Dämmernd liegt der Sommerabend...

Über Wald und grünen Wiesen;
Goldner Mond, im blauen Himmel,
Strahlt herunter, duftig labend.

An dem Bache zirpt die Grille,
Und es regt sich in dem Wasser,
Und der Wandrer hört ein Plätschern
Und ein Atmen in der Stille.

Dorten an dem Bach alleine,
Badet sich die schöne Elfe;
Arm und Nacken, weiß und lieblich,
Schimmern in dem Mondenscheine.

Heinrich Heine (1797-1856)
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Redaktion
Heidi Hoyermann (Leitung)
Verena Jäggin

Übersetzungen
Paula Cullen
Brigitte Schneider

Layout
Patrick Rosemary,
Morf Bimo Print AG, Binningen

Publikationen/Publications
Verena Jäggin (Layout)

Mitarbeitende/Colleagues
Verena Jäggin (Layout)

Fotos
Manuela Bernasconi (privat)
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Christoph Moroni (privat)
Patrick Rosemary (Rückseite)

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Anschrift
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Departement Biomedizin
Hebelstrasse 20
4031 Basel
dbfacts@unibas.ch
Liebe Leserinnen und Leser


Christoph Moroni hat mit seiner Emeritierung die Leitung des Instituts für Mikrobiologie abgegeben, setzt aber seine Forschungstätigkeit als Gastprofessor am Biozentrum fort. Simona Rossi Girard hat als SNF-Förderprofessorin ihre Tätigkeit aufgenommen, ihre Forschungsgruppe trägt den Namen «Immunoregulation». Zusätzlich wurden vom SNF eine neue Förderprofessur an Alfred Zippelius (Onkologie) und je ein SCORE Beitrag an Beat Kaufmann (Kardiologie) und David Semela (Hepatology) zugesprochen. Allen einen guten Start am DBM und viel Erfolg!

In der neuesten Ausgabe gibt uns Bernhard Bettler einen Einblick in die Aktivitäten seines Labors «Molecular Neurobiology Synaptic Plasticity» und Bettina Burger und Erwin Kump nehmen uns mit auf die Reise durch die Forschung der Gruppe «Dermatology». Ein ganz anderes Forschungsthema beleuchtet Daniel Haag-Wackernagel in seinem Beitrag über Tauben, Menschen und Wildschweine. Eine Auswahl der neuesten Publikationen aus dem DBM finden Sie ab Seite 18.

Das und vieles mehr finden Sie in der nun vorliegenden Ausgabe. Lassen Sie sich überraschen.

Viel Spass bei der Lektüre.
Radek Skoda

Dear Readers

Many of you have asked yourselves, and others have asked us directly: “When will the next issue of DBM Facts appear?” The reason behind the delay in the publication of the first issue for 2009 was the illness of Thomas Stebler, our long term designer and creative head, who, to our regret, had to resign suddenly and unexpectedly. Following this a promising collaboration with a new partner began to show delays due to their having severe back problems and this arrangement also had to be finally ended. On our third attempt we found Patrick Rosemary, an accomplished and competent designer, whom we with trust with DBM Facts due to his years of experience working in the printers where DBM facts is produced.

There is a lot to report since the last issue of DBM Facts. In February 2009 the DBM was visited and evaluated, for the first time, by the new Advisory Board. During this visit a research day was held, on which 12 research groups made presentations. A short portrait of the 8 international renowned scientists on our Advisory Board can be found on page 17. The report of the Advisory Board will be shared with the rectory, deanship and the directors of the university clinics involved and forms an important basis for future strategic decisions.

With his promotion to professor emeritus, Christoph Moroni has resigned his position as head of the Institute for Microbiology, but is continuing his research work as a guest professor at the Biozentrum. Simona Rossi Girard has taken up her position as SNF-professor, her research group carries the name “Immunoregulation”. Additionally, the SNF has awarded a new professorship to Alfred Zippelius (Oncology) and SCORE grants to Beat Kaufmann (Cardiology) and David Semela (Hepatology). We wish them all a good start at the DBM and much success.

In this latest issue Bernhard Bettler gives us an insight into the activities of his Laboratory “Molecular Neurobiology Synaptic Plasticity” and Bettina Burger and Erwin Kump take us on a journey through the research of their group “Dermatology”. A completely different research topic is highlighted by Daniel Haag-Wackernagel in his article on pigeons, people and boar. A selection of the latest publications from the DBM can be found on page 18.

All that and more can be found in this current issue. Let yourself be surprised.
Getting brains back in tune

Somehow the unstable stuff of which we are composed has learned the trick of maintaining stability.

Walter Cannon, The Wisdom of the Body
The “Synaptic plasticity” group at the Institute of Physiology is primarily interested in the mechanisms that control neuronal excitability, and to exploit these mechanisms for the treatment of neurological and psychiatric diseases. Our work largely focuses on G-protein coupled receptors (GPCRs) and their role in synaptic transmission and synaptic plasticity. Here I describe, from a historical perspective, how our efforts to make the GABA\(_{\beta}\) receptors amenable to drug discovery not only yielded novel synthetic GABA\(_{\beta}\) compounds, but also shaped our view of how GPCRs operate in general.

**The balance of excitation and inhibition**

A fine-tuned balance of excitation and inhibition in neuronal circuits is essential for nearly all brain functions, including representation of sensory information, cognitive processes such as decision making, sleep and motor control. Because a tightly controlled balance between excitation and inhibition is critical for brain functions, homeostatic plasticity mechanisms are in place to prevent excess excitation or inhibition. Loosely defined, homeostatic plasticity acts to stabilize the activity of a neuron or neural circuit in the face of perturbations, such as changes in cell size or in synapse number or strength. Neurons can compensate for perturbations by, for example, modulating the activity of ion channels and neurotransmitter receptors. Homeostatic plasticity assures that most of us make it through life without bringing our brains in for a tune-up, unlike our hardwired cars that once in a while need a trip to the shop. Genetic and epigenetic factors can cause homeostatic plasticity mechanisms to become maladaptive or dysfunctional, resulting in an imbalance of excitation and inhibition. This is believed to underlie many neurological and psychiatric diseases, including epilepsy, Parkinson’s disease, schizophrenia, anxiety, depression posttraumatic stress disorder and addiction. The Mendelian Inheritance in Man (OMIM) database currently lists ~500 genes associated with mental retardation and neuropsychiatric disorders, suggesting that a causal treatment for these disorders will be difficult. Currently available medication for neurological and psychiatric diseases therefore primarily aims at restoring neuronal homeostasis by tipping the balance of excitation and inhibition, by either manipulating the production and removal of neurotransmitters or by activating or blocking neurotransmitter receptors. A prime target for mental health disorders are the neurotransmitter systems that operate in the neural circuits involved in mood regulation (prefrontal cortex and limbic structures), fear and anxiety (amygdala-based circuits) and cognitive control of behaviour (frontal-striatal-thalamic circuits).

**Glutamate and GABA receptors as therapeutic targets**

The most abundant excitatory and inhibitory neurotransmitters in the brain are glutamate and GABA, respectively. The receptors for GABA and glutamate are therefore of importance for regulating the patterns of activity in almost all neural circuits. It is therefore not surprising that several drugs acting at these receptors have been introduced to the clinic. These drugs primarily target GABA\(_{A}\) (e.g. benzodiazepines for anxiety, insomnia, agitation, seizures, muscle spasms, alcohol withdrawal) and NMDA receptors (e.g. memantine for Alzheimer’s disease), which are neurotransmitter-gated ion channels. However, glutamate and GABA also signal via G-protein coupled metabotropic glutamate receptors (mGluRs) and GABA\(_{B}\) receptors. These GPCRs modulate rather than mediate synaptic transmission and profoundly affect synaptic plasticity mechanisms. They therefore represent another means for restoring the balance of excitation and inhibition.

The first mGluR was cloned in 1991, and seven other structurally related receptors were identified shortly thereafter. These receptors exhibit unique patterns of distribution in the brain and therefore allow a pharmacological manipulation of selective neuronal populations and neural circuits. The cloning of mGluRs therefore sparked drug discovery efforts in all major pharmaceutical companies and nowadays a whole range of subtype-specific synthetic agonists, antagonists and allosteric modulators are available. Compounds acting at mGluRs are currently being evaluated for the treatment of anxiety, schizophrenia, nicotine/alcohol dependence, cognitive dysfunctions and fragile X mental retardation. In contrast to the mGluRs, GABA\(_{B}\) receptors resisted cloning for a long time. Using an expression cloning approach based on radioligand binding, my lab (at the time at Novartis) eventually succeeded in cloning GABA\(_{B}\) receptors a decade ago. Surprisingly, we
found that GABA<sub>B</sub> receptors need to form heterodimers between GABA<sub>B1</sub> and GABA<sub>B2</sub> subunits to function, which explained why previous expression cloning attempts based on functional assays failed. This finding unsettled the classical view that GPCRs function as monomeric entities and lead to the discovery of a wide array of other homo- and heteromeric GPCRs<sup>1–4</sup>. Most people in the field will probably now agree that GPCRs form either homo- or heterodimers, and possibly higher order oligomers. GPCR heterodimers often differ in their properties from the homodimers and bind distinct ligands and/or initiate novel signaling pathways. Heterodimers therefore represent novel drug targets and offer the opportunity for a more selective therapeutic interference with cellular functions.

Following the cloning of GABA<sub>B</sub> receptors, we showed that the absence of functional GABA<sub>B</sub> receptors in knock-out mice produces overt phenotypes, including epileptiform activity, impaired memory, hyperalgesia, hypothermia, contextual hyperactivity and increased anxiety<sup>5,6</sup>. These phenotypes pointed at possible therapeutic indications for GABA<sub>B</sub> receptors. While still at Novartis, we identified the first positive allosteric modulators acting at GABA<sub>B</sub> receptors. These allosteric
modulators produced pronounced anxiolytic effects, in line with the anxious phenotype of GABA\textsubscript{B} knockout mice. In a separate line of experiments, we used GABA\textsubscript{B} knockout mice to demonstrate that \(\gamma\)-hydroxy butyrate (GHB), a metabolite of GABA and popular drug of abuse, mediates its typical physiological effects via the activation of GABA\textsubscript{B} receptors. This clearly established GHB as a second endogenously produced agonist at GABA\textsubscript{B} receptors.

**GABA\textsubscript{B} receptor heterogeneity**

Before cloning, it was generally assumed that the GABA\textsubscript{B} receptor system would include several pharmacologically distinct receptor subtypes, similar to the mGluRs. It came as a big surprise to many in the field that we only identified two receptor subtypes. Even more puzzling was the finding that the two subtypes, when expressed in vitro, did not exhibit pharmacological or functional differences. Receptor subtypes are based on the subunit isoforms GABA\textsubscript{B1a} and GABA\textsubscript{B1b}, both of which combine with GABA\textsubscript{B2} subunits to form heteromeric GABA\textsubscript{B(1a,2)} and GABA\textsubscript{B(1b,2)} receptors. Because of the lack of selective pharmacological tools it remained unclear whether the two receptors convey identical or separate functions in vivo. Likewise, it remained unclear why native GABA\textsubscript{B} receptors differ in their electrophysiological and pharmacological characteristics whereas the cloned receptors do not. During the past six years my laboratory has started to address these issues.

Because selective pharmacological tools are missing, we dissected the native functions of the GABA\textsubscript{B(1a,2)} and GABA\textsubscript{B(1b,2)} receptors by generating GABA\textsubscript{B1a}\textsuperscript{-/-} and GABA\textsubscript{B1b}\textsuperscript{-/-} mice that selectively express one or the other GABA\textsubscript{B} receptor subtype\textsuperscript{8}. Electrophysiological analysis of these mice demonstrated that GABA\textsubscript{B1a} exclusively assembles presynaptic receptors inhibiting glutamate release, while both GABA\textsubscript{B1a} and GABA\textsubscript{B1b} can form receptors inhibiting GABA release\textsuperscript{6, 8, 9}. We further showed that mainly receptors assembled with GABA\textsubscript{B1b} couple to postsynaptic effector K\textsuperscript{+} channels and Ca\textsuperscript{2+} channels. The genetic absence of GABA\textsubscript{B1a} resulted in a pronounced loss of long-term potentiation (LTP), a synaptic plasticity phenomenon essential for learning and memory formation\textsuperscript{6, 8}. The loss of LTP in GABA\textsubscript{B1a}\textsuperscript{-/-} mice is due to a decrease in “silent synapses”, which are synapses that normally can be recruited during synaptic plasticity processes\textsuperscript{8}. A physiological role of GABA\textsubscript{B1a} receptors is therefore to limit the loss of silent synapses and to ensure that plasticity processes are maintained in the dynamic range. In collaboration with K. Kaufmann and J. Cryan (Novartis), we found that GABA\textsubscript{B1a}\textsuperscript{-/-} and GABA\textsubscript{B1b}\textsuperscript{-/-} mice exhibit differences in several behavioural paradigms, including taste aversion\textsuperscript{10}, fear conditioning\textsuperscript{6} and cognition\textsuperscript{8}. Interestingly, GABA\textsubscript{B1b}\textsuperscript{-/-} mice are unable to extinguish aversive memories for months, which is reminiscent of posttraumatic stress disorder, a condition characterized by intense fear resulting from the exposure to extreme trauma. In collaboration with Novartis, we recently developed an assay system that allows screening for receptor subtype-specific compounds\textsuperscript{11}. Such compounds would allow a more selective therapeutic interference with the GABA\textsubscript{B} receptor system. For example, specific GABAB\textsubscript{1a(2)} receptor antagonists are predicted to be superior to non-specific GABA\textsubscript{B} receptor antagonists that are being tested in a Phase II clinical trial with Alzheimer’s disease patients.

A major focus in my laboratory is to investigate why GABA\textsubscript{B1a(2)} and GABA\textsubscript{B1b(2)} receptors do not exhibit pharmacological or functional differences in transfected non-neuronal cells, while native GABA\textsubscript{B} responses clearly differ in their electrophysiological and pharmacological characteristics. Differences could, for example, relate to direct modifications of the receptor (e.g. phosphorylation), to receptor-associated proteins, to distinct effector systems or a combination of the above. For drug discovery, it is important to clarify whether additional GABA\textsubscript{B} receptor subunits exist or not. To address this issue we started collaborating with B. Fakler (University Freiburg iBr) and used GABA\textsubscript{B}-specific antibodies to affinity purify GABA\textsubscript{B} receptor complexes from mouse brains. GABA\textsubscript{B} receptor-associated proteins were subsequently identified using mass spectrometry. These experiments revealed the interaction of GABA\textsubscript{B} receptors with a family of proteins with unknown function. We are currently analyzing whether these proteins alter functional and pharmacological properties of recombinant GABA\textsubscript{B} receptors. We are also generating knock-out mice to study how the absence of these proteins changes the properties of native GABA\textsubscript{B} receptors.
Devanx (http://devanx.vitamib.com/)

We have recently been awarded research funding for five years under the European Commission’s Framework Programme 7 (FP7) Health initiative. The funding will go towards the study of the molecular basis of anxiety disorders and how perturbations in early life prime the brain for altered emotionality in adulthood. The project, titled DEVANX (Serotonin and GABAB receptors in anxiety: from developmental risk factors to treatment), is undertaken by a pan-European consortium of seven laboratories involving investigators from INSERM (Paris, France), the Universidad Pablo de Olavide (Seville, Spain), the European Molecular Biology Laboratory (Monterotondo, Italy); the Central Institute of Mental Health (Mannheim, Germany) and our group. One of our main tasks will be to generate the genetic tools to inactivate and restore GABA\(_B\) receptor function in defined neural circuits.

**Additional projects**

In collaboration with M. Hoener (Roche) we have studied Trace Amine-Associated Receptor 1 (TAAR1), a member of a novel family of nine GPCRs expressed in monoaminergic systems. TAAR1 not only responds to trace amines, which are endogenous amine compounds present at low levels in the brain, but also to classical biogenic amines and amphetamine-related psychostimulants. Trace amine receptors have therefore been implicated in the etiology of a number of neuropsychiatric disorders. Our electrophysiological analysis of TAAR1 knock-out mice revealed that TAAR1 tonically activates dopaminergic neurons in the ventral tegmental area, a brain region that is part of the pleasure system, or reward circuit, one of the major sources of incentive and behavioural motivation\(^2\).

In collaboration with B. Hemmings (FMI) and A. Merlo (Neurosurgery) we have been awarded a research grant from Oncosuisse to develop molecular strategies for therapeutic interference with brain tumors. Based on the findings that Notch2 is lost in oligodendrogliomas and frequently amplified and overexpressed in glioblastoma and astrocytoma, we generated mouse models to test whether Notch2 can either act as a tumor suppressor gene or as an oncogene. The analysis of Notch2 conditional knock-out mice and Notch2 transgenic mice supports that both the lack and the ectopic expression of Notch2 can increase the number of proliferative cells in the brain. In addition to the transgenic experiments, we currently use cultured neural stem cells to investigate how Notch2 influences cell proliferation.

**References:**

WHICH OUTSIDE REVEALS YOUR INSIDE?

Introduction
The Dermatological Research group (lab 317) was founded in July 2007 on the Department of Biomedicine. We work closely together with the “Klinik und Poliklinik für Dermatologie” and react to the daily request of patients and external physicians at the hospital. Our research is focused on two different dermatological topics: genodermatosis and immunological reactions of the skin. Our genodermatosis project includes different small projects resulting from the daily questions as well as one major project. This focuses on the connections between skin lesions and hereditary colon cancer. The second project aims to shed light on the immunological basis of graft-versus-host disease in the skin.

