginous areas in ED group, which were almost absorbed in the hMSC group. The regenerated tendon-like tissues hMSC-treated tendons showed also a higher birefringence of aligned collagen fibres by polarized light microscopy. In conclusion, the addition of human mesenchymal stem cells promoted better repair of ruptured Achilles tendon.

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New methods/Translational Biochemistry

Measuring intermolecular FRET between talin-1 and (meta-)vinculin in living cells and sstablishing methods to measure intracellular FRET using organic dyes

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Integrin-based focal adhesions (FAs) are complex subcellular structures mediating cell adhesion to the extracellular matrix (ECM) and consisting of several hundred proteins. One key component in FAs is talin, an integrin binding protein essential for integrin activation and the connection to the f-actin cytoskeleton. Another critical adhesion protein is vinculin, which is recruited by talin-1 to FAs mediating additional binding to the cytoskeleton. Even though the talin-vinculin interaction is considered crucial for proper cell adhesion and mechanosensitivity, it is still unknown where and when this interaction takes place. In my master thesis, I used Förster resonance energy transfer (FRET) to measure the talin–vinculin interaction in living cells. I can show that talin-1 and vinculin as well as talin-1 and metavinculin, a muscle-specific splice isoform of vinculin, predominantly interact in FAs but not in the cytoplasm of cells.

In the second part of my master thesis, I established a technique to specifically label intracellular proteins with commercially available organic dyes. This method was successfully used to perform fluorescence lifetime imaging microscopy (FLIM)-measurements in cells and may be a first step towards single-molecule experiments in cells.

New methods/Translational Biochemistry

Development of a dynamic model of the alveolar interface for the study of aerosol deposition

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Interactions of nano and micro particles from the air with pulmonary epithelia (lung) are complex processes, mediated by the humidity of the alveolar environment as well as the diaphragm contraction. How this movement contributes to nanoparticle deposition and uptake by the epithelial wall is not clearly understood, partly because of the lack of appropriate models which can recapitulate the alveolar microenvironment.
To study the long and short term effects of aerosol phase nanoparticles on pulmonary tissue in the presence of cyclic motion, we developed an air-liquid interface bioreactor with a mobile elastic membrane to simulate physiological lung muscle stretching. The system consists of an aerosol generator with a moving membrane placed between an air-liquid interface.
Lung tissue is cultured on the stretchable porous membrane, activated by an electro-pneumatic regulator. The membrane is sandwiched between two compartments. The mechanical properties of the membranes were evaluated in dry and wet conditions as well as their ability to undergo reversible cyclic deformation for long term experiments.
A prototype based on the CFD (computational fluid-dynamic) model was realized in PDMS. The system was characterized in terms of aerosol deposition efficiency: a fluorescein solution was nebulized and particle concentrations on the membrane and in the liquid compartment were evaluated.

New methods/Translational Biochemistry

Immediate prediabetes and diabetes in women after gestational diabetes is associated with obesity, disordered eating behavior and low physical fitness

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Women after gestational diabetes (GDM) are at high risk for developing type 2 diabetes (T2D). Shortly after delivery, a subgroup of these women already shows prediabetes, defined as impaired fasting glucose (IFG) and/or impaired glucose tolerance (IGT) or even T2D. The knowledge of risk factors for this early deterioration of glucose metabolism could enable personalized lifestyle interventions.

In this cross-sectional explorative interim analysis, we looked at differences between women after GDM grouped by an oral glucose tolerance test (oGTT) into normoglycemic (NGT) and prediabetic/diabetic individuals (IFG/IGT/T2D). We measured body composition (body mass index, body fat and waist circumference), eating behavior (the Three Factor Eating Questionnaire, the Eating Disorder Examination and a Food Frequency Questionnaire) and physical fitness (spiroergometry). Group comparisons between NGT and IFG/IGT/T2D were analyzed using Mann-Whitney-U test and correlations were analyzed by Spearman coefficient. 74 women after DGM received an oGTT between 3.4 and 15.7 month after delivery. Our data suggest that overweight, disordered eating patterns and low physical fitness contribute to early prediabetes/diabetes after GDM. Consequently, nutrition advice, psychological counseling and encouragement of physical activity would be the major intervention tools to prevent or at least to slow down the development of T2D in this group of women.

37 New methods/Translational Biochemistry
Multispectral optoacoustic imaging - Molecular imaging engineering for translational medicine

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Optoacoustics is an emerging new hybrid bioimaging modality which combines acoustical detection and EM absorption contrast. Optoacoustic imaging not only provides structural but also functional information of biological tissues. Multi-Spectral Optoacoustic Tomography (MSOT) is capable of high resolution three dimensional (3D) visualizations of molecular probes located deep in scattering living tissues, with resolution and speed representative of ultrasound. This method can simultaneously deliver anatomical, functional and molecular information with both high resolution and penetration capabilities. We have recently developed a portable spherical array probe for volumetric real-time optoacoustic imaging at centimeter scale depths, which has successfully provided superior imaging speed and suitability for the 3D visualization of tissues, and also yielded detailed in-vivo volumetric images on a mesoscopic level. The system enables us to conduct intravital imaging of tumor masses and internal organs of small animal, and study perfusion profile in real time. Such studies allow us to investigate hypoxia and nutrition gradients as well as cell viability, proliferation and drug response potentials – which is vital in understanding the dynamics of living tissues and disease prognosis and progression. Further, the handheld approach allows convenient handling of both pre-clinical experiments as well as clinical measurements in human subjects. Hopefully this innovation will be instrumental in aiding the translation of the powerful and promising MSOT technology from the laboratory to clinical practice – and allow us to 'hear the molecules' in action.

38 New methods/Translational Biochemistry
Multispectral optoacoustic tomography: A highly promising optical imaging modality for clinical arthritis imaging

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Besides the established clinical imaging modalities like Computed Tomography or Magnetic Resonance Imaging, optical imaging modalities, owing to their versatile contrast, have greatly enriched clinical and basic biological discovery. However, due to the strong light scattering in living tissues, spatial resolution of the optical method quickly deteriorates when deep tissue imaging at the millimeter to centimeter penetration scale is considered. Multispectral Optoacoustic Tomography (MSOT) overcomes this resolution degradation by means of detection of acoustic pressure waves generated via the optoacoustic effect. MSOT is therefore capable of yielding anatomical, functional and molecular images with spatial resolutions on the order of several tens to several hundreds of microns deep in strongly scattering tissues. Thus far, MSOT has mainly been applied in small animal ex vivo and in vivo basic research studies, further assisted with functional and molecular agents. Clinical applications of the optoacoustic method are also emerging, e.g. in in vivo breast cancer studies or intravascular imaging applications. Herein, we briefly introduce the fundamentals of MSOT and some of its recent application developments and further showcase clinical potential of the method for visualization of rheumatoid arthritis.

New methods/Translational Biochemistry

Hepatitis C virus protein crystallization in outer space

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Hepatitis C virus (HCV) is a worldwide problem. It is estimated that 170 million individuals are infected with hepatitis C Virus. About 150 million people are chronically infected, and more than 350000 people die every year from hepatitis C-related liver diseases.
Egypt is reported to have the highest number of infected individuals worldwide due to the use of contaminated parental anti-schistosomal therapy. The dominant genotype in Egypt is HCV genotype 4. Nearly 100 crystal structures of HCV NS5B have been reported, covering genotypes 1a, 1b, 2a and 2b, although all structures lack the C-terminal membrane-anchoring tail, but not for genotype 4. Crystallization methods for proteins have been the subject of decades of development, yet protein crystallization remains the limiting step in structural studies. One of the used techniques is to perform the crystallization experiment in a Microgravity environment (Space shuttle or on the ISS). Gravity is considered a non-preferable condition for protein crystallization as it raises the convection of the samples. Some of the carried out experiments introduced better results than the ground based experiments, but still that is not always the case. We expect that crystallizing a certain type of HCV protein can give a clear, large crystal which shall be used designing new specific drugs for HCV genotype 4.