I. Skin and hereditary colon cancer
The term ‘hereditary colon cancer’ encompasses different types of colon cancer: hereditary nonpolyposis colon cancer (HNPCC) or Lynch syndrome, familial adenomatous polyposis (FAP), Peutz-Jeghers syndrome, juvenile polyposis, and PTEN-hamartoma tumor syndrome. All of them are caused by mutations in different genes and most types are accompanied by skin lesions. There are large differences between skin lesions for the different colon cancer types. In some diseases the special type of colon cancer in connection with skin lesions was even classified as a separate syndrome. As a consequence of molecular classification it was recognized that there are different phenotypes of the same disease. In

From left to right: Peter Häusermann, Erwin Kump, Bettina Burger and Peter Itin.
our project we are interested in one disease called Gardner syndrome, a special type of FAP.

**FAP**

FAP is an autosomal-dominant inherited disease characterised by multiple colorectal adenomas and specific extracolonic features. The typical manifestation is the development of hundreds to thousands of colorectal polyps, which progress from an early stage to advanced stages, finally resulting in an invasive carcinoma. The prevalence of FAP is estimated at 1 in 5000 – 10 000 (accounts for nearly 1% of all colorectal cancers) and more than 90% of families affected by FAP have a mutation in APC. For the patients a periodical screening of the colorectum by sigmoidoscopy is recommended, starting between 10 and 12 years, because of the complete penetrance of the disease.

**Gardner Syndrome (OMIM 175100)**

Gardner syndrome, a variant of FAP, was initially described in 1950 by Gardner and Stephens. Main features on the one hand are gastro-duodenal polyps, which have a nearly 100% progression rate to colorectal cancer if left untreated, and on the other hand there are epidermal cysts (node in the skin), fibroma (benign neoplasm of connective tissue), lipoma (benign neoplasm of adipose tissue), possibly pigment shifting, desmoids (low malign type of fibrosarcoma), osteomas (benign bone tumor), congenital hypertrophy of the retinal pigment epithelium (CHRPE) and dental abnormalities. In 1987 results of linkage analysis suggested that Gardner syndrome and familial colorectal cancer are allelic disorders. In 1991, the APC gene (chromosome 5q21-22) was identified and it was recognized that mutations in the APC gene were the underlying cause of both Gardner syndrome and FAP (1, 2).

**APC (Adenomatous Polyposis of the Colon)**

The APC gene is located on the long arm of chromosome 5 and its product is widely expressed in many tissues. The protein contains numerous functional domains mediating the regulation of β-catenin, protein-protein interactions in cell adhesion, formation of epithelial cell-cell contact, and maintenance of cytoskeletal microtubules. The APC gene has an 8538 bp open reading frame and consists of 15 transcribed exons, encoding a 312 kDa protein consisting of 2843 amino acids which acts as a tumor suppressor. Mutation in one allele is the first step following the classical two-hit model of tumor suppressor inactivation. Inactivation of the wildtype allele by somatic loss results in a loss of control of cell growth and proliferation of the affected cells. More than 300 different germline mutations of APC are described, whereas about 30% are de novo. Most mutations (more than 80%) are nonsense or frameshift mutations, which result from point mutations, insertions or deletions, leading to a premature stop codon and a truncated, functionally inactivated protein.

**Skin lesions**

In Gardner syndrome different skin lesions arise more often than in the normal population, benign as well as malign (see also chapter 'Gardner syndrome'). In the case of benign lesions in particular, the occurrence of extracolonic manifestations will not often be noted by the gastroenterologist treating the FAP patient, so exact prevalences are not known. They are often present many years before colorectal polyps develop and could give us the possibility of identifying mutation carriers before the occurrence of adenomas. Unfortunately, in the majority of cases the cause of these skin lesions is unknown and we are not able to differentiate people with skin lesions due to Gardner syndrome from patients with sporadic skin lesions. Furthermore, connections between benign type of skin lesion like epidermal cysts and mutation site in the APC are generally not investigated and are poorly understood.

**Epidermal cysts** (also known as epidermoid cysts or epithelial inclusion cysts) are dermal or subcutaneous nodules in which a keratin-filled cyst is lined by epithelium. In childhood these cysts are rare, they predominantly occur in young and middle-aged adults, but in Gardner syndrome they are one of the main-features. More than 50% of patients with Gardner develop cutaneous cysts (1) and multiple epidermal cysts in a child should remind the physician to check for the syndrome. Several factors differentiate cutaneous cysts associated with Gardner syndrome from ordinary cysts. Epidermal cysts of Gardner syndrome occur at an earlier stage (around puberty), in irregular distribution (over the
face, scalp, trunk and extremities), and tend to be multiple. These cysts can be seen many years before intestinal polyps develop and usually they are asymptomatic. So far, a correlation of developing epidermal cysts and the site of mutation is unknown, patients develop epidermal cysts regardless of their mutation localization. Therefore they do not necessarily occur in all affected members of a family with the same mutation in APC.

One further main feature of Gardner syndrome is two different types of fibromas, nuchal fibroma and Gardner fibroma. Fibromas are benign soft tissue lesions, consisting of collagen and fibroblasts. They show plaque-like growth structure and infiltration of surrounding tissue. Similar to epidermal cysts in association with FAP they have a predilection for childhood and adolescence. Nuchal fibromas are thick bundles of collagen which radiate into surrounding fat. As the name suggests, they occur mainly on the neck. Gardner fibromas are formless sheets of collagen and have been associated with subsequent development of desmoids (see beneath) at the same site, with or without a history of previous surgery. The most affected sites are the back and paraspinal region (61%), the second most affected sites are the head, neck and extremities (14%). 11% of Gardner fibromas affect the chest and abdomen. In contrast to nuchal fibromas, Gardner fibromas do not show increased number of small nerve bundles.

Further benign lesions, very common in Gardner syndrome are lipomas. Ordinary lipomas are benign neoplasms of adipose tissue and are the most common mesenchymal neoplasm in adults between the ages of 40 and 60 years. In Gardner syndrome they are described as appearing more often and at an early stage but reliable information do not exist.

Desmoid tumors, one of the main features of Gardner syndrome, are rare locally invasive fibromatoses. They are a major cause of morbidity in FAP patients who have undergone prophylactic colonic surgery. Between 10 and 15% of FAP patients probably develop desmoid tumors, correlating with an increased risk of ~850 times compared to the general population. Although they are histologically benign and without metastasis, they have a high recurrence rate after excision, cause obstruction and perforation of surrounding structures, and treatment is often unsuccessful. Usually, FAP-associated desmoids arise in the abdomen or the abdominal wall. In most cases they occur within two years of abdominal surgery or during pregnancy. Females have twice the odds of developing desmoids compared with males. Furthermore, a strong family history of desmoids is a risk factor, as well as the position of the APC germ-line mutation. Development of desmoids is connected to mutations at the 3’ end, in general downstream to codon 1444.

Lesions not directly linked to the skin but occurring in nearly 80% of Gardner patients are osteomas. Osteoma is a benign neoplasm of bone tissue consisting of well differentiated compact or cancellous bone that is characterized by slow continuous growth and is the most common accompanying bone lesion seen in Gardner syndrome. Most common location of osteomas is the mandible, however, osteomas may occur in the skull and long bones. Usually, osteomas manifest earlier than polyposis, so they may be sensitive markers for the disease. Many patients develop osteoma over years without knowing that they have Gardner syndrome.

A very important marker for the early non-invasive diagnosis of affected family members is the CHRPE. CHRPE is a harmless pigment lesion, present at birth in a subgroup of patients with mutations leading to stops between codon 463 and 1444. In families inheriting a mutation at this site an ophthalmological examination at early age and detection of the pigment lesions on the fundus can be the first marker of disease.

Genotype-phenotype correlation
Conspicuous phenotype of FAP usually develops in late childhood or early adulthood and is characterized by the appearance of hundreds or thousands of adenomatous polyps in the colon. The genotype-phenotype correlation in FAP starts to be recognized in such a way that determined phenotypical manifestations e.g. desmoids or CHRPE are related with mutations in specific areas of the APC gene. The first sign of a hereditary colon cancer disease like Gardner syndrome may often be the various extracolonic manifestations. The frequency of extracolonic manifestations is dependent on different factors like genetic and environmental. Some of them reflect the influence of different germline mutations, e.g. patients with mutation between codons 1445 and...
1560 have a high risk for developing desmoid tumors. Nevertheless, extracolonic manifestations also show phenotypic heterogeneity among patients carrying the same APC mutation (even intrafamilial) and did not correlate with phenotypic expression of colorectal polyps. At the time different explanations exist for this: first, that the type of ‘second hit’ might vary between individuals with the same germline APC mutation; and second, that factors other than APC genotype can affect disease expression in FAP. These can be modifier genes, which are not linked to APC, epigenetic factors or environment.

Until now, no clear explanation can be offered for the genotype-phenotype correlations in extracolonic manifestations. Due to the lack of studies, the correlation of benign extracolonic manifestations to the cancer predisposition remains unclear. Modifier genes for the development of extra-colonic manifestations are currently still enigmatic. Knowledge of such genes would essentially contribute to a better presymptomatic treatment of FAP patients and could highly improve the quality of care for these patients. To date it is impossible to make a prognosis for probability of developing skin lesions and to make a prediction for the patient.

**Project**

The aim of our project is to understand the development of skin lesions in some patients with a special kind of hereditary cancer. Such knowledge makes it possible to give a better consultation and make an individual risk evaluation for developing different skin lesions.

To perform our study we have contacted approximately 80 patients carrying genetic predisposition for familial adenomatous polyposis coli (FAP) in collaboration with the Department of Human Genetics Basel. The skin of all individuals is examined and, as far as possible, a biopsy is taken. Many patients show benign skin lesions characteristically for the single cancer syndrome, like epidermal cysts, fibroma, lipoma. First examinations confirm the knowledge that carriers of interfamilial identical mutations exhibit different phenotypes relating to skin lesions. Even in families with the same mutation in the APC gene the occurrence of such skin lesions varies between individuals. The specific skin lesions will be analyzed for a second hit or loss of heterozygosity especially in the APC gene, the gene expression, and chromosomal rearrangements. As a comparison we will analyze the number and type of the same skin lesions in individuals without a disposition to colorectal cancer. Likewise, we will study the skin lesions of healthy controls and elaborate the discrepancy between the mutation carrier and controls. (By the way, we are looking for the control group. If you are interested in please contact us for further information.) Subsequently we would like to clarify why some patients with a predisposition for colorectal cancer are affected by skin lesions and others are unaffected. Identification of associations between the skin lesion and the genomic background additional to the congenital APC mutation will help to assess the skin lesion. This results in an improved therapy and follow-up strategies, and a better consultation for the patients. Furthermore it may be helpful to identify patients with Gardner syndrome by their skin lesions and lead them in early prevention.

**II. Graft versus Host disease of the skin**

Graft-versus-Host disease (GVHD) is a major complication after allogeneic hematopoietic stem cell transplantation (HSCT) and the skin is one of the leading organs involved in that process. Clinically, GVHD presents either as an acute disease within days to weeks after allogeneic HSCT mainly during the inpatient phase, or, alternatively, as a more heterogeneous chronic syndrome that usually occurs months to years after discharge from the hospital.

Acute GVHD typically occurs between day 14 and day 42 after HSCT. Here, the skin can be the only target organ and if not, is often attacked as the first organ before the liver and/or intestinal tract. Acute cutaneous GVHD manifests as a maculopapular rash with a sudden onset that is first found on the upper back and lateral neck, later involves palms, soles, pinnae, and cheeks, and eventually gets generalized.

Chronic cutaneous GVHD often appears with milder and subtle lesions such as dryness of skin (xerosis), follicular prominence or ichthyosis. A subtype of chronic cutaneous GVHD is the sclerotic type cutaneous GVHD. This disease often presents with plaques of dermal sclerosis that resembles morphea and eventually progresses to generalized scleroderma.
Based on data that we collect by immunohistochemical analysis on skin biopsies from GVHD patients, we examine the role of leukocytes and apoptosis in acute and chronic GVHD of the skin. We especially want to depict the currently unknown mechanisms that navigate the course of cutaneous GVHD leading to acute or chronic disease. The sclerotic type of chronic cutaneous GVHD will be brought into sharper focus. Here we assume a dominant role of tissue macrophages in the development of this disease manifestation. Tissue macrophages are well known for their various roles i.e. as professional antigen presenting cells, as strong apoptosis inducers and as modulators of the cytokine environment in the surrounding tissue. In the context of sclerotic type of chronic cutaneous GVHD, macrophages are very interesting candidates involved in tissue remodeling, as on the one hand they can express matrix metalloproteinases such as collagenases, and on the other hand, they can serve as a major source of TGF-beta signaling which induces collagen expression in fibroblasts. These functions, and especially the causal relationships between macrophages and fibroblast-induced fibrosis in sclerotic type of cutaneous GVHD, have not been studied so far.

As a small research group, it is of crucial importance for us to establish ourselves in a well defined research niche. We are convinced that cutaneous GVHD is a field which gives us this opportunity. The molecular mechanisms that direct a sclerotic type of chronic cutaneous GVHD are only poorly understood to date, and the focus of interest on the role of tissue macrophages in this disease is novel.

In a first phase of our projects, we rely on skin biopsies from GVHD patients. Here we benefit a lot from the numerous patient samples that are stored in paraffin blocks. In this phase we want to study the numbers and kinds of leukocytes that infiltrate the skin during the course of a cutaneous GVHD, and also the rates of apoptosis that are induced in the different disease manifestations (see Figure 1).

We also work on a skin infiltration assay in order to study the infiltration rates and differentiation events specifically of peripheral blood monocytes from an allogeneic HSCT donor into the skin of the corresponding recipient (see Figure 2). This ex-vivo experiment shall provide us with information on the migration and differentiation behaviour of the monocytes in the course of a cutaneous GVHD. A prospective study is planned starting...
2009 with sequential skin biopsies, focusing on the role of macrophages to navigate the various chronic forms of cutaneous GVHD.

Being a small research group is our major drawback, however, luckily we are linked to the team of PD Dr. Barbara Biedermann, where we have the great opportunity to exchange expertise and knowledge, and to solve technical problems in a fruitful scientific environment. In order to optimize our research concepts and to most exactly define and update our goals, we are in a tight cooperation with the hematology unit of the UHBS. We kindly appreciate the collaboration with Prof. Gratwohl and his team, the conceptual advices and constructive critics of whom are of priceless importance for the success of our GVHD project.

Outlook
Both projects are in the starting phase of collecting samples and data as well as doing preliminary tests. The numerous opportunities for collaborations and reciprocal help one can find here in the ZLF and the UHBS afford very good starting conditions, so we look forward to the future.

Bettina Burger and Erwin Kump

References:
Von Tauben, Menschen und Schweinen


Optimale Anpassung

Es ist deshalb nur verständlich, dass die betroffenen Hausbesitzer die Tauben möglichst von ihrer Liegenchaft fernhalten wollen. Doch das ist nicht so einfach. Tauben sind intelligente Tiere, die sich optimal an die schwierigen Lebensbedingungen in der Stadt angepasst haben. Vogelscheuchen aller Art, wie z.B. die in Basel weit verbreiteten Kunststoffraben, werden innerhalb kürzester Zeit als ungefährlich erkannt und bleiben fortan ohne Wirkung. Eine ganze Industrie hat sich auf die Produktion so genannter Taubenabwehrsysteme spezialisiert. Mit mehr oder weniger grossem Erfolg wird versucht, die Tauben mit Vogelabwehrnetzen und -gittern, Spanndrahtsystemen, Spikes aus Metall und Kunststoff, Elektroschockanlagen, Ultraschall- und Magnetpulssystemen von Gebäuden fernzuhalten (Haag-Wackernagel 2000). In Basel bieten die Freie Strasse und die Steinenvorstadt ein buntes Bild an funktionierenden und weniger funktionierenden Taubenabwehrsystemen. Die Tauben finden bei weniger wirksamen Systemen meist...
einen Weg, ihre gewohnten Orte wieder zu nutzen. Bei Spanndrahstystemen setzen sich die Tauben einfach auf die Federn oder die Trägerelemente (Abb.1). Im Waaghof, dem Basler Stadtgefängnis, fütterten die Strafgefangenen Tauben vor den Fenstern ihrer Zellen. In der Folge wurden die Fenster mit einem Spanndrahtsystem geschützt, was die Tauben aber nicht hinderte, sich zwischen die Drähte zu zwängen um wieder an ihre gewohnten Orte zu gelangen (Abb. 2). In einer neueren Publikation (Haag-Wackernagel & Geigenfeind 2008) konnten wir zeigen, dass sich eine erwachsene Strassentaube mit einer Brustbreite von 7 cm durch eine Öffnung von 6 x 6 cm zwängen kann.