Immuology/Cancer/Epigenetics

The function of CD83 in the thymus

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The main players of adaptive immunity, Tcells, are established in the thymus. The thymic microenvironment accomplishes the generation of a highly diverse and self-tolerant Tcell repertoire in two steps: during positive selection cortical thymic epithelial cells (cTEC) allow thymocytes which have successfully rearranged their T cell receptor to proceed to negative selection, which is mediated by medullary TEC by deleting auto-reactive T cells. Mice lacking the CD83 gene show a severe reduction in $CD4^+$ T cells and reduced MHCII surface levels. The importance of CD83 for proper Tcell development is known, but its actual function is still unresolved. We addressed the question if the reduced MHCII level of cTEC is causative for the impaired $CD4^+$ Tcell development or if CD83 has additonal MHCII-independent functions, such as cell-to-cell signalling. To test this, we introduced truncated versions of CD83 into TEC. This fast and new method enables us to introduce genes of interest into TEC and study their effects in vivo. Using this method, we could demonstrate that reconstitution of the transmembrane domain of CD83 alone being suffient to rescue normal $CD4^+$ T cell development in the $CD83^{-/-}$ RTOC, whereas the extracellular domain is dispensable for positive selection of thymocytes. Therefore, we can exclude signalling in trans via CD83 being essential for Tcell development.

41 Immuology/Cancer/Epigenetics
T cell re-direction against Glypican-3 for immunotherapy of hepatocellular carcinoma (HCC)

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A new therapeutic approach for HCC is adoptive T-cell therapy. Glypican-3 (GPC3) as a tumour associated antigen is expressed in up to 60% of all HCC but not in healthy human liver tissue. Our goal is to isolate GPC3-specific T cell receptors and to express them in T cells to render them capable of recognizing and eliminating GPC3-expressing HCC.

Immunodominant epitopes for GPC3 have not been described. We used Ultra-Nano-HPLC coupled on-line to the Q exactive mass spectrometer to obtain a comprehensive HLA class I peptidome from a GPC3+/HLA-A2+ hepatoma cell line. Two predominant HLA-A2 bound GPC3 peptides were identified and used to target GPC3 epitopes that are presented on HCC cells.

To isolate tumour reactive T-cells, an allo-restricted stimulation approach was used. Dendritic cells from HLA-A2 negative donors were co-transfected with GPC3 and HLA-A2 RNA and used to stimulate and expand T-cells from the autologous naïve T-cell repertoire. We detected and expanded MHC-streptamer-positive $CD8^+$ T-cells specific for both targeted GPC3 epitopes, and grew T-cell clones from them. Adoptive T-cell therapy using GPC3-redirected T-cells provides a promising new approach for treatment of HCC.

42 Immuology/Cancer/Epigenetics
The MENX rats as an animal model for medullary thyroid carcinoma

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Medullary thyroid cancer (MTC) originates from the parafollicular C-cells of the thyroid gland. Hereditary MTCs carry RET mutations and are part of the multiple endocrine neoplasia (MEN) syndrome. MTC is one of the few malignancies with rising incidence and accounts for 8-15% of all deaths related to thyroid cancers because it has a high tendency to metastasise. C-cells do not concentrate radioactive iodine nor are sensitive to chemo-therapy. Therefore, total thyroidectomy is the current method of treatment. Animal models of MTC carrying RET specific mutations have been created, but their phenotype does not closely reflect the human disease. We recently identified a variant of the MEN syndromes that spontaneously developed in a Sprague-Dawley rat colony.
The main aim of this study is to determine the validity of the MENX-associated thyroid tumours as a model of human MTCs. The initial histological evaluation shows a high degree of homology between MENX associated thyroid lesions and human MTCs. Additionally, the MENX thyroid tumours were investigated at the molecular level by investigating their microRNA expression profile. A comparison of microRNA signatures between rat thyroid tumours and human MTC lesions showed similar expression patterns, making MENX rats a suitable model of MTC.

Immuology/Cancer/Epigenetics

Ubiquitylation in the regulation of heterochromatic boundaries

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The genetic information of eukaryotes is packaged into chromatin. This DNA-protein complex plays an important role, not only in compacting the genome inside the nucleus, but also in regulating gene accessibility and expression. The genome can be partitioned into active euchromatin and inactive heterochromatin domains. Ubiquitylation plays an important role in chromatin regulation and is not restricted to monoubiquitylation of histones H2A and H2B. The addition of ubiquitin also marks chromatin factors for degradation. In fact we are interested in studying the role of ubiquitylation in shaping the chromatin domains through proteolysis.
In *Schizosaccharomyces pombe* the boundary factor Epe1 prevents spreading from heterochromatin into neighboring euchromatin. Epe1 is selectively ubiquitylated and degraded within heterochromatic domains, confining his presence to the heterochromatic boundaries. Now, we seek to understand the mechanisms that protect Epe1 from being degraded at the boundaries. Currently, we focus on the possible role of deubiquitylating enzymes (DUBs) in reversing the ubiquitylation of Epe1 at the boundaries and deleted different DUBs in a reporter strain that contains the ura4 gene outside the boundary of the heterochromatic mating locus. We determined the spreading of heterochromatin by in vivo silencing assays and by quantitatively assessing the expression levels of the reporter gene. Our preliminary results suggest an involvement of specific DUBs in the control of heterochromatin-euchromatin boundaries. The ongoing experiments aim to determine the relation of these DUBs with Epe1 regulation and other chromatin factors.

Immuology/Cancer/Epigenetics

Immune evasion of hepatitis delta from CD8+ T cell immune response

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The immunopathology of hepatitis delta virus (HDV) infection remains unclear. However, CD8+ T-cell response seems to play a key role in the outcome of the infection. In the presence of cytotoxic T lymphocyte (CTL) immune response, those viruses with mutations within MHC class I restricted epitopes are able to evade effector T-cell recognition, resulting in chronic infection. Consequently, the aims of this Europe-wide study were: analysis of the genotype(s); characterization of the variability of the large hepatitis delta antigen (L-HDAg) and identification of HDV immune escape from CD8+ T-cell response. The sequences of L-HDAg from 146 chronically infected patients, and their HLA class I backgrounds were determined. Almost all of the sequences were branched under genotype 1. A higher frequency of mutations in a known HLA-B27 epitope and in a predicted HLA-A24 epitope in HLA-B27/A24 positive patients was detected, as compared to negative patients. Longitudinal studies indicate that the mutation rates depend on the time of onset of the super-infection. There was a higher mutation rate shortly after the onset of infection. The results suggest that immune escape from CD8+ T-cell response may contribute to the persistence of HDV in chronically-infected patients, and to the evolution of circulating HDV isolates.

45 Immuology/Cancer/Epigenetics
Structural studies on the *Drosophila* Polycomblike protein

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Polycomblike (Pcl) protein is an accessory component of Polycomb Repressive Complex 2 (PRC2), which methylates histone H3 at lysine K27 (H3-K27) to repress its target genes. The Pcl protein is required for generating high-levels of H3-K27 tri-methylation in the chromatin of Polycomb target genes by helping to anchor PRC2 at Polycomb Response Elements (PREs) of these target genes. However, the mechanism by which Pcl binds to PREs and how it tethers PRC2 are not well understood. Using X-ray crystallography, we determined the structure of the *Drosophila* Pcl protein construct containing the second PHD finger and an adjacent winged-helix domain ($Pcl_{PHD2\text{-}WH}$). In addition, we present biophysical analysis showing that the winged-helix domain of Pcl binds to DNA in a sequence non-specific manner with moderate binding affinity for PRE DNA. An equivalent human Pcl orthologue PHF1 protein construct (i.e. $PHF1_{PHD2\text{-}WH}$) also binds to DNA with similar affinity. Using DNA probes containing CpG di-nucleotides that were either methylated or non-methylated, we find that PHF1 binds both kinds of probes with comparable affinity.
Taken together, these studies establish that *Drosophila* and human Pcl proteins are DNA-binding proteins and suggest that this DNA-binding activity might contribute to PRC2 anchoring at PREs in Polycomb target genes.

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YadA mediated virulence in *Yersinia* is dependent on its collagen adherence, but not its autoagglutination capacity

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Yersinia enterocolitica and *Yersinia pseudotuberculosis* are enteropathogenic bacteria causing diseases ranging from self-limiting diarrhea to sepsis. In *Y. enterocolitica* but not in *Y. pseudotuberculosis*, the trimeric autotransporter *Yersinia* adhesin A (YadA) is an essential factor for mediating mouse virulence. The N-terminal tip domain (NTTD) of YadA shows high sequence variability between different *Yersinia* species, serotypes and strains. To study the impact of these differences on binding to extracellular matrix (ECM) proteins, autoagglutination and mouse virulence, we introduced genes of YadA species variants and NTTD deletion constructs into the *Yersinia* virulence plasmid pYV and transformed them into the *Y. enterocolitica* strain WA314. Our studies in an intraperitoneal mouse infection model showed that NTTD is crucial for virulence in *Y. enterocolitica* YadA but not in *Y. pseudotuberculosis* YadA. Deleting parts of the NTTD alters the autoagglutination behavior of the strains. Also NTTD-dependent changes in the characteristics of binding to different ECM proteins, like collagen and laminin, were detected. Modifications of the binding affinity of *Y. enterocolitica* YadA to ECM proteins seem to reduce mouse virulence to a higher degree than the same modifications in *Y. pseudotuberculosis* YadA. Interestingly, changes in autoagglutination seemed to have no impact at all.

Immuology/Cancer/Epigenetics

FancA overexpression and radiation resistance in cell lines of head and neck squamous cell carcinoma

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Head and neck squamous cell carcinoma (HNSCC) is worldwide one of the most frequent cancer types. Despite ongoing developments tumors are not always responding properly to therapies leading to a poor overall patient survival. Especially radioresistancy of tumors represent an important problem that causes therapy failure; thus, the identification of molecular markers predicting radiation response of HNSCC would allow the improvement of therapy by the application of personalized therapy approaches. DNA gains on chromosome 16q23-24 have been shown to correlate with a reduced progression-free survival rate of HNSCC patients after radiotherapy. The FancA gene, a member of the FA/BRCA pathway, is located on chromosome 16q24.3 and has been proposed as potential biomarker for HNSCC radiation response. Here, FancA overexpressing cells have been generated in order to elucidate the effect of FancA alterations on radiation response *in vitro*. FancA overexpressing cells showed enhanced radioresistance compared to control cells, supporting a role of FancA in the mechanisms leading to radiation resistance in HNSCC. Additionally, data on cell viability, senescence as well as measurements of DNA repair activity after *in vitro* irradiation prove a radioprotecting effect of FancA overexpression.

Immuology/Cancer/Epigenetics

DNA damage activates the chromatin remodeler ALC1 through ADP-ribosylation

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ATP-dependent chromatin remodeling enzymes alter nucleosome structure and facilitate transcription, replication and DNA repair. However, little is known about how DNA damage triggers their activity. ALC1 is a unique chromatin-remodeling enzyme whose activity is strictly regulated by a dynamic posttranslational modification (Poly-ADP ribosylation). ALC1 harbors a C-terminal macrodomain and an N-terminal snf2 like ATPase domain separated by a linker. This modular architecture provides a way to couple DNA damage-induced poly-ADP ribosylation (PARylation) with chromatin remodeling. The macrodomain mediates PARylation dependent recruitment to sites of DNA damage and the ATPase domain is thought to remodel chromatin at the damage site. Here, we show that the ATPase domain and the macrodomain physically interact. The interaction is lost upon PARylation, both in vivo and in vitro, and is dependent on the intact ADP-ribose binding pocket of the macrodomain. Also, poly-ADP ribose but not mono-ADP ribose is necessary and sufficient for the loss of interaction. We further observe that there are at least two bridging segments between the ATPase and the macrodomain. In addition, our fluorescence lifetime measurements indicate conformation rearrangements within ALC1 upon DNA damage dependent on intact ADP-ribose binding pocket. In summary, our data together with previous studies indicates that the macrodomain not only acts as a recruitment module but is also the repressor, activator and processivity factor during different stages of remodeling reaction. Our data thus supports the emerging concept of remodeler activation through intra-molecular interactions.

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α1-Antitrypsin regulates IL-8 release and CXCR2 expression in human neutrophils

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The modulation of the neutrophil CXC chemokine receptor-2 (CXCR2) is an important mechanism for controlling neutrophil responsiveness. The expression of CXCR2 is rapidly down-modulated by lipopolysaccharide (LPS) via tyrosine kinase-dependent mechanism, whereas IL-8 down-modulates surface CXCR2 expression through rapid receptor internalization. Acute phase protein α1-antitrypsin (A1AT) is a known regulator of neutrophil responses to LPS; however, A1AT's effect on CXCR2 expression has not been investigated. Priming of human neutrophils with LPS caused a time-dependent decrease in surface CXCR2 levels and reduction in CXCR2 mRNA. At 1h, A1AT enhanced while at 4h it abolished the effect of LPS on CXCR2 expression. Similarly, when compared to IL-8 primed cells, addition of A1AT caused an initial enhanced internalization and a subsequent re-expression of surface CXCR2 without affecting CXCR2 mRNA. Remarkably, A1AT enters into the neutrophil rapidly, induces transient ERK1/2 activation and localizes into plasma membranes, cytosol and secretary vesicles pushing out IL-8. Pretreatment with U0126, a highly specific MEK1/2 inhibitor, abolished A1AT induced ERK1/2 activation and IL-8 release, and attenuated effect of A1AT on CXCR2 expression in LPS or IL-8-primed neutrophils. Our data show that A1AT modulates surface CXCR2 expression via activation of ERK1/2 pathway and concomitant modulation of IL-8 release.

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Clinical manifestations of complicated and uncomplicated malaria in Aligarh, India

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Malaria remains as an important public health problem globally. Over a period of 2 years we observed patients admitted in the JNMC, Aligarh and collected 360 samples. *Plasmodium falciparum*, *Plasmodium vivax* and mixed infections happened to 64, 34 and 2 percent during years 2011-2012. The peak transmission period was observed following rainy season during the month of September and October which continued till November. Fever with chills, abdominal pain, vomiting and cough were the most frequent symptoms and were more prominent in falciparum malaria as compared to *P. vivax* infection. Thrombocytopenia, Anemia, splenomegaly, and respiratory distress were noticed in 69, 39, 60, 10% and 47, 15, 35, 3% *P. falciparum* and *P. vivax* cases. Jaundice was observed in 15 and 4% cases of *falciparum* and *vivax* malaria. Cerebral malaria was observed in only a few patients having *P. falciparum* infection but no fatality was observed.

Immuology/Cancer/Epigenetics

Comparative global characterization of microRNA-expression in radiation-associated and sporadic breast carcinomas

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Breast cancer is one of the most common cancers worldwide. Beside risk factors such as age and lifestyle, exposure to ionizing radiation is known to enhance the breast cancer risk. MicroRNAs are small noncoding RNAs that control gene expression at the post-transcriptional level. Further, microRNAs are now being used as prognostic and predictive biomarkers for human cancer. Present study aims at identification of microRNAs that are specifically deregulated in radiation-associated breast cancer. Here, we applied a case-case study design by investigating global microRNA-expression profiles using Agilent microarrays. Formalin-fixed paraffin-embedded clinical breast cancer samples from 46 radiation-exposed and 48 non-exposed Ukrainian breast cancer patients from the same region were analyzed. Most of the exposed patients worked as clean-up workers after the Chernobyl disaster at the site of accident. Amongst 1205 analyzed microRNAs, hsa-miR-4299 and hsa-miR-196a-5p were found to be differentially expressed and were validated using qRT-PCR. These microRNAs are the first potential radiation biomarkers in human breast cancer. We are now setting up a study for the validation of these microRNAs in an independent set of samples. In case of positive validation, the identified microRNAs could serve as predictive and prognostic biomarkers of radiation-induced breast cancer and as targets in personalized therapy approaches. Moreover, above results provide an important step towards elucidating molecular mechanisms of radiation-induced breast cancer.

Immuology/Cancer/Epigenetics

Transcriptional and translational library of the naïve T cell surface - An advanced view on the early activation process

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The cell surface is the bridge between the environment and the cell itself. To gain more knowledge about processes taking place during T cell differentiation, it is essential to monitor these changes of the surface composition. Since allergies and asthma are an increasing problem and it is known that an imbalanced differentiation process of naïve T cells (the common progenitor for all Th cell subsets) could be a reason for this, we wanted to elucidate this process in a more complete way. By combining RNA and corresponding surface protein expression data from naïve and activated T cells, we created a surface library of naïve T cells. This hypothesis-free analysis enabled us to identify 140 cell surface proteins on naïve and activated T cells, partly not described in an immunological context before. Functional studies of these proteins are ongoing to clarify their role on the surface of naïve T cells. The Liquid chromatography-tandem mass spectrometry (LC-MS/MS) based cell surface capturing (CSC) technology was used to enrich and identify cell surface glycoproteins and their quantitative changes during activation (αCD3/αCD28). The transcriptional state of naïve and activated T cells was investigated by microarray based gene expression profiling (8x60K) and analysis with GeneSpring Software (Agilent).