**Wildschweine legen zu**


«Schweineabwehrwürfel» im Selbstversuch


Unsere Erfahrungen mit Strassentauben und Wildschweinen haben uns eindrücklich gezeigt, dass sich diese Tiere nicht so einfach aus dem menschlichen Lebensraum vertreiben lassen. Beide Tierarten, auch wenn sie äußerlich noch so unterschiedlich sein mögen, besitzen viele Gemeinsamkeiten. Beide sind sie intelligent und anpassungsfähig. Sie lernen schnell, was eine wirkliche Gefahr für sie ist und lassen sich nicht durch falsche Signale vertreiben. Gerade diese enorme Lernfähigkeit hat ja zum Erfolg dieser Kulturfolger geführt. In den nächsten Monaten wollen wir Versuche mit einem selbst konzipierten Wildschweinabwehrsysten durchführen, das verschiedene negative Reize miteinander kombiniert. Wir werden es aber nicht persönlich nehmen, wenn sich auch in diesem Fall die Wildschweine als die Schläfer erweisen.

Daniel Haag-Wackernagel

Literatur


Advisory Board Meeting 2009

The newly formed Scientific Advisory Board visited the Department of Biomedicine for the first time during the 6th DBM-Research Day in February 2009. We are grateful that 8 internationally renowned scientists agreed to evaluate our research groups and advise the DBM on how to improve the quality and impact of our research. Each of the four focal areas of DBM research is advised by two Advisory Board members. Here are short portraits of the members and their research interests:

Mariano Barbacid, Spanish National Cancer Research Centre, Madrid: Ras proteins and Cyclin-dependent kinases in normal homeostasis and in tumor development.

Bob Löwenberg, Erasmus University, Rotterdam: treatment and the pathobiology of leukemia; hematopoietic stem cell transplantation.

Dimitris Kioussis, National Institute for Medical Research, London: molecular events that lead to the stage – and and cell type – specific opening or closing of genes during T cell development.

Kathryn Wood, John Radcliffe Hospital, University of Oxford: mechanisms involved in transplant rejection and immunological tolerance.

Greg Lemke, Salk Institute La Jolla, USA: development of the mammalian nervous and immune systems. Receptor protein-tyrosine kinases (ErbB, Eph, Tyro3) that mediate cellular interactions.

Christian Lüscher, University of Geneva: drug dependence and addiction: Neuroadaptive changes and altered synaptic plasticity in the mesolimbic dopamine system (Kir3/GIRK channels).

Paolo Bianco, Sapienza University, Rome: postnatal stem cells in human bone marrow and other mesoderm-derived tissues; modeling and treatment of bone and other connective tissue diseases, for which there is no cure.

Karl-Heinz Krause, University of Geneva: stem cells; Biology of ageing; NOX family of ROS-generating NADPH oxidases.

During this first visit of the Advisory Board, 12 DBM research groups presented their new results and their plans for the near future. The Advisory Board also visited these laboratories and spoke with lab members. The report of the Advisory Board will provide important guidance for making strategic decisions in the future. We sincerely thank the Advisory Board members for their time and input. The next visit of the Advisory Board is planned for January 2010.

Radek Skoda/Mark Melnyk

Selected publications by DBM members

Below you can find the abstracts of recent articles published by members of the DBM. The abstracts are grouped according to the impact factor of the journal where the work appeared. To be included, the papers must meet the following criteria:

1. The first author, last author or corresponding author (at least one of them) is a member of the DBM.
2. The DBM affiliation must be mentioned in the authors list as it appeared in the journal.
3. The final version of the article must be available (online pre-publications will be included when the correct volume, page numbers etc. becomes available).

We are primarily concentrating on original articles. Due to page constraints, abstracts of publications that appeared in lower ranked journals may not be able to be included. Review articles are generally not considered, unless they appeared in the very top journals (e.g. Cell, Science, Nature, NEJM, etc.). The final decision concerning inclusion of an abstract will be made by the chair of the Department of Biomedicine.

If you wish that your article will appear in the next issue of DBM Facts please submit a pdf file to the Departmental Assistant, Manuela Bernasconi: manuela.bernasconi@unibas.ch

Deadline for the next issue is September 30, 2009.

Reviews Immunology: Perspectives
Affinity threshold for thymic selection through a T-cell receptor–co-receptor zipper

E. Palmer¹ and D. Naeher¹

Abstract:
The affinity of the T-cell receptor (TCR) for self antigen is the basis for the selection of a useful (MHC-restricted) and safe (self-tolerant) T-cell repertoire. However, it has been difficult to understand how thymocytes measure ligand affinity and translate this signal into a cellular response.

In this Opinion article, we propose a new model that describes how the TCR discriminates between low- and high-affinity ligands, which is based on the duration of TCR-ligand interactions and a ‘zipper’ mechanism that mediates the interaction of the TCR and co-receptor molecules to initiate negative-selection signalling.
Different T Cell Receptor Signals Determine CD8\(^+\) Memory Versus Effector Development

E. Teixeiro\(^1\,2\), M. A. Daniels\(^1\,2\), S. E. Hamilton\(^3\), A. G. Schrum\(^1\,5\), R. Bragado\(^4\), S.C. Jameson\(^3\), Ed Palmer\(^1\)

Abstract:
Following infection, naïve CD8\(^+\) T cells bearing pathogen-specific T cell receptors (TCRs) differentiate into a mixed population of short-lived effector and long-lived memory T cells to mediate an adaptive immune response. How the TCR regulates memory T cell development has remained elusive. Using a mutant TCR transgenic model, we found that point mutations in the TCR \(\beta\) transmembrane domain (\(\beta\)TMD) impair the development and function of CD8\(^+\) memory T cells without affecting primary effector T cell responses. Mutant T cells are deficient in polarizing the TCR and in organizing the nuclear factor B signal at the immunological synapse. Thus, effector and memory states of CD8\(^+\) T cells are separable fates, determined by differential TCR signaling.

A Self-Regulatory System of Interlinked Signaling Feedback Loops Controls Mouse Limb Patterning

J.D. Bénazet\(^1\), M. Bischofberger\(^2\), E. Tiecke\(^1\), A. Gonçalves\(^1\), J. F. Martin\(^1\), A. Zuniga\(^1\), F. Naef\(^2\) and R. Zeller

Abstract:
Embryogenesis depends on self-regulatory interactions between spatially separated signaling centers, but few of these are well understood. Limb development is regulated by epithelial-mesenchymal (e-m) feedback loops between sonic hedgehog (SHH) and fibroblast growth factor (FGF) signaling involving the bone morphogenetic protein (BMP) antagonist Gremlin1 (GREM1). By combining mouse molecular genetics with mathematical modeling, we showed that BMP4 first initiates and SHH then propagates e-m feedback signaling through differential transcriptional regulation of Gremlin1 to control digit specification. This switch occurs by linking a fast BMP4/GREM1 module to the slower SHH/GREM1/FGF e-m feedback loop. This self-regulatory signaling network results in robust regulation of distal limb development that is able to compensate for variations by interconnectivity among the three signaling pathways.
Decreed levels of microRNA miR-122 in individuals with hepatitis C responding poorly to interferon therapy

M. Sarasin-Filipowicz1,2,4, J. Krol3,4, I. Markiewicz3, M. H Heim1,2 and W. Filipowicz1

Abstract:
Several microRNAs (miRNAs), including liver-specific miR-122, have been implicated in the control of hepatitis virus (HCV) RNA replication and its response to interferon (IFN) in human hepatoma cells. Our analysis of liver biopsies from subjects with chronic hepatitis C (CHC) undergoing IFN therapy revealed no correlation of miR-122 expression with viral load and markedly decreased pretreatment miR-122 levels in subjects who had no virological response during later IFN therapy; other investigated miRNAs showed only limited changes. These data have implications for the prospect of targeting miRNAs for CHC therapy.

A high-mobility, low-cost phenotype defines human effector-memory CD8+ T cells

G. Zenhaeusern1, P. Gubser1, P. Eisele2, O. Gasser1, A. Steinhuber1, A. Trampuz4, C. Handschin2, A. D. Luster5, and C. Hess1

Abstract:
We measured mRNA expression of genes key to mitochondrial energy metabolism (peroxisome proliferator–activated receptor γ coactivator 1β [PGC-1β], estrogen-related receptor α [ERRα], cytochrome C, ATP synthase, and the uncoupling proteins [UCPs] UCP-2 and -3), quantified ATP contents, and performed calorimetric analyses. Together these assays indicated a high energy efficiency of the high crawling frequency CD8+ T-cell population, and identified differentially regulated heat production among nonlymphoid versus lymphoid homing CD8+ T cells.

1 Department of Biomedicine, University of Basel, Hebelstrasse 20, CH-4031 Basel, Switzerland.
2 Division of Gastroenterology and Hepatology, University Hospital Basel, Petersgraben 4, CH-4031 Basel, Switzerland.
3 Friedrich Miescher Institute for Biomedical Research, Maulbeerstrasse 66, CH-4002 Basel, Switzerland.
Abstract:
We generated mice expressing a full-length Mpl transgene under the control of a 2-kb Mpl promoter in an Mpl–/– background, effectively obtaining mice that express full-length Mpl in the absence of other Mpl isoforms. These mice developed thrombocytosis with platelet levels approximately 5-fold higher than wild-type controls and markedly increased megakaryocyte numbers. The reintroduction of one wild-type Mpl allele restored normal platelet counts. We excluded the deletion of Mpl-tr, a dominant-negative isoform, as the underlying molecular cause for thrombocytosis. Instead, we found that transgene expression driven by the 2-kb Mpl promoter fragment was decreased during late megakaryocyte maturation, resulting in strongly diminished Mpl protein expression in platelets. Because platelets exert a negative feedback on thrombopoiesis by binding and consuming Tpo in the circulation through Mpl, we propose that the severe reduction of Mpl protein in platelets in Mpl-transgenic Mpl–/– mice shifts the equilibrium of this feedback loop, resulting in markedly elevated levels of megakaryocytes and platelets at steady state. Although the mechanism causing decreased expression of Mpl protein in platelets from patients with myeloproliferative disorders differs from this transgenic model, our results suggest that lowering Mpl protein in platelets could contribute to raising the platelet count.

Maintenance of a normal thymic microenvironment and T-cell homeostasis require Smad4-mediated signaling in thymic epithelial cells

L. T. Jeker1, T. Barthlott1, M. P. Keller1, S. Zikelys1, M. Hauri-Hohl1, C. X. Deng2, and G. A. Holländer1

Abstract:
Signals mediated by the transforming growth factor-β superfamily of growth factors have been implicated in thymic epithelial cell (TEC) differentiation, homeostasis, and function, but a direct reliance on these signals has not been established. Here we demonstrate that a block in canonical transforming growth factor-β signaling by the loss of Smad4 expression in TECs leads to qualitative changes in TEC function and a progressively disorganized thymic microenvironment. Moreover, the number of thymus resident early T-lineage progenitors is severely reduced in the absence of Smad4 expression in TECs and directly correlates with extensive thymic and peripheral lymphopenia. Our observations hence place Smad4 within the signaling events in TECs that determine total thymus cellularity by controlling the number of early T-lineage progenitors.

Pronounced thrombocytosis in transgenic mice expressing reduced levels of Mpl in platelets and terminally differentiated megakaryocytes

R. Tiedt1, J. Coers1, S. Ziegler4, A. Wiester4, H. Hao-Shen1, C. Bornmann1, J. Schenkel5, S. Karakhanova5, F. J. de Sauvage6, C. W. Jackson7, and R. C. Skoda1,2,4

Abstract:
We generated mice expressing a full-length Mpl transgene under the control of a 2-kb Mpl promoter in an Mpl–/– background, effectively obtaining mice that express full-length Mpl in the absence of other Mpl isoforms. These mice developed thrombocytosis with platelet levels approximately 5-fold higher than wild-type controls and markedly increased megakaryocyte numbers. The reintroduction of one wild-type Mpl allele restored normal platelet counts. We excluded the deletion of Mpl-tr, a dominant-negative isoform, as the underlying molecular cause for thrombocytosis. Instead, we found that transgene expression driven by the 2-kb Mpl promoter fragment was decreased during late megakaryocyte maturation, resulting in strongly diminished Mpl protein expression in platelets. Because platelets exert a negative feedback on thrombopoiesis by binding and consuming Tpo in the circulation through Mpl, we propose that the severe reduction of Mpl protein in platelets in Mpl-transgenic Mpl–/– mice shifts the equilibrium of this feedback loop, resulting in markedly elevated levels of megakaryocytes and platelets at steady state. Although the mechanism causing decreased expression of Mpl protein in platelets from patients with myeloproliferative disorders differs from this transgenic model, our results suggest that lowering Mpl protein in platelets could contribute to raising the platelet count.
Clonal analysis of deletions on chromosome 20q and JAK2-V617F in MPD suggests that del20q acts independently and is not one of the predisposing mutations for JAK2-V617F

F. X. Schaub¹, R. Jäger², R. Looser¹, H. Hao-Shen¹, S. Hermouet¹,², F. Girodon², A. Tichelli², H. Gisslinger², K. Ralovic²,³, and R. C. Skoda¹

Abstract:
We developed a real-time copy number polymerase chain reaction assay for deletions on chromosome 20q (del20q), screened peripheral blood granulocytes from 664 patients with myeloproliferative disorders, and identified 19 patients with del20q (2.9%), of which 14 (74%) were also positive for JAK2-V617F. To examine the temporal relationship between the occurrence of del20q and JAK2-V617F, we performed colony assays in methylcellulose, picked individual burst-forming units–erythroid (BFU-E) and colony-forming units–granulocyte (CFU-G) colonies, and genotyped each colony individually for del20q and JAK2-V617F. In 2 of 9 patients, we found that some colonies with del20q carried only wild-type JAK2, whereas other del20q colonies were JAK2-V617F positive, indicating that del20q occurred before the acquisition of JAK2-V617F. However, in colonies from 3 of 9 patients, we observed the opposite order of events. The lack of a strict temporal order of occurrence makes it doubtful that del20q represents a predisposing event for JAK2-V617F. In 2 patients with JAK2-V617F and 1 patient with MPL-W515L, microsatellite analysis revealed that del20q affected chromosomes of different parental origin and/or 9pLOH occurred at least twice. The fact that rare somatic events, such as del20q or 9pLOH, occurred more than once in subclones from the same patients suggests that the myeloproliferative disorder clone carries a predisposition to acquiring such genetic alterations.

¹ Department of Biomedicine, Experimental Hematology, University Hospital Basel, Basel, Switzerland
² Center for Molecular Medicine, Austrian Academy of Sciences, Vienna, Austria
³ Inserm Unite Mixte de Recherche (UMR) 892, Institut de Biologie, Nantes, France
⁴ Centre Hospitalier Universitaire, Nantes, France
⁵ Laboratoire d’Hematologie, Centre Hospitalier Universitaire de Dijon, Dijon, France
⁶ Division of Diagnostic Hematology, University Hospital Basel, Basel, Switzerland
⁷ Department of Internal Medicine I, Division of Hematology and Blood Coagulation, Medical University of Vienna, Vienna, Austria

Role of the ATP-Binding Cassette Transporter Abcg2 in the Phenotype and Function of Cardiac Side Population Cells

O. Pfister¹,², A. Oikonomopoulos³, K.-I. Sereti¹,³, R. L. Sohn¹, D. Cullen¹, G.C. Fine¹,², F. Mouquet¹,⁶, K. Westerman¹, R. Liao¹

Abstract:
Recently, the side population (SP) phenotype has been introduced as a reliable marker to identify subpopulations of cells with stem/progenitor cell properties in various tissues. We and others have identified SP cells from postmitotic tissues, including adult myocardium, in which they have been suggested to contribute to cellular regeneration following injury. SP cells are identified and characterized by a unique efflux of Hoechst 33342 dye. Abcg2 belongs to the ATP-binding cassette (ABC) transporter superfamily and constitutes the molecular basis for the dye efflux, hence the SP phenotype, in hematopoietic stem cells. Although Abcg2 is also expressed in cardiac SP (cSP) cells, its role in regulating the SP phenotype and function of cSP cells is unknown. Herein, we demonstrate that regulation of the SP phenotype in cSP cells occurs in a dynamic, age-dependent fashion, with Abcg2 as the molecular determinant of the cSP phenotype in the neonatal heart and another ABC transporter, Mdr1, as the main contributor to the SP phenotype in the adult heart. Using loss- and gain-of-function experiments, we find that Abcg2 tightly regulates cell fate and function. Adult cSP cells isolated from mice with genetic ablation of Abcg2 exhibit blunted proliferation capacity and augmented cell death. Conversely, overexpression of Abcg2 is sufficient to enhance cell proliferation, although with a limitation of cardiomyogenic differentiation. In summary, for the first time, we reveal a functional role for Abcg2 in modulating the proliferation, differentiation, and survival of adult cSP cells that goes beyond its distinct role in Hoechst dye efflux.

¹ Cardiac Muscle Research Laboratory, Cardiovascular Division, Department of Medicine, Harvard Medical School, Boston, Mass
² Department of Anesthesia, Brigham and Women’s Hospital, Harvard Medical School, Boston, Mass
³ School of Medicine, University of Crete, Greece
⁴ University of Washington School of Medicine, Seattle
⁵ Myocardial Research, Department of Biomedicine, and Division of Cardiology, University Hospital Basel, Switzerland
⁶ Cardiologie C, Lille University Hospital, France
Interferon-γ Regulates Idiopathic Pneumonia Syndrome, a Th17+CD4+ T-Cell–mediated Graft-versus-Host Disease

N. Mauermann1, J. Burian1, C. von Garnier1, S. Dirnhofer2, D. Germano1, C. Schuett1, M. Tamm4, R. Bingisser5, U. Eriksson1, and L. Hunziker1

Abstract:
Rationale: Pulmonary complications of hematopoietic stem cell transplantation include infections and graft-versus-host diseases, such as idiopathic pneumonia syndrome (IPS). Conflicting data exist regarding the role of the interferon (IFN)-γ–producing Th1 CD4+ T-cell subset and IL-17A in IPS.