53 Immuology/Cancer/Epigenetics
Endocytosis drives *Kras*-dependent tumor initiation in the pancreas

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In pancreatic ductal adenocarcimoma (PDAC), activating *Kras* mutations drive cancer initiation. Acinar cells undergo acinar-ductal metaplasia (ADM) and develop to PDAC in an epidermal growth factor receptor (EGFR)-dependent manner. We addressed the role of endocytosis in PDAC using primary 3D acinar epithelial explants from conditional mouse models based on $Kras^{G12D}$ (CK) and EGFR deletion (CKE). CK acinar explants mimic ADM *ex vitro* in an EGFR-dependent manner. Endocytosis increased during ADM in CK, but not in CKE, explants. Pharmacological inhibition of endocytosis inhibited ADM in a dose-dependent manner and reduced P-ERK activation, suggesting that endocytosis is required for sustained ERK signaling. Furthermore, endocytosis induced by hypoosmotic shock in explants lacking EGFR rescued ADM phenotype independently of *Kras* and induced P-ERK activation in presence of $Kras^{G12}$. In cancer cells, endocytosis was required for cell proliferation independent of EGFR and $Kras^{G12D}$, although cells lacking $Kras^{G12D}$ displayed reduced basal endocytosis. Blocking endocytosis did not compromise the growth of $Kras^{G12D}$-pancreatic tumor xenografts. Thus, we hypothesize that EGFR is essential for maintenance of the endocytic compartment, a *conditio sine qua non* for ADM. Additionally, endocytosis may be required together with oncogenic $Kras^{G12D}$ for sustained ERK signaling in acinar cells undergoing ADM, but not for tumor maintenance

54 Protein-Biochemistry/"-omics"
SLC26A2 in primary aldosteronism

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The molecular mechanisms that lead to autonomous aldosterone secretion are necessary to find biomarkers and therapeutic targets for primary aldosteronism. We have investigated SLC26A2, a gene at a locus that was associated with aldosterone to renin ratio in a genome-wide association study among participants of a population based survey. This gene encodes a sulfate transporter and had no reported adrenal function, yet we observed significantly high murine adrenal gland expression, which was down-regulated in vivo with angiotensin II and potassium. Also, SLC26A2 expression was lower in aldosterone producing adenomas compared to normal adrenal glands. A knockdown of the gene in adrenocortical NCI-H295R cells drastically increased aldosterone production as well as upstream events such as CAMK pathway and intracellular $Ca2^{+}$ content. Further substantiation of these observations came from a knock out mouse model which presented increased aldosterone output in a gender specific manner. These observations indicate that a possible impact of SLC26A2 in aldosterone secretion regulation may be present in the etiology of primary aldosteronism.

Protein-Biochemistry/"-omics"

External regulators of the proteasome in human platelets

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The ubiquitin-proteasome systems (UPS) is the major degradatory pathway for proteins. It not only maintains protein quality control but is also involved in critical cellular processes such as apoptosis, antigen presentation and the development of neurodegenerative and vascular diseases. Recently a proteasome complex has also been identified in platelets, however its regulation and biological function are not well known. In this work, we focus on the regulation of the 20S and 26S proteasome in human platelets and aim to identify novel regulators of the platelet proteasome.

We initially demonstrated that most genes of the UPS and of the proteasome subunits are expressed in platelets. Chymotrypsin-, trypsin- and caspase-like activity of the 26S and 20S proteasome was quantified in lysed platelets. Interestingly, catalytic activity differed among the proteasome subunits while trypsin-like activity of the 20S proteasome was the highest. Epoxomicin robustly inhibited the platelet proteasome and resulted in an increased accumulation of poly-ubiquitinated protein. To mimic an infectious disease scenario, we incubated $1x10^8$ platelets with $4x10^6$ *E. coli* bacteria for 4h and detected increased activity of the proteasome and accelerated degradation of polyubiquitinated proteins. This effect was reversible with inhibition of the proteasome. Supporting these findings, platelets isolated from sepsis patients reveal an overexpression of the proteasome activating subunit PSME1. We identified novel mediators that activate the proteasome in platelets. This data may help in understanding the functional role of the proteasome in platelets, its role for platelet functions and platelet-related diseases.

Protein-Biochemistry/"-omics"

Structure and RNA-binding properties of the Not1–Not2–Not5 module of the yeast Ccr4–Not complex

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Deadenylation is the first and rate-limiting step in mRNA turnover, an important step in gene expression. The evolutionarily conserved Ccr4-Not complex is the central molecular machinery that catalyzes the processive deadenylation of mRNA poly(A) tails. The Ccr4-Not complex is a multisubunit complex organized around a large scaffold protein, Not1. The complex has a modular architecture with at least two distinct modules: a "deadenylase module" and a "Not module". Previous studies have revealed the structure and have dissected the activities of the deadenylase module. To explore the function of the Not module, we have mapped the interacting domains of the yeast Not1, Not2 and Not5 proteins. We have determined the 2.8 Å structure of this ternary complex that forms the core of the Not module. The structure shows how the Sm-like domains of Not2 and Not5 dock on to an extensive HEAT-repeat region in Not1. Disrupting the interactions within the ternary complex shows severe growth defects in yeast. Based on structural information and *in vitro* assays, we found that the ternary complex forms a composite surface that binds poly(U) RNA, with a site at the Not5 Not-box. Our results suggest that the Not module forms a versatile platform for macromolecular interactions.

57 Protein-Biochemistry/"-omics"
Making protein structures discoverable

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Compare the ca. 32 Million protein sequences deposited in UniProt with the fewer than 90,000 solved structures in PDB, and you realize they constitute only 0.28%! Nevertheless, for about half of all proteins, some three-dimensional (3D) structural information is available, since they are significantly similar to proteins with known 3D structure. However, many scientists fail to benefit from this information because it is difficult to access and use. Out of this need was born SRS 3D, a module of SRS, that allows users to easily and rapidly find all related structures for a given target sequence. Structures can then be viewed together with sequences, alignments and sequence features. Over the course of the last year, we set out to update, redesign and extend this service, which we plan to present under the name Aquaria.

58 Protein-Biochemistry/"-omics"
Identifying individual differences of fluoxetine response in juvenile rhesus monkeys by metabolite profiling

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Fluoxetine (Prozac®) is a psychopharmacological agent that is commonly used therapeutically in a variety of developmental disorders. However, therapeutic response shows high individual variability and severe side effects in children (e.g. hyperactivity and cognitive abilities). Using metabolomics to study fluoxetine response in juvenile rhesus monkeys, this work aims to 1) identify biomarkers for the response to fluoxetine and find a potential role of monoamine oxidase A (MAOA) polymorphisms in altering the treatment response, and 2) evaluate "side effects" of fluoxetine therapy during juvenile development and detect metabolites associated with behavior. Juvenile male rhesus monkeys with high- or low-MAOA genotype were treated daily with fluoxetine or vehicle. After one year, monkeys were assessed for impulsivity (reward delay) and blood and cerebrospinal fluid (CSF) specimens were collected. The samples were analyzed with a GC-TOF-MS platform. Our results show that metabolomics can discriminate fluoxetine- from vehicle-treatment in both plasma and CSF. Some of the metabolites show a significant interaction effect between treatment and MAOA genotype. An impulsivity-related side effect after fluoxetine treatment was also observed and correlated with metabolite levels. Our study illustrates that the metabolic deviations detected in plasma and CSF may serve as biomarkers for fluoxetine response in juvenile monkeys.