Objectives: To determine the role of IFN-γ and IL-17A in the establishment of pulmonary graft-versus-host disease.

Methods: A semiallogeneic murine model based on C57BL/6 x BALB/c as recipients with transplantation of BALB/c RAG2–/– bone marrow and transfer of different genetic knockout T cells (T-bet –/–, IFN-γ –/–, IFN-γR–/–) on a BALB/c background. Lung tissue was examined for parenchymal changes and infiltrating cells by histology and fluorescence-activated cell sorter analysis.

Measurements and Main Results: After transfer of semiallogeneic bone marrow together with donor CD4+ T cells lacking IFN-γ or T-bet—a T-box transcription factor controlling Th1 commitment—we found severe inflammation in the lungs, but no enhancement in other organs. In contrast, wild-type donor CD4+ T cells mediated minimal inflammation only, and donor CD8+ T cells were not required for IPS development. Mechanistically, the absence of IFN-γ or IFN-γ signaling in pulmonary parenchymal cells promoted expansion of IL-17A–producing CD4+ T cells and local IL-17A release. In vivo depletion of IL-17A reduced disease severity.

Conclusions: One mechanism of IFN-γ protection against IPS is negative regulation of the expansion of pathogenic IL-17A–producing CD4+ T cells through interaction with the IFN-γ receptor on the pulmonary parenchymal cell population.

Impaired translation of CCAAT/enhancer binding protein α mRNA in bronchial smooth muscle cells of asthmatic patients

P. Borger1, N. Miglino1, M. Baraket2, J. L. Black2, M. Tamm1 and M. Roth1,3

Abstract:
Background: Bronchial smooth muscle (BSM) cells of asthmatic patients have an impaired expression of CCAAT/enhancer binding protein (C/EBP) α, which is associated with increased proliferation.

Objective: We sought to assess the translational regulation of CEBPA mRNA in cultured BSM cells of healthy control subjects (n = 11) and asthmatic patients (n = 12).

Methods: Translation efficiency was studied by using a translation control reporter system driven by the control elements present in the CEBPA mRNA. Translation efficiency was determined by the ratio of 2 artificial hemagglutinin (HA.11) proteins: p23 and p12. We also analyzed levels of proteins that control translation of CEBPA mRNA, namely heterogeneous nuclear ribonucleoprotein E2, calreticulin, eukaryotic translation initiation factor (eIF4E), and 4E binding protein.

Results: Compared with healthy control subjects, BSM cells of asthmatic patients proliferate faster (2.1-fold) and are primed for IL-6 secretion. Real-time RT-PCR showed that BSM cells of asthmatic patients express normal levels of CEBPA mRNA, whereas they express lower levels of C/EBPα (p42). Transient transfections with the translation control reporter system construct showed a disturbed p12/p23 ratio in BSM cells of asthmatic patients relative to healthy control subjects, which coincided with lower levels of eIF4E.

Conclusion: BSM cells of asthmatic patients have normal levels of CEBPA mRNA but inadequately reinitiate the translation into C/EBPα. Impaired translation control upstream of eIF4E might underlie the observed increased proliferation and priming of BSM cells of asthmatic patients.

1Pulmonary Cell Research, Department of Biomedicine, University Hospital Basel, Basel, Switzerland
2Pharmacology, University of Sydney, Sydney, Australia
3Woolcock Institute of Medical Research, University of Sydney, Sydney, Australia
Graafian ovarian follicles consist of follicular fluid, one single mature oocyte, and several hundred thousands of granulosa cells (GCs). Until now, luteinizing GCs have been considered to be terminally differentiated, destined to undergo death after ovulation. Present concepts of luteal function, endocrine regulation of early pregnancy, and the recruitment of new ovarian follicles are all based on the cyclical renewal of the entire population of GCs. We now demonstrate that luteinizing GCs isolated from the ovarian follicles of infertile patients and sorted with flow cytometry based upon the presence of their specific marker, the follicle-stimulating hormone receptor (FSHR), can be maintained in culture over prolonged periods of time in the presence of the leukemia-inhibiting factor (LIF). Under those conditions the markers of GC function such as FSHR and aromatase gradually disappeared. POU5F1 (POU domain, class 5, homeobox 1), a typical stem cell marker, was expressed throughout the culture, but germ line cell markers such as nanog, vasa, and stellar were not. Mesenchymal lineage markers such as CD29, CD44, CD90, CD105, CD117, and CD166, but not CD73, were expressed by substantial subpopulations of GCs. The multipotency of a subset of GCs was established by in vitro differentiation into other cell types, otherwise not present within ovarian follicles, such as neurons, chondrocytes, and osteoblasts. Follicle-derived stem cells were also able to survive when transplanted into the backs of immunocompetent mice, in vivo generating tissues of mesenchymal origin. The unexpected findings of multipotency of cells with prolonged lifespans originating from ovarian follicles are likely to have a significant impact on evolving theories in ovarian pathophysiology, particularly with reference to ovarian endometriosis and ovarian cancer.

Abstract:
Loss of expression of the cell–cell adhesion molecule E-cadherin is a hallmark of epithelial–mesenchymal transition (EMT) in development and in the progression from epithelial tumours to invasive and metastatic cancers. Here, we demonstrate that the loss of E-cadherin function up-regulates expression of the neuronal cell adhesion molecule (NCAM). Subsequently, a subset of NCAM translocates from fibroblast growth factor receptor (FGFR) complexes outside lipid rafts into lipid rafts where it stimulates the non-receptor tyrosine kinase p59F110 leading to the phosphorylation and activation of focal adhesion kinase and the assembly of integrin-mediated focal adhesions. Ablation of NCAM expression during EMT inhibits focal adhesion assembly, cell spreading and EMT. Conversely, forced expression of NCAM induces epithelial cell delamination and migration, and high NCAM expression correlates with tumour invasion. These results establish a mechanistic link between the loss of E-cadherin expression, NCAM function, focal adhesion assembly and cell migration and invasion.

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1 Department of Biomedicine, Institute of Biochemistry and Genetics, University of Basel, Basel, Switzerland
2 Institute of Pathology, University of Basel, Basel, Switzerland
3 Division of Molecular Biology, The Netherlands Cancer Institute, Amsterdam, The Netherlands
4 Division of Molecular Genetics, The Netherlands Cancer Institute, Amsterdam, The Netherlands

The Multipotency of Luteinizing Granulosa Cells Collected from Mature Ovarian Follicles

K. Kossowska-Tomaszczuk1,2, C. De Geyter1,2, M. De Geyter1, I. Martin2, W. Holzgreve1, A. Scherberich2 and H. Zhang2

Abstract:
Graafian ovarian follicles consist of follicular fluid, one single mature oocyte, and several hundred thousands of granulosa cells (GCs). Until now, luteinizing GCs have been considered to be terminally differentiated, destined to undergo death after ovulation. Present concepts of luteal function, endocrine regulation of early pregnancy, and the recruitment of new ovarian follicles are all based on the cyclical renewal of the entire population of GCs. We now demonstrate that luteinizing GCs isolated from the ovarian follicles of infertile patients and sorted with flow cytometry based upon the presence of their specific marker, the follicle-stimulating hormone receptor (FSHR), can be maintained in culture over prolonged periods of time in the presence of the leukemia-inhibiting factor (LIF). Under those conditions the markers of GC function such as FSHR and aromatase gradually disappeared. POU5F1 (POU domain, class 5, homeobox 1), a typical stem cell marker, was expressed throughout the culture, but germ line cell markers such as nanog, vasa, and stellar were not. Mesenchymal lineage markers such as CD29, CD44, CD90, CD105, CD117, and CD166, but not CD73, were expressed by substantial subpopulations of GCs. The multipotency of a subset of GCs was established by in vitro differentiation into other cell types, otherwise not present within ovarian follicles, such as neurons, chondrocytes, and osteoblasts. Follicle-derived stem cells were also able to survive when transplanted into the backs of immunocompetent mice, in vivo generating tissues of mesenchymal origin. The unexpected findings of multipotency of cells with prolonged lifespans originating from ovarian follicles are likely to have a significant impact on evolving theories in ovarian pathophysiology, particularly with reference to ovarian endometriosis and ovarian cancer.

1 Woman’s Hospital, University of Basel, Basel, Switzerland
2 Department of Research, University of Basel, Basel, Switzerland
Pharmacokinetics and Pharmacodynamic Effects of Oral GLP-1 and PYY\textsuperscript{3-36}: A Proof-of-concept Study in Healthy Subjects

C. Beglinger\textsuperscript{1,2}, B. Poller\textsuperscript{1,3}, E. Arbit\textsuperscript{1}, C. Ganzoni\textsuperscript{1}, S. Gass\textsuperscript{1}, I. Gomez-Orellana\textsuperscript{4} and J. Drewe\textsuperscript{1,3}

Abstract:
This proof-of-concept study was performed in order to establish the pharmacokinetics and pharmacodynamics of increasing oral doses of the satiety peptides glucagon-like peptide-1 (GLP-1) and peptide YY\textsubscript{3-36} (PYY\textsubscript{3-36}). Six healthy male subjects were given oral doses of either a placebo or GLP-1 in a dose-escalating schedule (doses of 0.5, 1.0, 2.0, and 4.0 mg). Next, another group of six healthy male subjects were given oral doses of either a placebo or PYY\textsubscript{3-36} in the same pattern of escalating doses (doses of 0.25, 0.5, 1.0, 2.0, and 4.0 mg). In healthy male volunteers, (i) oral administration of either of the peptides induced a rapid and dose-dependent increase in plasma drug concentrations; (ii) oral administration of GLP-1 induced a potent effect on insulin release; and (iii) both peptides suppressed ghrelin secretion. In conclusion, this study showed, for the first time, that satiety peptides such as GLP-1 and PYY\textsubscript{3-36} can be orally delivered safely and effectively in humans.

Neuregulin Signaling Is Dispensable for NMDA- and GABAA-Receptor Expression in the Cerebellum In Vivo

N. Gajendran\textsuperscript{1}, J. P. Kapfhammer\textsuperscript{2}, E. Lain\textsuperscript{1}, M. Canepari\textsuperscript{3}, K. Vogt\textsuperscript{3}, W. Wisden\textsuperscript{4}, and H. R. Brenner\textsuperscript{1}

Abstract:
Neuregulin-1s (NRG-1s) are a family of growth and differentiation factors with multiple roles in the development and function in different organs including the nervous system. Among the proposed functions of NRG-1s in the nervous system is the regulation of genes encoding certain neurotransmitter receptors during synapse formation as well as of other aspects of synaptic function. Here, we have examined, in granule cells of the cerebellum in vivo, the role of NRGs in the induction of NMDA receptor (NMDA-R) and GABA\textsubscript{A} receptor (GABA\textsubscript{A}-R), which are thought to be induced by NRG-1 secreted by the synaptic inputs. To this end, we used the Cre/loxP system to genetically ablate the NRG receptors ErbB2 and ErbB4 selectively in these cells, thus eliminating all NRG-mediated signaling to them. Unlike previous reports using cultured granule cells to address the same question, we found that the developmental expression patterns of the mRNAs encoding the NR2C subunit of the NMDA-R and the \(\beta_2\)-subunit of the GABA\textsubscript{A}-R is normal in mice lacking the NRG receptors ErbB2 and ErbB4. Likewise, no alterations in cerebellar morphology nor in certain aspects of cerebellar wiring were resolved in these mutants. We conclude that NRG/Erb signaling to the granule cells is dispensable for the normal development of their synaptic inputs.
The GABA$\text{B}_{1a}$ Isoform Mediates Heterosynaptic Depression at Hippocampal Mossy Fiber Synapses

N. Guetg$^{1,3}$, R. Seddik$^1$, R. Vigot$^1$, R. Turecek$^1$, M. Gassmann$^1$, K. E. Vogt$^2$, H. Bräuner-Osborne$^{1,4}$, R. Shigemoto$^{5,6,7}$, O. Kretz$^3$, M. Frotscher$^1$, A. Kulik$^3$, and B. Bettler$^1$

Abstract:
NC$\text{B}_{1a}$ receptor subtypes are based on the subunit isoforms G$\text{B}_{1a}$ and G$\text{B}_{1b}$, which associate with G$\text{B}_{2}$ subunits to form pharmacologically indistinguishable G$\text{B}_{1a,2}$ and G$\text{B}_{1b,2}$ receptors. Studies with mice selectively expressing C$\text{B}_{1a}$ or C$\text{B}_{1b}$ subunits revealed that C$\text{B}_{1a,2}$ receptors are more abundant than C$\text{B}_{1b,2}$ receptors at glutamatergic terminals. Accordingly, it was found that C$\text{B}_{1a,2}$ receptors are more efficient than C$\text{B}_{1b,2}$ receptors in inhibiting glutamate release when maximally activated by exogenous application of the agonist baclofen. Here, we used a combination of genetic, ultrastructural and electrophysiological approaches to analyze to what extent G$\text{B}_{1a,2}$ and G$\text{B}_{1b,2}$ receptors inhibit glutamate release in response to physiological activation. We first show that at hippocampal mossy fiber (MF) terminal synapses more C$\text{B}_{1a,2}$ than C$\text{B}_{1b,2}$ protein is present at presynaptic sites, consistent with the findings at other glutamatergic synapses. In the presence of baclofen at concentrations $\geq 1 \mu\text{M}$, both C$\text{B}_{1a,2}$ and C$\text{B}_{1b,2}$ receptors contribute to presynaptic inhibition of glutamate release. However, at lower concentrations of baclofen, selectively C$\text{B}_{1a,2}$ receptors contribute to presynaptic inhibition. Remarkably, exclusively C$\text{B}_{1a,2}$ receptors inhibit glutamate release in response to synaptically released GABA. Specifically, we demonstrate that selectively C$\text{B}_{1a,2}$ receptors mediate heterosynaptic depression of MF transmission, a physiological phenomenon involving transsynaptic inhibition of glutamate release via presynaptic C$\text{B}_{1a,2}$ receptors. Our data demonstrate that the difference in C$\text{B}_{1a,2}$ and C$\text{B}_{1b,2}$ protein levels at MF terminals is sufficient to produce a strictly C$\text{B}_{1a,2}$-specific effect under physiological conditions. This consolidates that the differential subcellular localization of the C$\text{B}_{1a}$ and C$\text{B}_{1b}$ proteins is of regulatory relevance.

1 Department of Biomedicine, Institute of Physiology, Pharmazentrum, University of Basel, 4056 Basel, Switzerland
2 Division of Pharmacology and Neurobiology, Biozentrum, University of Basel, 4056 Basel, Switzerland
3 Institute of Anatomy and Cell Biology, Department of Neuranatomy, University of Freiburg, 79104 Freiburg, Germany
4 Division of Medical Chemistry, Faculty of Pharmaceutical Sciences, University of Copenhagen, 2100 Copenhagen, Denmark
5 Division of Cellular Structure, National Institute for Physiological Sciences, and
6 Department of Physiological Sciences, The Graduate University of Advanced Studies (Sokendai), Myodai, Okazaki 444-8787, Japan
7 Solution Oriented Research for Science and Technology, Japan Science and Technology Corporation, Kawaguchi 332-0012, Japan

Immediate and Sustained Blood Pressure Lowering by Urocortin 2
A Novel Approach to Antihypertensive Therapy?

T. Dieterle$^{1,2}$, S. Meili-Butz$^1$, K. Bühler$^1$, C. Morandi$^1$, D. John$^1$, P. T. Buser$^1$, J. Rivier$^3$, W. W. Vale$^1$, K. L. Peterson$^4$ and M. Brink$^1$

Abstract:
Recently, novel corticotropin-releasing factor-related peptides, named urocortin 1, 2, and 3, and a distinct cardiac and peripheral vascular receptor (corticotropin-releasing factor receptor 2) were described being part of a peripheral corticotropin-releasing factor system modulating cardiovascular function in response to stress. Vasorelaxation and blood pressure lowering have been reported after acute administration of these peptides. No data are available on the acute and chronic effects of urocortin 2 on blood pressure in models of arterial hypertension. To test these effects, hypertensive salt-sensitive and normotensive salt-resistant Dahl rats were randomly assigned to twice-daily applications of urocortin 2 or vehicle for 5 weeks. Blood pressure, heart rate, and left ventricular dimension and function were recorded at baseline, after initial application, and, together with cardiac and aortic expression of urocortin 2 and its receptor, after 5 weeks of treatment. Urocortin 2 significantly reduced blood pressure in hypertensive rats without affecting heart rate. Long-term urocortin 2 treatment in hypertensive rats induced sustained blood pressure reduction and diminished the development of hypertension-induced left ventricular hypertrophy and the deterioration of left ventricular contractile function. Corticotropin-releasing factor receptor 2 expression was preserved despite chronic stimulation by urocortin 2. In conclusion, our study shows that, in an animal model of arterial hypertension, urocortin 2 has immediate and sustained blood pressure-lowering effects. Beneficial effects on blood pressure, left ventricular dimension, and function, together with preserved receptor expression, suggest that corticotropin-releasing factor receptor 2 stimulation by urocortin 2 may represent a novel approach to the treatment of arterial hypertension.

1 Department of Biomedicine, University of Basel and University Hospital Basel, Basel, Switzerland
2 Division of Cardiology, University Hospital Basel, Basel, Switzerland
3 Clayton Foundation Laboratories, salk Institute, La Jolla, Calif
4 Institute of Molecular Medicine, University of California San Diego, La Jolla
Gli2 upregulates cFlip and renders basal cell carcinoma cells resistant to death ligand-mediated apoptosis

E. Kump1, J. Ji1, M. Wernli1, P. Häusermann2 and P. Erb1

Abstract:
Mutations in the Hedgehog signaling pathway is responsible for the formation of various cancers, including some forms of basal cell carcinoma (BCC). Uncontrolled Hedgehog signaling leads to overexpression of the zinc-finger Gli transcription factors, among which Gli2 plays a central role. We found that high Gli2 expression induced the concomitant high expression of the caspase 8 inhibitor, cFlip, and thereby counteracts death-ligand-mediated apoptosis. By investigating the cFlip promoter, Gli2 binding sites were identified and confirmed. Gli2 gene silencing by RNA interference broke the apoptosis resistance via cFlip downregulation. The direct functional connection between Gli2 and cFlip was not only demonstrated in a keratinocytic cell line but also in BCC tissue. As cFlip and Bcl-2 are highly expressed in BCCs, as a consequence of high Gli2 expression, this may explain the marked resistance of the tumor to the extrinsic and intrinsic apoptotic pathway. We could now demonstrate that Gli2 gene silencing in BCC tissues made the tumor sensitive to TRAIL (tumor necrosis factor-related apoptosis-inducing ligand)-mediated cell death by downregulating cFlip. As Gli2 silencing does not only downregulate cFlip, but also Bcl-2, Gli2 could be a key target for a novel therapeutic approach in tumors with dysregulated Hedgehog signaling.

1 Department of Biomedicine, Institute for Medical Microbiology, University of Basel, Petersplatz, Basel, Switzerland
2 Department of Dermatology, University Hospital Basel, Basel, Switzerland

Increasing the number of diagnostic mutations in malignant hyperthermia

S. Levano1,2, M. Vukcevic1,2, M. Singer1,2, A. Matter1,2, S. Treves1,2, A. Urwyler1,2 and T. Girard1,2

Abstract:
Malignant hyperthermia (MH) is an autosomal dominant disorder characterized by abnormal calcium homeostasis in skeletal muscle in response to triggering agents. Today, genetic investigations on ryanodine receptor type 1 (RYR1) gene and 1 subunit of the dihydropyridine receptor (DHPR) (CACNA1S) gene have improved the procedures associated with MH diagnosis. In approximately 50% of MH cases a causative RYR1 mutation was found. Molecular genetic testing based on RYR1 mutations for MH diagnosis is challenging, because the causative mutations, most of which are private, are distributed throughout the RYR1 gene. A more comprehensive genetic testing procedure is needed. Therefore, we aim to expand the genetic information related to MH and to evaluate the effect of mutations on the MH phenotype. Performing an in-depth mutation screening of the RYR1 transcript sequence in 36 unrelated MH susceptible (MHS) patients, we identified 17 novel, five rare, and eight non-disease-causing variants in 23 patients. The 13 remaining MHS patients presented no known variants, neither in RYR1 nor in the CACNA1S binding regions to RYR1. The 17 novel variants were found to affect highly conserved amino acids and were absent in 100 controls. Excellent genotype-phenotype correlations were found by investigating 21 MHS families - a total of 186 individuals. Epstein-Barr virus (EBV) lymphoblastoid cells carrying four of these novel mutations showed abnormal calcium homeostasis. The results of this study contribute to the establishment of a robust genetic testing procedure for MH diagnosis.

1 Department of Biomedicine, University Hospital Basel, Basel, Switzerland
2 Department of Anesthesia, University Hospital Basel, Basel, Switzerland
FTY720 therapy exerts differential effects on T cell subsets in multiple sclerosis

M. Mehling1, V. Brinkmann2, J. Antel3, A. Bar-Or1, N. Goebels4, C. Vedrine5, C. Kristofic2, J. Kuhle1, R.L.P. Lindberg1, and L. Kappos1

Abstract:
Background: The oral immunomodulator FTY720 has shown efficacy in patients with relapsing multiple sclerosis (MS). FTY720 functionally antagonizes sphingosine 1-phosphate receptor-1 (S1P1) on T cells and consequently inhibits S1P/S1P1-dependent lymphocyte egress from secondary lymphoid organs. Little is known about the phenotype and function of T cells remaining in peripheral blood during long-term FTY720 treatment.

Methods: T cells from FTY720-treated, interferon-beta (IFNβ)-treated and untreated patients with MS, and healthy donors (HD) were analyzed with respect to T cell subpopulation composition, proliferation, and cytokine production.

Results: In FTY720-treated patients (n = 16), peripheral blood CD4+ and CD8+ T cell counts were reduced by approximately 80% and 60% when compared to the other groups (IFNβ: n = 7; untreated: n = 5; HD: n = 10). This related to selective reduction of naive (CCR7+CD45RA+) and central memory (CCR7+CD45RA–) T cells (TCM), and resulted in a relative increase of peripheral effector memory (CCR7–CD45RA– [TEM] and CCR7–CD45RA+ [TEMRA]) T cells. The remaining blood T cell populations displayed a reduced potential to secrete IL-2 and to proliferate in vitro, but rapidly produced interferon-gamma upon reactivation, confirming a functional TEM/TEMRA phenotype. Neither FTY720 nor FTY720-P directly suppressed proliferation or cytokine production by T cells.

Conclusion: Therapeutic dosing of FTY720 reduces naive T cells and TCM, but not TEM, in blood, without affecting T cell function. This is presumably because naive T cells and TEM express the homing receptor CCR7, allowing recirculation to secondary lymphoid tissues on a regular basis and, thus, trapping of the cells by FTY720 in lymph nodes.

Polyomavirus BK Replication Dynamics In Vivo and In Silico to Predict Cytopathology and Viral Clearance in Kidney Transplants

G. A. Funk1, R. Gosert1, P. Comoli2, F. Ginevri3 and H. H. Hirsch4

Abstract:
Fast BK virus (BKV) replication in renal tubular epithelial cells drives polyomavirus-BK-associated nephropathy (PVAN) to premature kidney transplant (KT) failure. BKV replicates or replicates in urothelial cells, but remains asymptomatic in two-thirds of affected KT patients. Comparing 518 day-matched plasma-urine samples from 223 KT patients, BKV loads were ~3000-fold higher in urine than in plasma (p < 0.000001). Molecular and quantitative parameters indicated that >95% of urine BKV loads resulted from urothelial replication and <5% from tubular epithelial replication. Fast BKV replication dynamics in plasma and urine with half-lives of <12 h accounted for daily urothelial and tubular epithelial cell loss of ~x104 and 6-x107, respectively. BKV dynamics in both sites were only partly linked, with full and partial discordance in 36% and 32%, respectively. Viral expansion was best explained by models where BKV replication started in the kidney followed by urothelial amplification and tubular epithelial cell cross-feeding reaching a dynamic equilibrium after ~10 weeks. Curtailing intrarenal replication by 50% was ineffective and >80% was required for clearing viremia within 7 weeks, but viruria persisted for >14 weeks. Reductions >90% cleared viremia and viruria by 3 and 10 weeks, respectively. The model was clinically validated in prospectively monitored KT patients supporting >80% curtailing for optimal interventions.
Regulation of Fat-Stimulated Neurotensin Secretion in Healthy Subjects

J. Drewe1,3, S. Mihailovic1,2, M. D’Amato4, and C. Beglinger1,2

Abstract:

Context: Cholecystokinin (CCK) and neurotensin are stimulated during meal intake by the presence of fat in the small intestine. The sequence of events suggests that fat hydrolysis is crucial for triggering the release.

Objective: The aim of this study was to investigate whether CCK mediated the effect of intraduodenal (ID) fat on neurotensin secretion via CCK-1 receptors.

Setting: This was a single center study; 34 male volunteers were studied in consecutive, randomized, double-blind, cross-over studies.

Subjects and Methods: CCK and neurotensin release were quantified in: 1) 12 subjects receiving an ID fat infusion with or without 60 mg orlistat, an irreversible inhibitor of gastrointestinal lipases, in comparison to vehicle; 2) 12 subjects receiving ID long chain fatty acids (C18s), ID medium chain fatty acids, or ID vehicle; and 3) 10 subjects receiving ID C18 with and without the CCK-1 receptor antagonist dexloxiglumide or ID vehicle plus iv saline (placebo). Hormone concentrations were measured by specific RIA systems.

Results: ID fat induced a significant increase in CCK and neurotensin concentrations ($P < 0.001$–$0.002$). Inhibition of fat hydrolysis by orlistat abolished both effects. C18 stimulated CCK and neurotensin release ($P < 0.001$, respectively), whereas medium chain fatty acid was ineffective. Dexloxiglumide administration partially blocked the effect of C18 on neurotensin; the effect was only present in the first phase of neurotensin secretion.

Conclusions: Generation of C18 through hydrolysis of fat is a critical step for fat-induced stimulation of neurotensin in humans; the signal is in part mediated via CCK release and CCK-1 receptors.

CD11b+ Monocytes Abrogate Th17 CD4+ T Cell-Mediated Experimental Autoimmune Myocarditis

A. Valaperti1, R. R. Marty1, G. Kania1, D. Germano1, N. Mauermann1, S. Dirnhofer2, B. Leinenstoll1, P. Blyszczuk1, C. Dong1, C. Mueller1, L. Hunziker1,3, and U. Eriksson1,3

Abstract:

Experimental autoimmune myocarditis (EAM) represents a Th17 T cell-mediated mouse model of postinflammatory heart disease. In BALB/c wild-type mice, EAM is a self-limiting disease, peaking 21 days after α-myosin H chain peptide (MyHC-α)/CFA immunization and largely resolving thereafter. In IFN-γR–/– mice, however, EAM is exacerbated and shows a chronic progressive disease course. We found that this progressive disease course paralleled persistently elevated IL-17 release from T cells infiltrating the hearts of IFN-γR–/– mice 30 days after immunization. In fact, IL-17 promoted the recruitment of CD11b+ monocytes, the major heart-infiltrating cells in EAM. In turn, CD11b+ monocytes suppressed MyHC-α-specific Th17 T cell responses IFN-γ-dependently in vitro. In vivo, injection of IFN-γR–/–CD11b+ monocytes, suppressed MyHC-α-specific T cells, and abrogated the progressive disease course in IFN-γR–/– mice. Finally, cojection of MyHC-α-specific, but not OVA-transgenic, IFN-γ-releasing CD4+ Th1 T cell lines, together with MyHC-α-specific Th17 T cells protected RAG2–/– mice from EAM. In conclusion, CD11b+ monocytes play a dual role in EAM: as a major cellular substrate of IL-17-induced inflammation and as mediators of an IFN-γ-dependent negative feedback loop confining disease progression.
Prominin-1\(^+\)/CD133\(^+\) bone marrow-derived heart-resident cells suppress experimental autoimmune myocarditis

G. Kania\(^1\), P. Blyszczuk\(^1\), A. Valaperti\(^1\), T. Dieterle\(^2\), B. Leimenstoll\(^3\), S. Dirnhuber\(^4\), H. Zulewski\(^5\) and U. Eriksson\(^6\)

Abstract:
Aims: Experimental autoimmune myocarditis (EAM) is a CD4\(^+\) T cell-mediated mouse model of inflammatory heart disease. Tissue-resident bone marrow-derived cells adopt different cellular phenotypes depending on the local milieu. We expanded a specific population of bone marrow-derived prominin-1-expressing progenitor cells (PPC) from healthy heart tissue, analysed their plasticity, and evaluated their capacity to protect mice from EAM and heart failure.

Methods and results: PPC were expanded from healthy mouse hearts. Analysis of CD45.1/CD45.2 chimera mice confirmed bone marrow origin of PPC. Depending on in vitro culture conditions, PPC differentiated into macrophages, dendritic cells, or cardiomyocyte-like cells. In vivo, PPC acquired a cardiac phenotype after direct injection into healthy hearts. Intravenous injection of PPC into myosin alpha heavy chain/complete Freund’s adjuvant (MyHC-/CFA)-immunized BALB/c mice resulted in heart-specific homing and differentiation into the macrophage phenotype. Histology revealed reduced severity scores for PPC-treated mice compared with control animals (treated with phosphate-buffered saline (PBS) or crude bone marrow at day 21 after MyHC-/CFA immunization). Echocardiography showed preserved fractional shortening and velocity of circumferential shortening in PPC but not PBS-treated MyHC-/CFA-immunized mice. In vitro and in vivo data suggested that interferon-\(\gamma\)-signalling on PPC was critical for nitric oxide-mediated suppression of heart-specific CD4\(^+\) T cells. Accordingly, PPC from interferon-\(\gamma\)-receptor-deficient mice failed to protect MyHC-/CFA-immunized mice from EAM.

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Mycolic Acids Constitute a Scaffold for Mycobacterial Lipid Antigens Stimulating CD1-Restricted T Cells

E. Layre\(^1,2\), A. Collmann\(^1\), M. Bastian\(^1\), S. Mariotti\(^3\), J. Czaplicki\(^1,2\), J. Prandi\(^1,2\), L. Mori\(^1\), S. Stenger\(^4\), G. De Libero\(^1\), G. Puzo\(^1,2\) and M. Gilleron\(^1,2\)

Abstract:
CD1-restricted lipid-specific T lymphocytes are primed during infection with Mycobacterium tuberculosis, the causative agent of tuberculosis. Here we describe the antigenicity of glycerol monomycolate (GroMM), which stimulates CD1b-restricted CD4\(^+\) T cell clones. Chemical characterization of this antigen showed that it exists as two stereoisomers, one synthetic isomer being more stimulatory than the other. The hydroxyl groups of glycerol and the mycolic acid length are critical for triggering the T cell responses. GroMM was presented by M. tuberculosis-infected dendritic cells, demonstrating that the antigen is available for presentation during natural infection. Ex vivo experiments showed that GroMM stimulated T cells from vaccinated or latently infected healthy donors but not cells from patients with active tuberculosis, suggesting that GroMM-specific T cells are primed during infection and their detection correlates with lack of clinical active disease.

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1 Experimental Critical Care, Department of Biomedicine, University Hospital, Hebelstrasse 20, CH-4031 Basel, Switzerland
2 Clinical Cardiology, Department of Internal Medicine, University Hospital, Hebelstrasse 20, CH-4031 Basel, Switzerland
3 General Internal Medicine, Department of Internal Medicine, University Hospital, Hebelstrasse 20, CH-4031 Basel, Switzerland
4 Institute of Pathology, University Hospital, Hebelstrasse 20, CH-4031 Basel, Switzerland
5 Division of Endocrinology, Diabetes and Clinical Nutrition, University Hospital, Hebelstrasse 20, CH-4031 Basel, Switzerland
6 Department of Cardiology, University Hospital, Rämistrasse 100, CH-8091 Zurich, Switzerland

1 CNRS; IPBS (Institut de Pharmacologie et de Biologie Structurale); 205 route de Narbonne, F-31077 Toulouse, France
2 Université de Toulouse; UPS; IPBS; F-31077 Toulouse, France
3 Experimental Immunology, Department of Biomedicine, University Hospital, 4031 Basel, Switzerland
4 Institut für Medizinische Mikrobiologie und Hygiene, 89081 Ulm, Germany
The Sushi Domains of Secreted GABA<sub>B1</sub> Isoforms Selectively Impair GABA<sub>B</sub> Heteroreceptor Function

J.Y. Tiao<sup>1</sup>, A. Bradaia<sup>1</sup>, B. Biermann<sup>1</sup>, K. Kaupmann<sup>2</sup>, M. Metz<sup>1</sup>, C. Haller<sup>1</sup>, A. G. Rolink<sup>3</sup>, E. Pless<sup>4</sup>, P.N. Barlow<sup>4</sup>, M. Gassmann<sup>1</sup>, and B. Bettler<sup>1</sup>

Abstract:

GABA<sub>B</sub> receptors are the G-protein-coupled receptors for γ-aminobutyric acid (GABA), the main inhibitory neurotransmitter in the brain. GABA<sub>B</sub> receptors are promising drug targets for a wide spectrum of psychiatric and neurological disorders. Receptor subtypes exhibit no pharmacological differences and are based on the subunit isoforms GABA<sub>B1a</sub> and GABA<sub>B1b</sub>. GABA<sub>B1a</sub> differs from GABA<sub>B1b</sub> in its ectodomain by the presence of a pair of conserved protein binding motifs, the sushi domains (SDs). Previous work showed that selectively GABA<sub>B1a</sub> contributes to heteroreceptors at glutamatergic terminals, whereas both GABA<sub>B1a</sub> and GABA<sub>B1b</sub> contribute to autoreceptors at GABAergic terminals or to postsynaptic receptors. Here, we describe GABA<sub>B1j</sub>, a secreted GABA<sub>B1</sub> isoform comprising the two SDs. We show that the two SDs, when expressed as a soluble protein, bind to neuronal membranes with low nanomolar affinity. Soluble SD protein, when added at nanomolar concentrations to dissociated hippocampal neurons or to acute hippocampal slices, impairs the inhibitory effect of GABA<sub>B</sub> heteroreceptors on evoked and spontaneous glutamate release. In contrast, soluble SD protein neither impairs the activity of GABA<sub>B</sub> autoreceptors nor impairs the activity of postsynaptic GABA<sub>B</sub> receptors. We propose that soluble SD protein scavenges an extracellular binding partner that retains GABA<sub>B1a</sub>-containing heteroreceptors in proximity of the presynaptic release machinery. Soluble GABA<sub>B1</sub> isoforms like GABA<sub>B1j</sub> may therefore act as dominant-negative inhibitors of heteroreceptors and control the level of GABA-mediated inhibition at glutamatergic terminals. Of importance for drug discovery, our data also demonstrate that it is possible to selectively impair GABA<sub>B1a</sub> heteroreceptors by targeting their SDs.