Pathways/Receptors/Biochemistry

Agonist and mechanically induced receptor activations evoke distinct active receptor conformations

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The FRET technique is widely used to monitor interactions between two proteins. Here, we inserted two fluorochromes in one protein to detect dynamic intramolecular conformational changes of G-protein coupled receptors. To analyze differences between agonist-induced and mechanically induced active receptor conformations, we analyzed the Gq/11 protein coupled histamine H1 receptor, which showed the highest mechanosensitivity in previous studies. This receptor regulates many physiological processes like ileum contraction, modulation of circadian cycle, systemic vasodilatation, allergy-induced itching and bronchoconstriction. The receptor was C-terminally linked to cerulean, a stable cyan fluorescent protein, and a small tetracysteine-binding motif was inserted at different positions of the third intracellular loop, which allows binding of the small fluorescent arsenical hairpin binder FlAsH, a yellow fluorophore. Agonist stimulations with histamine and mechanical stimulations with hypotonic bath solutions were performed using a focal pressurized perfusion system. Interestingly, mechanical stimulation resulted in a significantly greater decrease of the FRET signal than agonist stimulation with higher amplitudes and faster kinetics. Furthermore, the amplitude of mechanically induced FRET signals showed a concentration dependency since hypotonic solution with 150 mOsmol evoked higher FRET amplitudes than 250 mOsmol bath solution. Altogether, our results indicate that agonist stimulation induces a different active receptor conformation than mechanical stimulation.

Pathways/Receptors/Biochemistry

Investigating the chemistry behind red wine astringency - What is the role of protein/polyphenol interactions?

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Consuming beverages like tea or wine, nuts and certain fruits (especially when unripe), one can feel a puckering and dry sensation in the oral cavity. Polyphenols have been identified as the key food constituents causing this phenomenon of astringency. Mediated by hydrophobic forces and hydrogen bonding, polyphenols rapidly bind to salivary proline-rich proteins (PRPs) and histatins forming soluble aggregates which can grow to colloidal size. As a consequence of this complex formation, friction of oral surfaces is believed to increase and salivary lubrication is lost. From an evolutionary point of view, the precipitation mechanism is proposed to function as a defence strategy against harmful dietary phenolics, which might otherwise reduce the nutritional value of foodstuffs by inhibiting iron absorption and digestive enzymes. The concomitant unpleasant mouthfeel can be considered as an alarm system to prevent the consumer from further food intake. To investigate the mechanisms of astringency on a molecular level we performed psychophysical experiments in combination with high performance instrumental analytics of saliva-astringent mixtures. Results of the experimental study will be discussed.

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Pathways/Receptors/Biochemistry

Bayesian analysis of a series of FRAP experiments with mixed-effect priors

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FRAP (Fluorescence Recovery after Photobleaching) is a frequently used imaging technique, with which the binding behaviour of molecules in various cellular compartments, in our case in cell nuclei, can be investigated in vivo. The molecules of interest are therefore fluorescently tagged, a part of the cell nucleus of the cell of interest is bleached, and the recovery of the bleached part of the nucleus is observed by taking images of the nucleus in predefined time intervals. However, there is still a lack of methodology for the quantitative analysis of FRAP data, especially for the joint analysis of multiple images resulting from a series of experiments. We propose a Bayesian approach allowing for mixed-effect priors on the parameters of a nonlinear regression model in order to analyse the recovery curves of several cell nuclei altogether. By using a somatic mouse cell line expressing GFP-tagged DNA methyltransferase, we show that with the proposed method, the data of a series of FRAP experiments on multiple cell nuclei can be analysed simultaneously by a single model. We get joint parameter estimates for all recovery curves as well as a measure for the variation of the parameters which is due to the biological variation between cell nuclei.

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Pathways/Receptors/Biochemistry

Phospholipase C-ε (PLCε) induced TRPC6 activation: A common but redundant mechanism in podocytes

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In all eukaryotic cells, hormonal activation of phospholipase C(PLC)-coupled membrane receptors by hormones leads to increased intracellular Ca^{2+}concentration. PLCs catalyze the hydrolysis of phosphatidylinositol 4,5-bisphosphate to generate inositol 1,4,5-trisphosphate (IP3) and diacylglycerol (DAG) which opens DAG sensitive classical transient receptor potential channels e.g. TRPC6, initiating Ca^{2+} influx from extracellular space. While TRPC6 activation by PLCβ- and PLCγ-isozymes was extensively studied, the role of PLCε remains elusive. Most interestingly, for both proteins, mutations in patients with focal segmental glomerulosclerosis (FSGS) were identified, indicating that DAG production by PLCε might induce TRPC6 activation in podocytes in vivo. TRPC6 co-immunoprecipitated with PLCε when overexpressed in HEK293 cells as well as in freshly isolated podocytes. Receptor-operated TRPC currents in HEK293 cells stably expressing TRPC6 were reduced by a specific PLCε siRNA and by a PLCε loss of function mutant isolated from an FSGS patient. PLCε induced TRPC6 activation was also identified in murine embryonic fibroblasts (MEFs), with deleted $G_{\alpha q/11}$ genes. Further analysis revealed that a $G_{\alpha 12/13}$ RhoGEF activation induces Rho mediated PLCε stimulation. However, TRPC6-/- podocytes, but not PLCε-/- podocytes, show decreased cation influx and increased actin polymerization and proliferation rates, suggesting a redundant role of PLCε-mediated TRPC6 activation in MEF and podocytes.

Pathways/Receptors/Biochemistry

Characterization of $G_{i/o}$-coupled receptors using a cAMP-sensor based FRET and a Kir channel based electrophysiological approach

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To functionally characterize different GPCR-subtypes ($G_{i/o}$, G_s), we performed a fluorescent approach with a cAMP-sensitive reporter coupled to a FRET-pair, the ECFP/EYFP-flanked cAMP sensor Epac. Commonly used to measure cAMP increases upon GsPCR activation, we probed whether this sensor might also serve to monitor $G_{i/o}$-protein activation. $G_{i/o}$-protein coupled adrenergic α2A receptor and the cAMP sensors EYFP-Epac1-ECFP or EYFP-Epac2-ECFP were transiently over-expressed in HEK293 cells. After incubation with forskolin, agonist stimulation with noradrenalin caused an increase in FRET signals, corresponding to reduced cAMP levels. Additionally, we replaced ECFP with mTurquoise2, for higher quantum efficiency, photostability and strictly single-exponential fluorescence decay and EYFP with mVenus to obtain less sensitivity to pH and Cl^- changes. Measurements with HEK293 cells expressing G_s-protein coupled adrenergic β2 receptor with mVenus-Epac1/2-mTurquoise indicate that the new sensors are functional. We further performed whole-cell measurements with HEK293 cells over-expressing Gi/o-protein coupled 5-HT1B- or M2mACh receptors and Kir3.1/Kir3.2 or Kir3.1/Kir3.4 channel complexes that are activated by βγ dimers of $G_{i/o}$-proteins following receptor activation. Agonist stimulations with serotonin or carbachol caused current increases. Altogether, our results demonstrate that a FRET-based approach using cAMP sensor Epac is as suitable to determine $G_{i/o}$-protein activation as an electrophysiological approach monitoring Kir channel activation.

Pathways/Receptors/Biochemistry

Modified messenger RNA and its application in bone tissue engineering

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The aim of gene therapy is to alter protein expression within a cell to gain therapeutic or preventive effect. Very recently an alternative platform technology, called transcript therapy, has been developed. Here, messenger RNA (mRNA) is delivered instead of its counterpart DNA into the target cells. mRNA transfection is a useful approach to target slowly dividing cells with minimal risk of transgene-mediated mutagenesis. In the last decade, it was shown that exogenous mRNA can be modified to increase its stability and enhance translational efficiency. One strategy is to generate stabilized non-immunogenic messenger RNAs, including chemically modified nucleotides within the mRNA. Another strategy is the insertion of untranslated regions (UTRs) into the mRNA sequence to regulate mRNA's stability and its translation. The aim of this project is to investigate a set of UTR sequences as "stability" and/or "translation enhancers". To achieve this, various cellular UTRs were selected based on mRNA stability data and cloned in five different combinations along the reporter gene. Initial screening results, in different cell systems, demonstrate increased and longer expression with some of the tested UTRs compared to controls. The best working combination of UTRs is currently being incorporated into a vector coding for hBmp7 and the resulting mRNAs would then be transfected in mesenchymal stem cells to induce osteogenic differentiation.