1 From the Department of Biomedicine, Institute of Physiology, Pharmazentrum, University of Basel, CH-4056 Basel, Switzerland
2 Novartis Institutes for BioMedical Research, Novartis Pharma AG, CH-4002 Basel, Switzerland
3 Department of Biomedicine, Developmental and Molecular Immunology, University of Basel, CH-4058 Basel, Switzerland
4 Edinburgh Biomolecular NMR Unit, University of Edinburgh, Edinburgh EH9 3JJ, Scotland

Ryanodine Receptor Activation by Ca<sub>V</sub>1.2 Is Involved in Dendritic Cell Major Histocompatibility Complex Class II Surface Expression

M. Vukcevic<sup>1</sup>, G. C. Spagnoli<sup>2</sup>, G. Iezzi<sup>2</sup>, F. Zorzato<sup>3</sup>, and S. Treves<sup>1</sup>

Abstract:

Dendritic cells express the skeletal muscle ryanodine receptor (RyR1), yet little is known concerning its physiological role and activation mechanism. Here we show that dendritic cells also express the Ca<sub>V</sub>1.2 subunit of the L-type Ca<sup>2+</sup> channel and that release of intracellular Ca<sup>2+</sup> via RyR1 depends on the presence of extracellular Ca<sup>2+</sup> and is sensitive to ryanodine and nifedipine. Interestingly, RyR1 activation causes a very rapid increase in expression of major histocompatibility complex II molecules on the surface of dendritic cells, an effect that is also observed upon incubation of mouse BM12 dendritic cells with transgenic T cells whose T cell receptor is specific for the I-Abm12 protein. Based on the present results, we suggest that activation of the RyR1 signaling cascade may be important in the early stages of infection, providing the immune system with a rapid mechanism to initiate an early response, facilitating the presentation of antigens to T cells by dendritic cells before their full maturation.

1 Departments of Anaesthesia and Biomedicine, University Hospital, 4031 Basel, Switzerland
2 Institute of Surgical Research, Basel, University Hospital, 4031 Basel, Switzerland
3 Department of Experimental and Diagnostic Medicine, General Pathology section, University of Ferrara, 44100 Ferrara, Italy
A Novel Role for Embigin to Promote Sprouting of Motor Nerve Terminals at the Neuromuscular Junction

E. Lain, S. Carnejac, P. Escher, M. C. Wilson, T. Lømo, N. Gajendran, and H. R. Brenner

Abstract:
Adult skeletal muscle accepts ectopic innervation by foreign motor axons only after section of its own nerve, suggesting that the formation of new neuromuscular junctions is promoted by muscle denervation. With the aim to identify new proteins involved in neuromuscular junction formation we performed an mRNA differential display on innervated versus denervated adult rat muscles. We identified transcripts encoding embigin, a transmembrane protein of the immunoglobulin superfamly (IgSF) class of cell adhesion molecules to be strongly regulated by the state of innervation. In innervated muscle it is preferentially localized to neuromuscular junctions. Forced overexpression in innervated muscle of a full-length embigin transgene, but not of an embigin fragment lacking the intracellular domain, promotes nerve terminal sprouting and the formation of additional acetylcholine receptor clusters at synaptic sites without affecting terminal Schwann cell number or morphology, and it delays the retraction of terminal sprouts following re-innervation of denervated endplates. Conversely, knockdown of embigin by RNA interference in wild-type muscle accelerates terminal sprout retraction, both by itself and synergistically with deletion of neural cell adhesion molecule. These findings indicate that embigin enhances neural cell adhesion molecule-dependent neuromuscular adhesion and thereby modulates neuromuscular junction formation and plasticity.

Immunoglobulin M deposition in cutaneous nerves of anti-myelin-associated glycoprotein polynueropathy patients correlates with axonal degeneration.


Abstract:
Anti-myelin-associated glycoprotein (MAG) neuropathy is an antibody-mediated polyneuropathy. We correlated clinical features, immunoglobulin (Ig) M blood levels, IgM deposition and axonal degeneration in skin biopsies of anti-MAG neuropathy patients. By confocal microscopy, IgM deposits were found exclusively within perineurium-enclosed nerves; they were not found on single, non-perineurium-ensheathed myelinated axons. There was a linear correlation between IgM accumulation in nerve fascicles with IgM blood levels but not with anti-MAG antibody titer or disease duration. Axons with specific IgM deposits had signs of axonal damage, including neurofilament disintegration. Nodal structures were intact even at sites where the axons showed pathologic changes. Ultrastructural analysis revealed degeneration of myelinating Schwann cells. Taken together, these findings suggest that in anti-MAG neuropathy patients, IgM deposits are entrapped within cutaneous perineurium-ensheathed nerve bundles where they accumulate in the endoneurial space. High local IgM levels in the endoneurium may be required for IgM deposition on myelin and subsequent axonal injury and degeneration. This study underlines the importance of early, effective anti-B-cell treatments for preventing progression of this neuropathy.
Interaction with the hERG channel and cytotoxicity of amiodarone and amiodarone analogues

K. M. Waldhauser1, K. Brecht1, S. Hebeisen2, H. R. Ha1, D. Konrad1, D. Bur4 and S. Krähenbühl1

Abstract:
Background and purpose: Amiodarone (2-n-butyl-3-[3,5 diiodo-4-diethylaminoethoxybenzoyl]-benzofuran, B2-O-CH2CH2-OH) is an effective class III antiarrhythmic drug demonstrating potentially life-threatening organ toxicity. The principal aim of the study was to find amiodarone analogues that retained human ether-a-go-go-related protein (hERG) channel inhibition but with reduced cytotoxicity.

Experimental approach: We synthesized amiodarone analogues with or without a positively ionizable nitrogen in the phenolic side chain. The cytotoxic properties of the compounds were evaluated using HepG2 (a hepatocyte cell line) and A549 cells (a pneumocyte line). Interactions of all compounds with the hERG channel were measured using pharmacological and in silico methods.

Key results: Compared with amiodarone, which displayed only a weak cytotoxicity, the mono- and bis-desethylated metabolites, the further de-graded alcohol (B2-O-CH2-CH2-CH2-CH2-N-CH2-CH2-N-diisopropyl and the B2-O-CH2-CH2-N-disopropyl) and the B2-O-CH2-CH2-N-piperidine were equally or more toxic. Conversely, structural analogues such as the B2-O-CH2-CH2-N-pyrolidine and the B2-O-CH2-CH2-N-pyrolidine were significantly less toxic than amiodarone. Cytotoxicity was associated with a drop in the mitochondrial membrane potential, suggesting mitochondrial involvement. Pharmacological and in silico investigations concerning the interactions of these compounds with the hERG channel revealed that compounds carrying a basic nitrogen in the side chain display a much higher affinity than those lacking such a group. Specifically, B2-O-CH2-CH2-N-pyrolidine and B2-O-CH2-CH2-N-pyrolidine revealed a higher affinity towards hERG channels than amiodarone.

Conclusions and implications: Amiodarone analogues with better hERG channel inhibition and cytotoxicity profiles than the parent compound have been identified, demonstrating that cytotoxicity and hERG channel interaction are mechanistically distinct and separable properties of the compounds.

1 Division of Clinical Pharmacology & Toxicology, Department of Research, University Hospital Basel, Basel, Switzerland
2 Bsys Ltd., Wittenwil, Switzerland
3 Cardiovascular Therapy Research Unit, University Hospital of Zürich, Zürich, Switzerland
4 Department of Drug Discovery Chemistry, Actelion Ltd., Allschwil, Switzerland

Antinucleosome Antibodies as a Marker of Active Proliferative Lupus Nephritis

C. Bigler1, M. Lopez-Trascasa2, E. Potlukova3, S. Moll4, D. Danner3, M. Schaller1 and M. Trendelenburg1,5

Abstract:
Background: Antinucleosome autoantibodies were previously described to be a marker of active lupus nephritis. However, the true prevalence of antinucleosome antibodies at the time of active proliferative lupus nephritis has not been well established. Therefore, the aim of this study is to define the prevalence and diagnostic value of antinucleosomes against nucleosomes as a marker for active proliferative lupus nephritis.

Study Design: Prospective multicenter diagnostic test study.

Setting & Participants: 31 adult patients with systemic lupus erythematosus (SLE) at the time of the renal biopsy showing active class III or IV lupus nephritis compared with 59 control patients with SLE.

Index Test: Levels of antinucleosome antibodies and anti-double-stranded DNA (anti-dsDNA) antibodies.

Reference Test: Kidney biopsy findings of class III or IV lupus nephritis at the time of sampling in a study population versus clinically inactive or no nephritis in a control population.

Results: Increased concentrations of antinucleosome antibodies were found in 31 of 35 patients (89%) with active proliferative lupus nephritis compared with 47 of 59 control patients (80%) with SLE. No significant difference between the 2 groups with regard to number of positive patients ($P = 0.2$) or antibody concentrations ($P = 0.2$) could be found. The area under the receiver operating characteristic curve as a marker of the accuracy of the test in discriminating between proliferative lupus nephritis and inactive/no nephritis in patients with SLE was 0.581 (95% confidence interval, 0.47 to 0.70; $P = 0.2$).

Conclusions: Antinucleosome antibodies have a high prevalence in patients with severe lupus nephritis. However, our data suggest that determining antinucleosome antibodies is of limited help in the distinction of patients with active proliferative lupus nephritis from patients with SLE without active renal disease.

1 Clinical Immunology Laboratory, University Hospital Basel, Basel, Switzerland
2 Department of Immunology, University Hospital La Paz, Madrid, Spain
3 Third Clinic of Medicine, General Faculty Hospital, First Faculty of Medicine, Charles University, Prague, Czech Republic
4 Institute of Pathology, University Hospitals of Geneva and Lausanne, Switzerland
5 Institute of Internal Medicine B, University Hospital Basel, Basel, Switzerland

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DEPARTMENT OF BIOMEDICINE PUBLICATIONS

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High expression of indoleamine 2,3-dioxygenase gene in prostate cancer

C. Feder-Mengus¹, S. Wyler¹, T. Hudolin¹, R. Ruszat¹, L. Bubendorf¹, A. Chiarugi¹, M. Pittelli¹, W. P. Weber¹, A. Bachmann⁵, T. C. Gasser³, T. Sulser³, M. Heberer³, G. C. Spagnoli⁴ and M. Provenzano¹,⁶

Abstract:
Arginase 2, inducible- and endothelial-nitric-oxide synthase (iNOS and eNOS), indoleamine 2,3-dioxygenase (IDO) and TGF-β might impair immune functions in prostate cancer (PCA) patients. However, their expression was not comparatively analysed in PCA and benign prostatic hyperplasia (BPH). We evaluated the expression of these genes in PCA and BPH tissues.

Seventy-six patients (42 BPH, 34 PCA) were enrolled. Arginase 2, eNOS and iNOS gene expression was similar in BPH and PCA tissues. TGF-β₁ gene expression was higher in BPH than in PCA tissues (p = 0.035). IDO gene expression was more frequently detectable (p = 0.00007) and quantitatively higher (p = 0.00001) in PCA tissues than in BPH. IDO protein, expressed in endothelial cells from both BPH and PCA, was detectable in tumour cells in PCA showing evidence of high specific gene expression. In these patients, IDO gene expression correlated with kynurenine/trypophan ratio in sera.

Thus high expression of IDO gene is specifically detectable in PCA.

¹ICFS, Departments of Surgery and Biomedicine, Basel University Hospital, Basel, Switzerland
²Department of Urology, Clinical Hospital Center Zagreb, Croatia
³Department of Pathology, Basel University Hospital, Basel, Switzerland
⁴Department of Preclinical and Clinical Pharmacology, University of Florence, Italy
⁵Department of Urology, Basel University Hospital, Basel, Switzerland
⁶Department of Urology, Zürich University Hospital, Zürich, Switzerland

Comparison of Adhesion and Virulence of Two Predominant Hospital-Acquired Methicillin-Resistant Staphylococcus aureus Clones and Clonal Methicillin-Susceptible S. aureus Isolates

H. Karauzum¹, T. Ferry²,³, S. de Bentzmann⁴ G. Lina²,³, M. Bes²,³, F. Vandenesch²,³, M. Schmaler¹, B. Berger-Bäch¹, J. Etienne²,³ and R. Landmann¹

Abstract:
The virulence of SCCmec type IV hospital-acquired methicillin-resistant Staphylococcus aureus (MRSA) clinical isolates belonging to the major sequence type 8 (ST8 [Lyon clone]) and to a minor upcoming clone, ST5, was compared with that of methicillin-susceptible S. aureus (MSSA) isolates of matching sequence types. In vitro adhesion to human airway epithelial cells (HAECs) as an indicator of dissemination and mortality in a murine sepsis model as an indicator of virulence were evaluated. Ten MRSA isolates and 8 MSSA isolates of ST8 and 8 MRSA isolates and 8 MSSA isolates of ST5 were characterized with respect to multilocus sequence type, agr, spa, and capsule typing; in vitro doubling time; toxin and adhesin gene profiles; and adherence to HAECs. Adherence was significantly lower in the MRSA ST5 group than in the ST8 groups. Infections with MRSA and MSSA isolates ST8 and ST5 were compared. No change in virulence related to the presence of SCCmec was observed, since ST8 but not ST5 caused a significantly lower mortality in its presence. Despite their similar genetic backgrounds, individual clonal MRSA and MSSA isolates were heterogeneous in adherence and virulence. No one of these specific virulence factors determined in vitro was related to mouse mortality. In conclusion, in a bacteremic model, mortality was dependent on the ST and was differentially modulated by SCCmec; within an ST, clonality was not associated with a homogenous outcome.

¹Division of Infection Biology, Department of Biomedicine, University Hospital, Basel, Switzerland
²INSERM, U851, Lyon, F-69008 France
³Université Lyon 1, Centre National de Référence des Staphylocoques, Faculté Laennec, 7 rue Guillaume Paradin, Lyon, F-69008 France
⁴Unité Propre de Recherche 9027, Laboratoire d’ingénierie des systèmes Macromoléculaires, Marseille, France
⁵Institute of Medical Microbiology, University of Zurich, Zurich, Switzerland

1 European Journal of Cancer 44, 2266–2275, 2008 IF 4.5

2 Infection and Immunity 76, 5133–5138, 2008 #4.0
The septin cytoskeleton in myelinating glia

A.M. Buser¹, B. Erne¹, H.B. Werner², K.-A. Nave³ and N. Schaeren-Wiemers¹

Abstract:
Myelin is organized in subdomains with distinct protein and lipid composition. How these domains are established and maintained is currently unknown. Cytoskeletal elements interacting with membrane components could generate and sustain such structural domains. Here, we demonstrate that the transmembrane myelin protein MAL interacts with the cytoskeleton protein septin 6. Septins represent a fourth filamentous system involved in membrane compartmentalization, vesicle transport and scaffold formation. We report that multiple septin complexes are associated with myelin, and that they display an overlapping but non-identical composition in the central and peripheral nervous system. The expression of distinct subsets of septins was upregulated during myelin formation in peripheral nerves and oligodendrocytes. In the PNS, septins were highly enriched in non-compact myelin compartments, particularly in the paranodal loops and the microvilli at the node of Ranvier. Importantly in myelin lacking Septin 6, the abundance of its closest homolog Sept11 was increased, suggesting a functional compensatory role. Our data demonstrate that the septin cytoskeleton is an integral component of the myelin sheath and interacts with distinct myelin constituents such as MAL. We suggest that septins are intriguing candidates for membrane compartmentalization in myelin internodes.