65 Pathways/Receptors/Biochemistry

Mesenchymal cells regulate growth of intestinal crypts by a Wnt independent mechanism in 3D culture system

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Intestinal stem cells (ISC) reside at the bottom of each crypt and give rise to all lineages of the intestinal epithelium. Crypts are surrounded by pericryptal fibroblasts, which are believed to create a stem cell niche. The mechanisms by which the local environment regulates ISC proliferation and differentiation are unknown so far. In order to investigate this, we established several 3D culture systems which involve intestinal crypts and mesenchymal cells. The effects of four different murine and human types of mesenchymal cells were analysed. Co-culture studies revealed that these different types of mesenchymal cells induce a sphere-like phenotype in intestinal organoids. Analysis of mRNA levels by RT-PCR showed that mesenchymal cells express ligands for Wnt, Notch, Hedgehog and BMP pathways. Organoids co-cultured with mesenchymal cells exhibited increased proliferation and reduced differentiation as shown by Ki-67 and PAS staining. Similar effects were observed when fibroblast conditioned medium was used. Intestinal organoids derived from tumor tissue of the Apc +/1638N mouse phenotypically resembled crypts co-cultured with mesenchymal cells. However, Wnt inhibition studies revealed that mesenchymal cells regulate growth of intestinal organoids by a Wnt independent mechanism. Mass spectrometry analysis of the supernatant from co-culture uncovered the activation of ECM-receptor and focal adhesion pathways.

66 Pathways/Receptors/Biochemistry

The role of Cab45 in secretory cargo sorting at the Trans Golgi Network

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The Trans Golgi Network (TGN) is the central sorting organelle for newly synthesized proteins in the cell. Incoming cargo is selected and transported to its final cellular destination. Due to the involvement of numerous different cargos and pleomorphic carriers, this process is highly sophisticated and as a consequence dys-regulation or loss-of-function leads to severe disorders like diabetes, cancer and neurological diseases. While the sorting of lysosomal hydrolases at the TGN via a mannose-6-phosphate receptor and clathrin-coated vesicles to the lysosome is well characterised, the relevant mechanism for secreted proteins at the TGN remains largely unknown.

We have recently reported the requirement of Ca^{2+} in this process. A defect in Ca^{2+} homeostasis of the TGN results in missorting of secretory cargo and the secretion of a Golgi-resident, soluble protein called Cab45. But what is the fate of the Ca^{2+} in the lumen of the TGN and how does Ca^{2+} help in the sorting process? We found that Cab45 plays a crucial role in Ca^{2+} homeostasis of Golgi membranes and in the sorting of secretory cargo. However, it is not known how Cab45 is retained within the Golgi, how it sorts secretory cargo and what is the in vivo relevance of the protein.

Pathways/Receptors/Biochemistry

Investigation of self-renewal and senescence of tenomodulin-deficient tendon stem/progenitor cells

67

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Proper regulation of self-renewal and senescence is important for the maintenance of tissue-resident stem cells. In this study we investigated whether deletion of tenomodulin (Tnmd), a marker gene for tendon and ligament lineage, will result in altered self-renewal and senescence properties of tendon stem/progenitor cells (TSPC). To generate Tnmd-ablated TSPC, we used the available Tnmd knockout mouse strain. Self-renewal was studied by long-term calculation of population doubling time, clonogenicity assay and PCR analysis of Cyclin D1. Cellular senescence was investigated by beta galactosidase staining and quantification, and expression analyses of the cell cycle inhibitor genes p16, p21 and p53. The obtained results clearly demonstrated i) a significantly reduced proliferative ability of Tnmd knockout TSPC and ii) an earlier switch of the senescence program, which was accompanied by increased levels of p16 and p53. Taken together, we concluded that loss of Tnmd negatively regulates TSPC self-renewal.

Pathways/Receptors/Biochemistry

Transcriptome surveillance by selective termination of noncoding RNA synthesis

68

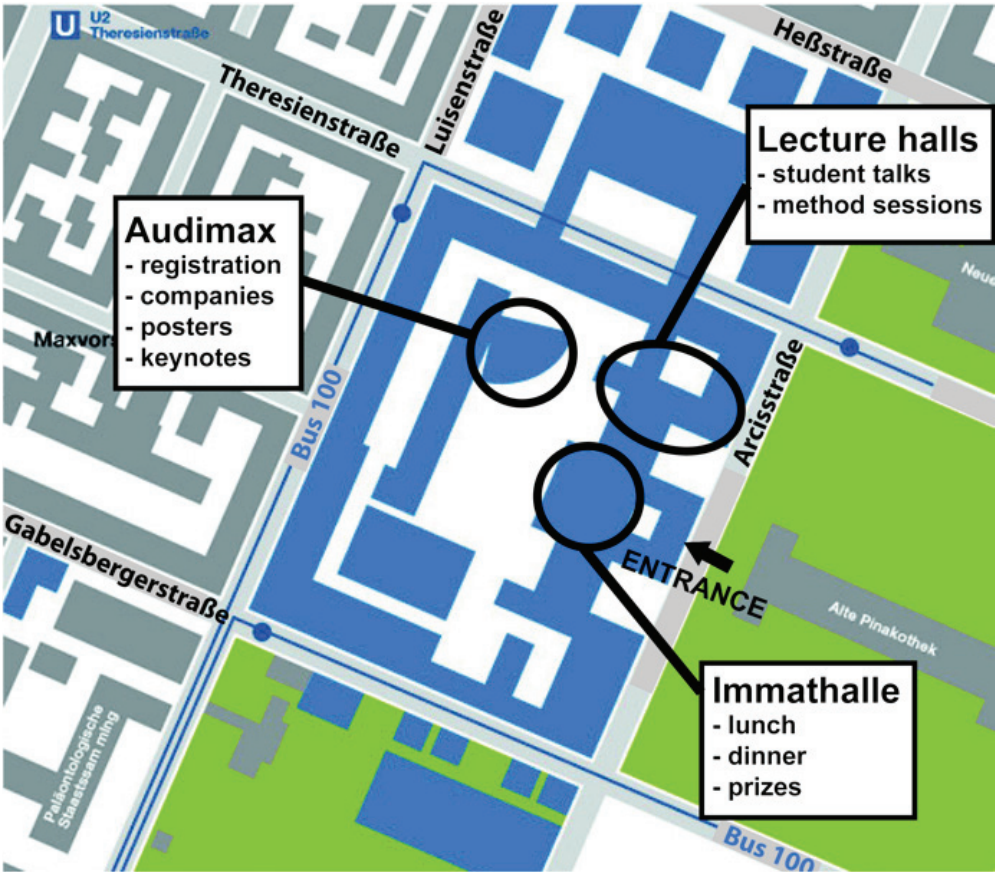
Björn Schwalb[1], Daniel Schulz, Anja Kiesel, Carlo Baejen, Phillipp Torkler, Julien Gagneur, Johannes Soeding, Patrick Cramer

[1]Gene Center Munich, Ludwig Maximilians Universität München

Pervasive transcription of eukaryotic genomes stems to a large extent from bidirectional promoters that synthesize mRNA and divergent noncoding RNA (ncRNA). Here, we show that ncRNA transcription in the yeast S. cerevisiae is globally restricted by early termination that relies on the essential RNA-binding factor Nrd1. Depletion of Nrd1 from the nucleus results in 1,526 Nrd1-unterminated transcripts (NUTs) that originate from nucleosome-depleted regions (NDRs) and can deregulate mRNA synthesis by antisense repression and transcription interference. Transcriptome-wide Nrd1-binding maps reveal divergent NUTs at most promoters and antisense NUTs in most 3' regions of genes. Nrd1 and its partner Nab3 preferentially bind RNA motifs that are depleted in mRNAs and enriched in ncRNAs and some mRNAs whose synthesis is controlled by transcription attenuation. These results define a global mechanism for transcriptome surveillance that selectively terminates ncRNA synthesis to provide promoter directionality and to suppress antisense transcription.

Map

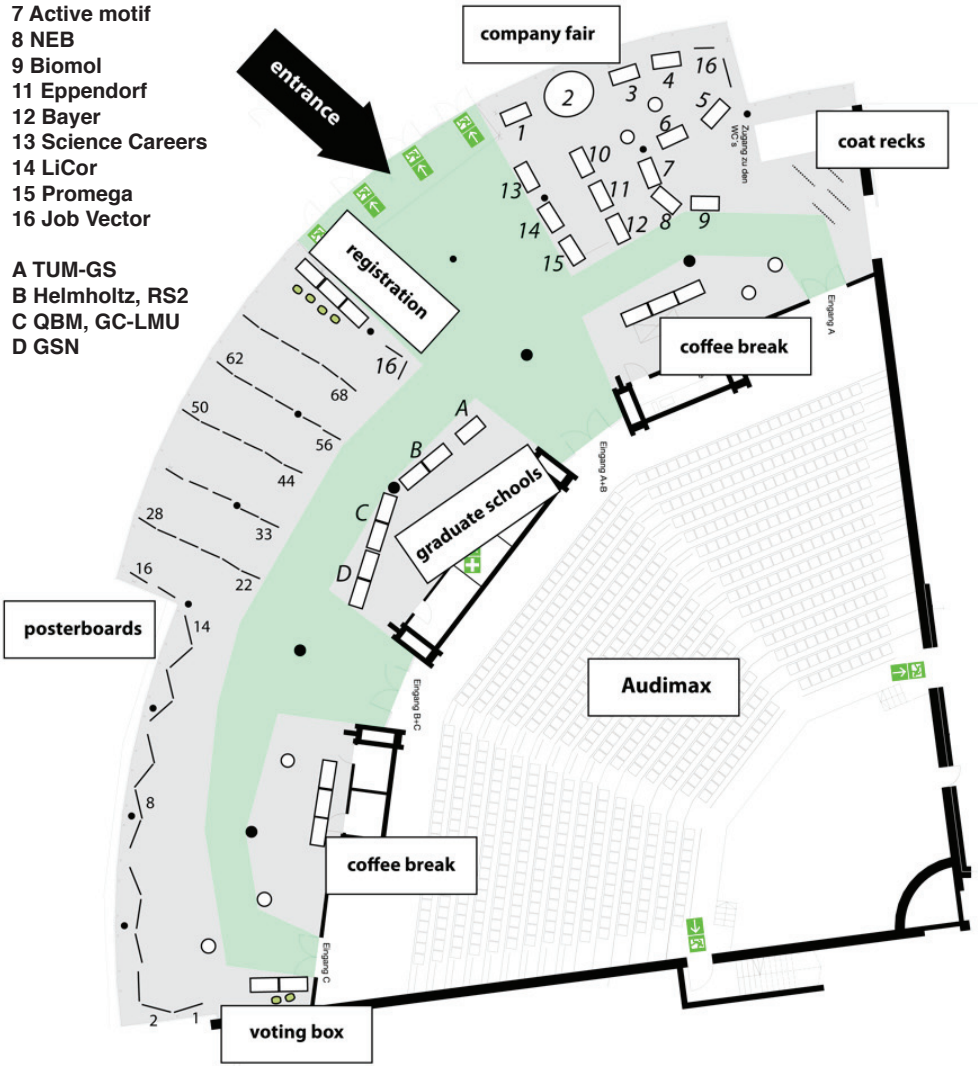
TUM downtown campus



Audimax

1 Amgen
2 Academics
3 Perkin Elmers
4 Merck
5 Adam Ruben (selling books) / mixed
6 Chroma
7 Active motif
8 NEB
9 Biomol
11 Eppendorf
12 Bayer
13 Science Careers
14 LiCor
15 Promega
16 Job Vector

A TUM-GS
B Helmholtz, RS2
C QBM, GC-LMU
D GSN



MENU

Lunch

Two sorts of Tortellini and Gnocchi with several sauces
and mediterranean vegetables
Zweierlei gefüllte Tortellini und Gnocchi mit verschiedenen Soßen und mediterranem Gemüse

Salad buffet
Salatbuffet

Weihenstephaner yoghurt
Weihenstephaner Joghurt

Dinner

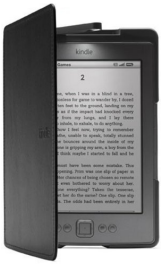
Freshly baked “Leberkäs” with potato salad
Ofenfrischer Leberkäs mit Kartoffelsalat

“Käsespätzle” with fried onions and salad
Käsespätzle mit Röstzwiebeln und Salat

Best Talk & Poster

Awards and Prizes

Be sure to vote for the best scientific contributions!
The best talks and posters will be awarded with:

	Talk	Poster
First prize **Sponsored by academics.de**	 **iPad mini**	 **Samsung GALAXY SIII**
Second prize **Sponsored by Olympus**	 **Olympus PEN Lite E-PL5**	 **Olympus Stylus XZ-2**
Third prize	 **Kindle Fire**	 **Kindle with case**

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Simon Golin,
„Project Management - interact Symposium“
May 7th - 8th, 2014

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http://www.munich-interact.org
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We are looking forward to welcome you in the next <interact> organizing team!

Acknowledgements

The <interact> 2014 organizing team would like to thank all our supporters. This symposium has only been possible with all your help!

Our special thanks go to:

- our four keynote speakers Prof. Dr. Hans Peter Peters, Dr. Adam Ruben, Prof. Dr. Thilo Stehle and Prof. Dr. Paul Frankland
- our four method session speakers Dr. Tobias Straub, Thorsten Abs, Prof. Dr. Heinrich Leonhardt and Prof. Dr. Don Lamb
- all members of our advisory board
- our generous donors
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- Christian Ude (Mayor of Munich)
- Prof. Dr. Wolfgang A. Herrmann (President of the TUM)
- Prof. Dr. Wolfgang M. Heckl (General Director of Deutsches Museum)
- all the participating institutes
- and YOU for participating! Special thanks to all our talk and poster presenters!

Notes

Presentation of Industry and Academia

academics is the leading job listing site for scientists and researchers in academia and industry. Whether you are looking for a PhD programme, a postdoc placement or a professorship - or perhaps considering a career in industry or research companies in Germany - academics has all the answers.
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Amgen is committed to unlocking the potential of biology for patients suffering from serious illnesses by discovering, developing, manufacturing and delivering innovative human therapeutics. This approach begins by using tools like advanced human genetics to unravel the complexities of disease and understand the fundamentals of human biology.

Amgen focuses on areas of high unmet medical need and leverages its biologics manufacturing expertise to strive for solutions that improve health outcomes and dramatically improve people's lives. A biotechnology pioneer since 1980, Amgen has grown to be the world's largest independent biotechnology

company, has reached millions of patients around the world and is developing a pipeline of medicines with breakaway potential.

Amgen strives to serve patients by transforming the promise of science and biotechnology into therapies that have the power to restore health or even save lives. In everything we do, we aim to fulfill our mission to serve patients.
Our Mission: To Serve Patients

Be Science-Based
Our success depends on superior scientific innovation, integrity and continuous improvement in all aspects of our business through the application of the scientific method. We see the scientific method as a multi-step process that includes designing the right experiment, collecting and analyzing data and rational decision making. It is not subjective or emotional, but rather a logical, open and rational process. Applying the scientific method in all parts of the organization is expected and highly valued.

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BC develops future leaders within Bayer as a launch pad for an exciting global executive career that provides abundant opportunities for team members to add value across different industries, functions, and countries.

Bio^{M} is a not-for-profit organisation with the mission to support the Bavarian biotechnology sector. Bio^{M} offers a wide variety of services, partly at no charge, to all regional biotech and life science companies.

This includes:
- Consulting for start-ups
- Business development support
- Funding opportunities
- PR and trade fairs
- Sector-specific job site
- Events and vocational training

Through networking, location marketing, consultancy and other services, we help to bring the Munich m^{4} Biotech Cluster further to the international forefront and make it visible on the global stage. Since winning the Leading-Edge Cluster Competition in 2010, Bio^{M} has been entrusted with managing the leading-edge cluster m^{4} - Personalized Medicine and Targeted Therapies

Biomax was founded in 1997 with the mission to develop software solutions based on the actual needs of life science organizations.
In pursuit of this goal, Biomax developed a systematic client consultation process that includes evaluation, feedback and optimization. As a result, Biomax has developed software solutions which address both highly specific and general research needs along the discovery-process pipeline. These software tools and content offer a robust foundation for expanding and building new bioinformatics solutions.
Today, life science organizations need access to relevant knowledge at the right time and in the right context to cut costs and enable efficient prioritization of research and production tasks based on well-defined objectives. To meet this need, Biomax provides computational and knowledge solutions which infer and manage the knowledge indispensable for efficient decision making.

Biomol GmbH is a key player in the life science reagent market. Based in Hamburg, Biomol supplies biochemicals, assay kits, proteins and antibodies for research areas like gene regulation, signal transduction, inflammation and apoptosis.
Biomol presents with pleasure its product highlights at the Interact 2014.