¹Neurobiology, Department of Biomedicine and Neurology, University Hospital Basel, Pharmacenter 7001, Klingelbergstrasse 50-70, CH-4056 Basel, Switzerland
²Department of Neurogenetics, Max-Planck-Institute of Experimental Medicine, D-37075 Göttingen, Germany

Pan-Dengue Virus Detection by PCR for Travelers Returning from the Tropics

A. Dumoulin¹, H. Marti², M. Panning², C. Hatz³ and H. H. Hirsch¹

Abstract:
We performed dengue virus (DENV) serology and quantitative real-time pan-DENV reverse transcription-PCR (RT-PCR) on 186 sera of 171 patients returning from the tropics. DENV loads significantly decreased with increasing times of disease and were higher in immunoglobulin M-negative samples. In the first week of disease, pan-DENV RT-PCR is the test of choice.
Weak effect of metal type and ica genes on staphylococcal infection of titanium and stainless steel implants

D. Hudetz1,2, S. U. Hudetz1, L. G. Harris3, R. Luginbühl4, N. F. Friederich2 and R. Landmann1

Abstract:
Currently, ica is considered to be the major operon responsible for staphylococcal biofilm. The effect of biofilm on susceptibility to staphylococcal infection of different implant materials in vivo is unclear. The interaction of ica-positive (wild-type (WT)) and ica-negative (ica-) Staphylococcus aureus and Staphylococcus epidermidis strains with titanium and both smooth and rough stainless steel surfaces was studied by scanning electron microscopy in vitro and in a mouse tissue cage model during 2 weeks following perioperative or postoperative inoculation in vivo. In vivo, WT S. epidermidis adhered equally and more strongly than did WT S. aureus to all materials. Both WT strains, but not ica strains, showed multilayered biofilm. In vivo, 300 CFUs of WT and ica S. aureus led, in all metal cages, to an infection with a high level of planktonic CFUs and only 0.89% adherent CFUs after 8 days. In contrast, 106 CFUs of the WT and ica strains were required for postoperative infection with S. epidermidis. In all metal types, planktonic numbers of S. epidermidis dropped to <100 WT, and adherent CFUs were low in WT-infected cages and absent in ica-infected cages after 14 days. Perioperative S. epidermidis inoculation resulted in slower clearance than postoperative inoculation, and in titanium cages adherent WT bacteria survived in higher numbers than ica bacteria. In conclusion, the metal played a minor role in susceptibility to and persistence of staphylococcal infection; the presence of ica genes had a strong effect on biofilm in vitro and a weak effect in vivo; and S. epidermidis was more pathogenic when introduced during implantation than after implantation.

Matrix-assisted laser desorption/ionization time-of-flight mass spectrometry for genotyping of human platelet-specific antigens

H.S.P. Garritsen1, A. Xiu-Cheng Fan2, N. Bosse1, H. Hannig1, R. Kelsch1, H. Kroll1, W. Holzgreve2, and X.Y. Zhong2

Abstract:
BACKGROUND: Genotyping of single-nucleotide polymorphisms (SNPs) using matrix-assisted laser desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) is an emerging technique, where finally tools for end users have become available to design primers and analyze SNPs of their own interest. This study investigated the potential of this technique in platelet (PLT) genotyping and developed a validated method for genotyping of clinical relevant human PLT antigens (HPAs).

STUDY DESIGN AND METHODS: A multiplex assay using MALDI-TOF MS to analyze six HPA loci (HPA-1, HPA-2, HPA-3, HPA-4, HPA-5, and HPA-15, respectively) simultaneously in a single reaction was applied for the genotyping of 100 DNA samples from a cohort of plateletpheresis donors and a patient population (n = 20) enriched for rare alleles. The genotyping results using MALDI-TOF MS were validated by the comparison with the results from typing by polymerase chain reaction with sequence-specific primers and conventional DNA sequencing.

RESULTS: Both homozygous and heterozygous genotypes of HPA-1 to -5 and -15 of the 120 individuals were easily identified by a six-plexed assay on MALDI-TOF MS. The three approaches achieved a 100 percent concordance for the genotyping results of the six HPA loci.

CONCLUSION: Compared to conventional methods, the MALDI-TOF MS showed several advantages, such as a high velocity, the ability to perform multiplexed assays in a single reaction, and automated high-throughput analysis of samples. This enables cost-efficient large-scale PLT genotyping for clinical applications.

1 From the Institute for Clinical Transfusion Medicine, Städtisches Klinikum Braunschweig gGmbH, Braunschweig, Germany
2 Laboratory for Prenatal Medicine and Gynecologic Oncology, Women’s Hospital/Department of Research, University of Basel, Basel, Switzerland
3 Institute of Transfusion Medicine and Transplantation Immunology, Westfaelische Wilhelms-University Münster, Germany
4 Institute of Transfusion Medicine Dessau, Red Cross Blood Transfusion Service NISTO, Dessau Germany
Requirements for CD8 T-cell migration into the human arterial wall

J. Gewaltig1,2, M. Kummer1, C. Koella1, G. Cathomas1 and B. C. Biedermann1,2

Abstract:
Atherosclerotic lesions develop in the arterial intima. Among the leukocytes that accumulate in advanced atherosclerotic plaques, CD8 T cells play a quantitatively important role. They may be involved in disease progression and plaque destabilization, leading to plaque rupture or erosion. These events finally precipitate cardiovascular events. Therefore, we wished to determine the accessibility of the human arterial wall, particularly the arterial intima, for CD8-positive, cytotoxic T lymphocytes. We quantified the number of CD8-positive T cells in the arterial wall using human arterial tissue microarrays. The conditions for efficient cytotoxic T-lymphocyte migration into the arterial wall were determined in an in vitro tissue invasion assay. The invasion pattern of resting or activated cytotoxic T-lymphocyte clones was morphometrically analyzed by confocal microscopy. CD8 T cells represented up to 50% of the lymphocytes in advanced atherosclerotic lesions. Resting CD8-positive cytotoxic T lymphocytes were able to migrate into the arterial intima when it was affected by advanced lesions but not at the earliest stages of the disease. After T-cell receptor and/or proinflammatory cytokine activation, cytotoxic T lymphocytes migrated efficiently into the arterial intima, even in the healthy or mildly affected sites. This in vitro tissue invasion assay mimics conditions under which effector cytotoxic T lymphocytes migrate into the arterial wall to reach similar cell densities as observed in arterial tissue sections from autopsies. Interference with T-cell activation may be important to inhibit cytotoxic T-lymphocyte invasion into the unaffected, healthy artery but may not prevent cytotoxic T-lymphocyte invasion into arteries that are severely affected by atherosclerotic lesions.

Oligohis-tags: mechanisms of binding to Ni2+-NTA surfaces

S. Knecht1,2, D. Ricklin1, Alex N. Eberle2 and B. Ernst1

Abstract:
Since immobilized metal ion affinity chromatography (IMAC) was first reported, several modifications have been developed. Among them, Ni2+ immobilized by chelation with nitrilotriacetic acid (NTA) bound to a solid support has become the most common method for the purification of proteins carrying either a C- or N-terminal histidine (His) tag. Despite its broad application in protein purification, only little is known about the binding properties of the His-tag, and therefore almost no thermodynamic and kinetic data are available. In this study, we investigated the binding mechanism of His-tags to Ni2+-NTA. Different series of oligohistidines and mixed oligohistidines/oligoalanines were synthesized using automated solid-phase peptide synthesis (SPPS). Binding to Ni2+-NTA was analyzed both qualitatively and quantitatively with surface plasmon resonance (SPR) using commercially available NTA sensor chips from Biacore. The hexahistidine tag shows an apparent equilibrium dissociation constant (KD) of 14 ± 1 nM and thus the highest affinity of the peptides synthesized in this study. Furthermore, we could demonstrate that two His separated by either one or four residues are the preferred binding motifs within hexahis tag. Finally, elongation of these referred motifs decreased affinity, probably due to increased entropy costs upon binding.
Gene expression analysis of normal appearing brain tissue in an animal model for multiple sclerosis revealed grey matter alterations, but only minor white matter changes

T. Zeis¹, J. Kinter¹, E. Herrero-Herranz¹, R. Weissert¹ and N. Schaeren-Wiemers¹

Abstract:
Multiple sclerosis (MS) is a chronic inflammatory demyelinating disease of the central nervous system (CNS). Recent studies suggest that, beside focal lesions, diffuse inflammatory and degenerative processes take place throughout the MS brain. Especially, molecular alterations in the so-called normal appearing white matter suggest the induction of neuroprotective mechanisms against oxidative stress preserving cellular homeostasis and function. In this study we investigated whether in an animal model for MS, namely in experimental autoimmune encephalomyelitis (EAE), similar changes occur. We isolated normal appearing white and grey matter from the corpus callosum and the above lying cerebral cortex from DA rats with rMOG-induced EAE and carried out a gene expression analysis. Examination of corpus callosum revealed only minor changes in EAE rats. In contrast, we identified a number of gene expression alterations in the cerebral cortex even though morphological and cellular alterations were not evident. One of the most striking observations was the downregulation of genes involved in mitochondrial function as well as a whole set of genes coding for different glutamate receptors. Our data imply that molecular alterations are present in neurons far distant to inflammatory demyelinating lesions. These alterations might reflect degenerative processes induced by lesion-mediated axonal injury in the spinal cord. Our results indicate that the MOG-induced EAE in DA rats is a valuable model to analyze neuronal alterations due to axonal impairment in an acute phase of a MS-like disease, and could be used for development of neuroprotective strategies.

¹ Neurobiology, Department of Biomedicine and Neurology, University Hospital Basel, Pharmacenter, Klingelbergstrasse 50-70, 4056 Basel, Switzerland
² Therapeutic Area Neurodegeneration, Serono Laboratories, 3 Chemin de Mines, 1202 Geneva, Switzerland

Suppression of short interfering RNA-mediated gene silencing by the structural proteins of hepatitis C virus

J. Ji¹, A. Glaser¹, M. Wernli¹, J. M. Berke², D. Moradpour² and P. Erb¹

Abstract:
Viruses have evolved strategies to overcome the antiviral effects of the host at different levels. Besides specific defence mechanisms, the host responds to viral infection via the interferon pathway and also by RNA interference (RNAi). However, several viruses have been identified that suppress RNAi. We addressed the question of whether hepatitis C virus (HCV) suppresses RNAi, using cell lines constitutively expressing green fluorescent protein (GFP) and inducibly expressing HCV proteins. It was found that short interfering RNA-mediated GFP gene silencing was inhibited when the entire HCV polyprotein was expressed. Further studies showed that HCV structural proteins, and in particular envelope protein 2 (E2), were responsible for this inhibition. Co-precipitation assays demonstrated that E2 bound to Argonaute-2 (Ago-2), a member of the RNA-induced silencing complex, RISC. Thus, HCV E2 that interacts with Ago-2 is able to suppress RNAi.

¹ Department of Biomedicine, Institute for Medical Microbiology, University of Basel, Petersplatz 10, CH-4031 Basel, Switzerland
² Division of Gastroenterology and Hepatology, Centre Hospitalier Universitaire Vaudois, University of Lausanne, CH-1011 Lausanne, Switzerland
Pharmacokinetics of intravenous and oral midazolam in plasma and saliva in humans: usefulness of saliva as matrix for CYP3A phenotyping

B. Link, M. Haschke, N. Grignaschi, M. Bodmer, Y. Zysset Aschmann, M. Wenk and S. Krähenbühl

Abstract:

Aims: To compare midazolam kinetics between plasma and saliva and to find out whether saliva is suitable for CYP3A phenotyping.

Methods: This was a two way cross-over study in eight subjects treated with 2 mg midazolam IV or 7.5 mg orally under basal conditions and after CYP3A induction with rifampicin.

Results: Under basal conditions and IV administration, midazolam and 1'-hydroxymidazolam (plasma, saliva), 4-hydroxymidazolam and 1'-hydroxymidazolam-glucuronide (plasma) were detectable. After rifampicin, the AUC of midazolam (mean differences plasma 53.7 (95% CI 4.6, 102.9) ng ml⁻¹ h⁻¹ and saliva 0.83 (95% CI 0.52, 1.14) ng ml⁻¹ h⁻¹) and 1'-hydroxymidazolam (mean difference plasma 11.8 (95% CI 7.9, 15.7) ng ml⁻¹ h⁻¹) had decreased significantly. There was a significant correlation between the midazolam concentrations in plasma and saliva (basal conditions: r = 0.864, P < 0.0001; after rifampicin: r = 0.842, P < 0.0001). After oral administration and basal conditions, midazolam, 1'-hydroxymidazolam and 4-hydroxymidazolam were detectable in plasma and saliva. After treatment with rifampicin, the AUC of midazolam (mean difference plasma 104.5 (95% CI 74.1, 134.9) ng ml⁻¹ h⁻¹ and 1'-hydroxymidazolam (mean differences plasma 51.9 (95% CI 34.8, 69.1) and saliva 2.3 (95% CI 1.9, 2.7) ng ml⁻¹ h⁻¹) had decreased significantly. The parameters separating best between basal conditions and post-rifampicin were: (1'-hydroxymidazolam + 1'-hydroxymidazolam-glucuronide)/midazolam at 20-30 min (plasma) and the AUC of midazolam (plasma) and of 1'-hydroxymidazolam (plasma and saliva) after oral administration.

Conclusions: Saliva appears to be a suitable matrix for non-invasive CYP3A phenotyping using midazolam as a probe drug, but sensitive analytical methods are required.

Whole blood assessment of antigen specific cellular immune response by real time quantitative PCR: a versatile monitoring and discovery tool

E. Schultz-Thater¹, D. M. Frey¹, D. Margelli², N. Raafat¹, C. Feder-Mengus¹, G.C. Spagnoli¹ and P. Zajac¹

Abstract:

Background: Monitoring of cellular immune responses is indispensable in a number of clinical research areas, including microbiology, virology, oncology and autoimmunity. Purification and culture of peripheral blood mononuclear cells and rapid access to specialized equipment are usually required. We developed a whole blood (WB) technique monitoring antigen specific cellular immune response in vaccinated or naturally sensitized individuals.

Methods: WB (300 µl) was incubated at 37°C with specific antigens, in the form of peptides or commercial vaccines for 5–16 hours. Following RNAlater addition to stabilize RNA, the mixture could be stored over one week at room temperature or at 4°C. Total RNA was then extracted, reverse transcribed and amplified in quantitative real-time PCR (qRT-PCR) assays with primers and probes specific for cytokine and/or chemokine genes.

Results: Spiking experiments demonstrated that this technique could detect antigen specific cytokine gene expression from 50 cytotoxic T lymphocytes (CTL) diluted in 300 µl WB. Furthermore, the high sensitivity of this method could be confirmed ex-vivo by the successful detection of CDB+ T cell responses against HCMV, EBV and influenza virus derived HLA-A201 restricted epitopes, which was significantly correlated with specific multimer staining. Importantly, a highly significant (p = 0.000009) correlation between hepatitis B surface antigen (HBsAg) stimulated IL-2 gene expression, as detectable in WB, and specific antibody titers was observed in donors vaccinated against hepatitis B virus (HBV) between six months and twenty years before the tests. To identify additional markers of potential clinical relevance, expression of chemokine genes was also evaluated. Indeed, HBsAg stimulated expression of MIP-1β (CCL3) gene was highly significantly (p = 0.0006) correlated with specific antibody titers. Moreover, a longitudinal study on response to influenza vaccine demonstrated a significant increase of antigen specific IFN-γ gene expression two weeks after immunization, declining thereafter, whereas increased IL-2 gene expression was still detectable four months after vaccination.

Conclusion: This method, easily amenable to automation, might qualify as technology of choice for high throughput screening of immune responses to large panels of antigens from cohorts of donors. Although analysis of cytokine gene expression requires adequate laboratory infrastructure, initial antigen stimulation and storage of test probes can be performed with minimal equipment and time requirements. This might prove important in „field“ studies with difficult access to laboratory facilities.

¹Institute of Surgical Research and Hospital Management, Dept. of Biomedicine, University Hospital of Basel, Basel, Switzerland
²Personnel Medical Service, University Hospital of Basel, Basel, Switzerland
Computational evaluation of oxygen and shear stress distributions in 3D perfusion culture systems: Macro-scale and micro-structured models

M. Cioffi1,2, J. Küffer3, S. Ströbel2, G. Dubini1, I. Martin2 and D. Wendt2

Abstract:
We present a combined macro-scale/micro-scale computational approach to quantify oxygen transport and flow-mediated shear stress to human chondrocytes cultured in three-dimensional scaffolds in a perfusion bioreactor system. A macro-scale model was developed to assess the influence of the bioreactor design and to identify the proper boundary conditions for the micro-scale model. The micro-scale model based on a micro-computed tomography (µCT) reconstruction of a poly(ethylene glycol terephthalate)/poly(butylene terephthalate) (PEGT/PBT) foam scaffold, was developed to assess the influence of the scaffold micro-architecture on local shear stress and oxygen levels within the scaffold pores. Experiments were performed to derive specific oxygen consumption rates for constructs perfused under flow rates of 0.3 and 0.03 ml min\(^{-1}\). While macro-scale and micro-scale models predicted similar average oxygen levels at different depths within the scaffold, µCT models revealed small local oxygen variations within the scaffold micro-architecture. The combined macro-scale/micro-scale approach indicated that 0.3 ml min\(^{-1}\), which subjected 95% of the cells to less than 6.3 mPa shear, would maintain the oxygen supply throughout the scaffold above anoxic levels (>1%), with 99.5% of the scaffold supplied with 8–2% O\(_2\). Alternatively, at 0.03 ml min\(^{-1}\), the macro-scale model predicted 6% of the cells would be supplied with 0.5–1% O\(_2\), although this region of cells was confined to the periphery of the scaffold. Together with local variations predicted by the micro-scale model, the simulations underline that in the current model system, reducing the flow below 0.03 ml min\(^{-1}\) would likely have dire consequences on cell viability to pronounced regions within the engineered construct. The presented approach provides a sensitive tool to aid efficient bioreactor optimization and scaffold design.

1 Laboratory of Biological Structure Mechanics, Department of Structural Engineering, Politecnico di Milano, Italy
2 Department of Surgery and of Research, University Hospital Basel, Switzerland
3 IRCCS Instituto Ortopedico Galeazzi, Milano, Italy
4 Institute for Product and Production Engineering, University of Applied Sciences Northwestern Switzerland, Switzerland

Journal of Biomechanics

Effects of fluid flow and calcium phosphate coating on human bone marrow stromal cells cultured in a defined 2D model system.