The Center for NanoScience (CeNS) was founded in 1998 at the Ludwig-Maximilians-University (LMU) Munich. The mission of CeNS is to promote and coordinate interdisciplinary research in the field of nanoscience in the Munich area. CeNS is an association of junior and senior scientists from basic research and industry and is conceived as a network, joining people from various institutions (LMU, TU Munich, University of Augsburg and others).

Chroma Technology Corp. is a leading manufacturer of optical filters and related products. Our mission is to provide the best products and solutions for our customers' ever-changing needs. To that end we develop lasting relationships with our customers by providing applications expertise and exceptional customer service. Chroma Technology is 100% employee-owned: each employee has a share of the responsibility for Chroma's success, and each earns a fair share of the rewards. We value the economic and social needs of the individual as well as the needs of the company as a whole. This is how we create the committed, involved and healthy employee body that is key to achieving our mission. Chroma Technology is an active and caring member of the community in which we live as well as the scientific and technical communities that we serve. We give back to these communities by providing jobs, offering financial support to local non-profit service agencies, sponsoring educational opportunities for students of science and technology, and promoting employee ownership.

eBioscience, an Affymetrix company is a lead supplier in flow cytometry and offers besides a large selections of antibodies, also immunoassays and proteins for Life Science research and diagnostic.
Frequently first to market the company develops more than 800 innovative products every year including antibodies, fluorochromes and further reagents for immunology, oncology, cell biology, stem cell biology and diagnostics.
In addition a broad spectrum of reagents for the analysis of cytokines, growth factors and other soluble proteins as well as bead-based multiplex

immunoassays for the flow cytometer, ready-to-use ELISA kits, matched antibody pairs, proteins and standards enables eBioscience, an Affymetrix company to provide complete solutions for biological systems analysis.

Eppendorf is a leading life science company that develops and sells instruments, consumables and services for liquid, sample and cell handling in laboratories worldwide. Its product range includes pipettes and automated pipetting systems, dispensers, centrifuges, mixers, spectrometers and DNA amplification equipment as well as ultra-low temperature freezers, fermenters, bioreactors, CO_2 incubators, shakers and cell manipulation systems. Associated consumables like pipette tips, test tubes, microliter plates and disposable bioreactors complement the instruments for highest quality workflow solutions.

Eppendorf products are most broadly used in academic and commercial research laboratories, e.g., in companies from the pharmaceutical and biotechnological as well as the chemical and food industries. They are also aimed at clinical and environmental analysis laboratories, forensics and at industrial laboratories performing process analysis, production and quality assurance.

Eppendorf was founded in Hamburg, Germany in 1945 and has about 2,700 employees worldwide. The company has subsidiaries in 25 countries and is represented in all other markets by distributors.

The **GraduateCenter**^{LMU} , the central unit for doctoral studies at LMU Munich, offers comprehensive services for doctoral students, coordinators of doctoral programs and professors. Its mission is to strengthen the framework for doctoral studies at LMU Munich and to ensure optimal research training for all junior scientists at the highest level.

The **GSN** is the teaching entity for the Munich Center of Neurosciences - Brain & Mind (MCN), offering an integrated program of study which takes students from their bachelor to a master or doctoral degree.

Under the umbrella of the Munich Center for Neurosciences - Brain & Mind, the GSN looks to link research groups from behavioral & cognitive neuroscience, biomedical neuroscience, cellular & systems neuroscience, molecular & developmental neuroscience, neurophilosophy and theoretical neuroscience & technical applications. Through creating an interdisciplinary network of research the GSN provides a stimulating environment for students and faculty to produce novel formulations of current concepts and theories.

The newly established **Graduate School of Quantitative Biosciences Munich (QBM)** is funded by the German Excellence Initiative and seeks to prepare young life scientists for the emerging era of quantitative, system-oriented bioscience. It provides an innovative, international doctoral training program that bridges the divide between traditionally separate disciplines, from biochemistry and medicine to bioinformatics, experimental and theoretical biophysics, and applied mathematics.

Key elements of the program are an interdisciplinary research project jointly supervised by two PIs from different fields, and an educational curriculum centered around an intensive core course that integrates a wide range of approaches to biological problems. A multi-facetted mentoring and professional skills program support the students' growth as independent scientists. www.qbm.lmu.de

HELMHOLTZ GEMEINSCHAFT

HELENA
Helmholtz Graduate School Environmental Health

Helmholtz Zentrum München, the German Research Center for Environmental Health, pursues the goal of developing personalized medicine, i.e. a customized approach to the diagnosis, treatment and prevention of widespread diseases such as diabetes mellitus and lung diseases. To that end, it investigates the interaction of genetics, environmental factors and lifestyle. The head office of the center is located in Neuherberg in the north of Munich. Helmholtz Zentrum München has a staff of approximately 2200 people and is a member of the Helmholtz Association, a community of scientific-technical and medical-biological research centers with some 34,000 staff members. www.helmholtz-muenchen.de.

Since 2010 Helmholtz Zentrum München ensures with the **Helmholtz Graduate School Environmental Health (HELENA)** an optimal education for young research talents. HELENA is a joint initiative for the promotion of graduate students of the Helmholtz Zentrum München, the Ludwig-Maximilians-Universität München (LMU) and the Technische Universität München (TUM). www.helmholtz-helena.de.

The **Postdoctoral Fellowship Program (PFP)** continues this successful effort for scientific educational training being an attractive research funding for postdocs. It is a newly established program for excellent early-career scientists (up to 3 years after graduation) at Helmholtz Zentrum München. Talented postdocs who are accepted into the three-year program receive 3-year fully funded postdoctoral positions and can focus on a research project in an institute of the Center. http://www.helmholtz-muenchen.de/fellows

IMPRS
From Biology to Medicine

The **IMPRS-LS PhD Program** is jointly organized by Munich-based Max Planck Institutes and Universities and is committed to first class training and education of life science graduate students. The program covers the areas of biochemistry, cell biology, molecular medicine, neurobiology and structural biology and takes an integrative and interdisciplinary approach to connect these disciplines and the participating players. Networking, communication and scientific interactions are promoted as an integral part of graduate training. The interdisciplinary setting of IMPRS-LS exposes students to a wide range of different topics and technologies thereby promoting the ability of cross-frontier thinking. While such broadly-based perspectives characterize the framework of the program, the research of an individual doctoral student is tightly anchored within the scope and focus of the chosen research group, providing ample room for specialization and in depth training.

Thesis research at IMPRS-LS is embedded into a structured curriculum offering a variety of training opportunities including lecture series, seminars, advanced courses, method and career development workshops and participation in international conferences.

Doctoral degrees are typically awarded by one of the two Munich partner universities, the Ludwig-Maximilians-Universität (LMU) München or the Technische Universität München (TUM).

More than 250 doctoral students from all over the world are currently working at the Max Planck Institutes of Biochemistry and Neurobiology and, together with numerous doctoral students from close by university laboratories, create a lively and dynamic international atmosphere at the research campus.

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The three-year PhD program in **Medical Life Science and Technology at TUM** provides high-level scientific training for students with a background in medicine as well as for those with a background in natural and life sciences or engineering. It consists primarily of an experimental scientific project that is performed in the laboratory of a faculty member. The practical work is complemented by interdisciplinary lectures, seminars and practical courses. The projects cover a diverse range of topics within and continuously expanding the boundaries of the program's main research areas: cardiovascular, imaging, immunology and infection, neuroscience, oncology and molecular medicine. The goal of the program is to give our students a thorough scientific training and the tools to become successful and independent researchers.
For more details, please visit our website: http://www.phd.med.tum.de/.

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The Life Science division Merck Millipore offers solutions that enable scientists to conduct life science research easily, efficiently and economically. With a range of more than 60,000 products, Merck Millipore is one of the top three suppliers of tools to the life science industry.
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A supplier-of-choice for scientists across the globe, NEB offers the largest selection of recombinant and native enzymes for genomic research. While restriction enzymes remain part of our core product portfolio, our ever-expanding catalog also includes products related to PCR, gene expression, cellular analysis, RNA analysis and epigenetics. Currently, NEB supplies over 276 restriction enzymes. These recombinant enzymes are available at a lower cost/unit and offer improved purity and lot-to-lot consistency. A recognized leader in the field of restriction enzymes, NEB has earned a world-wide reputation for setting the highest standards for quality and value.

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