S. Scaglione1, D. Wendt1, S. Miggino1, A. Papadimitropoulos1,2, M. Fato1, R. Quarto2, I. Martin1

Abstract:
In this study, we investigated the effect of the long-term (10 days) application of a defined and uniform level of fluid flow (uniform shear stress of 1.2 \times 10^{-3} \text{N/m}^2) on human bone marrow stromal cells (BMSC) cultured on different substrates (i.e., uncoated glass or calcium phosphate coated glass, Osteologic\(^\text{TM}\)) in a 2D parallel plate model. Both exposure to flow and culture on Osteologic significantly reduced the number of cell doublings. BMSC cultured under flow were more intensely stained for collagen type I and by von Kossa for mineralized matrix. BMSC exposed to flow displayed an increased osteogenic commitment (i.e., higher mRNA expression of cbfa-1 and osterix), although phenotype changes in response to flow (i.e., mRNA expression of osteopontin, osteocalcin and bone sialoprotein) were dependent on the substrate used. These findings highlight the importance of the combination of physical forces and culture substrate to determine the functional state of differentiating osteoblastic cells. The results obtained using a simple and controlled 2D model system may help to interpret the long-term effects of BMSC culture under perfusion within 3D porous scaffolds, where multiple experimental variables cannot be easily studied independently, and shear stresses cannot be precisely computed.

1 Laboratory of Biological Structure Mechanics, Department of Structural Engineering, Politecnico di Milano, Italy
2 Department of Surgery and of Research, University Hospital Basel, Switzerland
3 IRCCS Instituto Ortopedico Galeazzi, Milano, Italy
4 Institute for Product and Production Engineering, University of Applied Sciences Northwestern Switzerland, Switzerland
Dreizack-Brunnen «Spittelsprung-Brunnen»

Ecke Münsterberg / Freie Strasse

Dissertationen


Linda Kenins von der Forschungsgruppe Experimental Hematology (Departement Biomedizin USB) hatte ihre Dissertationsprüfung am 1. Dezember 2008. Sie definierte in ihrer Dissertation «The Role of Flt3 ligand in immune reconstitution».

Am 10. Dezember 2008 haben Michel Mallaun von der Forschungsgruppe Transplantation Immunology (Departement Biomedizin USB) und Balasubramanian Sivasankaran von der Forschungsgruppe Neurooncology (Departement Biomedizin USB) ihre Dissertationen erfolgreich abgeschlossen. Der Titel der Doktorarbeit von Michel lautete: «Proximal TCR signaling in self tolerance», Bala beschäftigte sich mit «The role of NOTCH2 gene in Human malignant glial brain tumours».

bietet «Immunomodulation by Ectosomes» und Davide be- schäftigte sich mit dem Thema: «Identification of a Novel Population of Bone Marrow-Derived Prominin-1/CD133+ Lung Progenitors with Regenerative Capacity».


Ebenfalls im Dezember 2008 hat Adrian Egli von der Forschungsgruppe Transplantation Virology (Institut für Medizinische Mikrobiologie) seine Doktorarbeit abgeschlossen. Der Titel seiner Dissertation lautete: «In vitro and in vivo characterization of the Cytomegalovirus and Polyomavirus BK specific immune response».

Marco Lepore von der Forschungsgruppe Experimental Immunology (Departement Biomedizin USB) hat am 29. April 2009 seine Doktorandenzzeit erfolgreich beendet. Er hat sich in seiner Dissertation mit dem Thema: “CD1-

Restricted Autoreactive T Cell Response: Characterization in Humans and Implication for Leukemia Targeting” beschäftigt.


Ting Liu von der Forschungsgruppe Childhood Leukemia (Departement Biomedizin USB) stellte sich am 11. Mai 2009 mit Erfolg den Fragen des Dissertationskomitees. Ihr beschäftigte das Thema "Identification of cooperating genetic events in acute leukemia".


Seit dem 27. Mai 2009 darf sich auch Svenja Landweer von der Forschungsgruppe Neural-immune Interactions (Institut für Physiologie) Frau Dr. nennen. Der Titel ihrer Doktorarbeit lautete: "Role of neurotrophins and neuropeptides in Genetic Absence Epilepsy Rats from Strasbourg (GAERS), a model for human generalized absence seizures".

Herzlichen Glückwunsch an alle!
Habilitationen

**Venia legendi für Henryk Zulewski**
Dr. med. Henryk Zulewski von der Forschungsgruppe Metabolism (Departement Biomedizin USB) hat von der Medizinischen Fakultät die Venia legendi für Innere Medizin und Endokrinologie erhalten. Der Titel seiner Habilitationsvorlesung vom 6. März 2009 lautete: „Potential of human adult stem cells to acquire a pancreatic endocrine phenotype“.

**Venia legendi für Thomas Dieterle**
Dr. med. Thomas Dieterle von der Forschungsgruppe Cardiobiology (Departement Biomedizin USB) hat sich im Dezember 2008 an der Medizinischen Fakultät habilitiert und die Venia legendi für Kardiologie erhalten.

Beförderungen

**Titularprofessur für Thomas Klimkait**
PD Dr. Thomas Klimkait von der Forschungsgruppe Research in Translational Medicine (Institut für Mikrobiologie) ist vom Universitätsrat zum Titularprofessor im Bereich Virologie an der Medizinischen Fakultät ernannt worden.

**SNF-Förderprofessuren für Mirjam Christ-Crain und Alfred Zippelius**
PD Dr. Mirjam Christ-Crain von der Forschungsgruppe Metabolism (Departement Biomedizin USB) hat eine SNF-Förderprofessur erhalten. Ebenfalls über eine SNF-Förderprofessur darf sich PD Dr. Alfred Zippelius von der Forschungsgruppe Medical Oncology (Departement Biomedizin USB) freuen.

**Christoph Hess Ordinarius für Ambulante Innere Medizin**
Prof. Christoph Hess von der Forschungsgruppe Immunobiology (Departement Biomedizin USB) ist zum Ordinarius für Ambulante Innere Medizin an der Medizinischen Fakultät und Chefarzt der Medizinischen Poliklinik am USB ernannt worden.

**Markus Heim Extraordinarius für Hepatologie**
Prof. Markus Heim von der Forschungsgruppe Hepatology (Departement Biomedizin USB) ist zum Extraordinarius für Hepatologie ernannt worden. Gleichzeitig wird Markus Heim Stellvertretender Chefarzt für Gastroenterologie am USB.

**Thomas Szucs Extraordinarius für Pharmazeutische Medizin**
Prof. Thomas Szucs, Co-Direktor am Europäischen Zentrum für Pharmazeutische Medizin (ECPM), ist zum Extraordinarius für Pharmazeutische Medizin ernannt worden. Ausserdem ist Prof. Szucs mit seinem MBA-Abschluss als Gesundheitsökonom am Institut für Sozial- und Präventivmedizin der Universität Zürich tätig.

**Alex N. Eberle wird Vizerektor**
Regine Landmann wird Vizedekanin Nachwuchsförderung


Albert Urwyler und Gerhard Christofori bestätigt


Auszeichnungen

David Leuenberger und Simone Salzmann erhalten Fakultätsauszeichnung


Preise

Fakultätspreis an Simone Ehrhard

Am Dies Academicus 2008 ist Simone Ehrhard für ihre Doktorarbeit „Immune Response in Lymph Notes of HIV-1 infected Individuals under Antiretroviral Therapy“, die sie an der Klinik für Infektiologie und Spitalhygiene und in der Forschungsgruppe Experimental Immunology (Institut für Mikrobiologie) durchgeführt hat, mit dem Fakultätspreis der Medizinischen Fakultät der Universität Basel ausgezeichnet worden. Der Preis wurde von der Roche Research Foundation gestiftet, das Preisgeld beträgt 10’000 CHF.

Fakultätspreis für höchsten Impactfactor 2008 an Aleksandra Wodnar-Filipowicz

DBM-Preis an Linda Kenins

Posterpreis an Bettina Burger

Poster-Award an Nadine Hardel
Am BioValley Science Day 2008 hat Nadine Hardel von der Forschungsgruppe Synapse Formation (Institut für Physiologie) den Poster-Award Kategorie Silber erhalten. Der Preis geht an Erstautor/innen, wurde von Roche und Actelion gestiftet, die Preissumme beträgt 1’000 CHF.

Best Poster Preis an Ulrike Hopfer

Pfizer Forschungspreis 2009 an Rainer Gosert und Magdalena Sarasin-Filipowicz

Preis der Schweizerischen Gesellschaft für Infektiologie an Nina Khanna

Hematologic Malignancies Award 2008 an Radek Skoda
Radek Skoda von der Forschungsgruppe Experimental Hematology (Departement Biomedizin USB) hat für seine Studie „Ratio of mutant JAK2-V617F to wild-type Jak2 determines the MPD phenotypes in transgenic mice“ den mit 100’000 CHF dotierten Hematologic Malignancies Award 2008 erhalten. Erstautor der Studie ist Ralph Tiedt. Die Firma Bristol-Myers Squibb Switzerland stiftet den Award alle zwei Jahre zur Förderung Schweizer Spitzenforschung auf dem Gebiet der Hämato-Onkologie. Der Award wurde 2008 zum ersten Mal vergeben.

Herzliche Gratulation!

DBM Facts 1|2009 Departement Biomedizin
Marcin Maj
Perioperative Patient Safety

Guido Flum
Animal Facility

Georg Funk
Immunobiology

Kseniya Maslova
Signaling

Olivier Gasser
Immunobiology

Oliver Gordon
Infection Biology

Mark Melnyk
Leitung Dienste DBM

Réjane Morand
Clinical Pharmacology

Annette Orleth
Medical Oncology

Dennis Pfaff
Signaling

Valentina Mele
Oncology Surgery

Raphael Thurnheer
Perioperative Patient Safety

Ludwig Villiger
Cardiobiology

Verena Widmer
Immunonephrology

Fayaz Ahmad Mir
Exp. Immunology
Ausserdem haben angefangen:

DEPARTEMENT BIOMEDIZIN USB

Oliver Härschnitz, Tissue Engineering
Chong Teck S’ng, Pneumology
Maria Luz Bellido Diaz, Prenatal Medicine
Yvonne Achermann, Infectious Diseases
Oliver Boss, Tissue Engineering
Martin Clauss, Infectious Diseases
Helena Lima, Tissue Engineering
Anja Zahno, Clinical Pharmacology
Seeta Ramanjaneyulu Gundimeda, Exp. Immunology
Cédric Hysek, Clinical Pharmacology

INSTITUT FÜR MEDIZINISCHE MIKRO-BIOLGIE

Piotr Kardas, Transplantation Virology
Zorica Slavnic, Reinigungsdienst
Marielle Bieri, Molecular Diagnostics
Simone Edelmann, Molecular Diagnostics
Jolanda Kamber, Molecular Diagnostics

INSTITUT FÜR ANATOMIE

Murat Bilici, Musculoskeletal Research Group

INSTITUT FÜR BIOCHEMIE UND GENETIK

Matthias Kreuzaler, Developmental and Molecular Immunology
Nadine Gehre, Developmental and Molecular Immunology

INSTITUT FÜR PHYSIOLOGIE

Rostislav Turecek, Synaptic Plasticity

Mark Melnyk neuer Leiter Dienste


Radek Skoda
3. DBM Badminton Open

Friday, Sept 25 2009, 19:30 – 24:00

Where: Badminton Halle Oberwil (www.badminton-halle.ch)

Costs: 30.– per person incl. one sandwich and one drink.

Category: Women-, Men-Doubles and Mixed play in one category

Matches: 4–5 matches for each team

Registration: 11. Sept 2009
per e-mail to Christian.Kalberer@unibas.ch
(Fee have to be paid at the time of registration)

There is a limit of 24 teams for this event (= 48 people).

I am looking forward to a fun evening!

Christian Kalberer, Lab 310, Exp. Hematology.

BADMINTONHALLE
OBERWIL
Congratulations

Das DBM gratuliert ganz herzlich!

Ivan
Christoph
Radimerski
Geboren am 27.9.2008

Paola Lucille
Schreiner-King
Geboren am 8.10.2008

Philipp Joaquim
Ritschard
Geboren am 28.8.2008
Anna Josephine Duss-Oser
Geboren am 27.2.2009

Sarah Anina Bachmann
Geboren am 24.3.2009

Herzlich willkommen, allerseits!

Manuela Satu Dubler
Geboren am 23.4.2009

Julian David Mild-Schneider
Geboren am 25.3.2008
Prof. Christoph Moroni,

Vorsteher des Institutes für Medizinische Mikrobiologie (IMM) der Medizinischen Fakultät der Universität Basel, ist Ende 2008 in den wohlverdienten Ruhestand gegangen, oder doch nicht?

Man nimmt die Nachricht der Emeritierung von Prof. Moroni mit Überraschung auf, ist man doch gewohnt, dass er in Forschung, Lehre und akademischer Selbstverwaltung immer überdurchschnittlich engagiert und präsent ist. Die erste Reaktion gilt der Lücke, die schwer zu füllen sein wird. In Wien, einer meiner früheren Arbeitsorte, wird erzählt, dass man tot sein muss, um eine verdiente Ehrung zu erhalten. Dies ist im Fall von Christoph Moroni sicherlich nicht der Fall. Die Liste seiner Leistungen in Forschung, Lehre und akademischem Milizdienst ist eindrucksvoll und scheint noch lange nicht abgeschlossen. Ich beschränke mich daher auf einen «Progress Report».


In Zeiten der AIDS Epidemie hat das Institut als erstes diagnostisches Labor die PCR-Methode für die molekulare Diagnostik von HIV und TB angewandt. Heute bietet das Institut neben der herausragenden Forschung und dem breiten Spektrum in der Lehre eine Vielzahl an diagnostischen Analysen, die ein nicht unbedeutendes
Einkommen für die Universität ausmachen. In diesen Jahren des Aufbaus und der Konsolidierung hat Prof. Moroni weiterhin erfolgreich geforscht und hervorragende Arbeiten über die Funktion von Retroviren und Onkogenen und später über die Regulation von mRNA-Stabilität durchgeführt, alles Arbeiten, die zum grundlegenden Verständnis der Regulation von Onkogenen in physiologischen Prozessen und in der Krebsentstehung beigetragen haben. Viele seiner ehemaligen Doktoranden, Postdoktoranden und wissenschaftlichen Mitarbeiter sind inzwischen Professoren der Universität Basel und anderswo, eine Liste zu lang, um hier aufgeführt zu werden.

Trotz der erreichten Ziele hat es sich Prof. Moroni nicht gemütlich eingerichtet. Im Gegenteil, von 1994 bis 1995 war er Dekan der Medizinischen Fakultät und von 2000-2006 hat er die Herausforderung angenommen, die klinische und die vorläufige Forschung in einem Department zu vereinen und zu führen, dem damaligen Departement für Klinisch-Biologische Wissenschaften (DKBW), aus dem unser heutiges Departement für Biomedizin (DBM) hervorgegangen ist. Diese administrativen Anstrengungen boten ihm auch kreative Spielräume, so die Neuorientierung der vorläufigen Institute im Rahmen von fakultären Schwerpunkten, eine Reihe wichtiger Neuerungen, sowie die Akquirierung und Renovierung des Gebäudes an der Mattenstrasse für das junge Department. So nebenbei war Prof. Moroni auch Mitglied mehrerer wichtiger Gremien, wie zum Beispiel Forschungsrat des Schweizerischen Nationalfonds, Mitglied des Swiss Boards of Consultants des Basel Institute of Immunology, Mitgründer und Verwaltungsrat der Firma InPheno und Mitglied des Universitätsrates der Medizinischen Universität Graz.


Liebe Christoph, wir hoffen, Deine Pläne gehen wie gewünscht in Erfüllung und Du bleibst uns noch lange als Forscherkollege erhalten. Wir wünschen Dir alles Gute für den neuen Lebensabschnitt.

Gerhard Christofori

Danke Christoph Moroni: Radek Škoda überreicht seinem Vorgänger im Amt des DBM-Leiters ein Geschenk zum Abschied.
A Quantum of Solace

The building was not at all what I had expected. In the corner of a park-like square with high beech trees, opposite to the marble-coloured University building, there was a two-story timbered house with medieval-style crown glass windows. At first sight, its somewhat armoured appearance was repelling, but the sign clearly indicated that this was “The Institute of Medical Microbiology of the University of Basel”. There seemed to be some heavy construction going on and, with all doors open, I went inside and asked directions to the office of the Professor. It was a bit murky there, and in the air, there was a damp smell reminiscent of the past century of microbiology. I became uncertain whether or not this could really be the place for any future training in molecular and medical sciences. After a short wait with the secretary, the Professor welcomed me and we sat down. Although my past research on yeast proteases had no overlap with his interests in viral transformation or cancer, the Professor seemed at ease with the aspects of cell biology. He explained some aspects of his current research on ras – oncogene and the role of posttranscriptional control of gene expression. He surprised me with his engaging optimism, not only concerning the research, but surprisingly enough, also regarding the modernisation of the Institute. He indicated that there might be the position of an assistant doctor in diagnostics in the next couple of months. Three weeks later, I received a letter stating that I could start in December 1988.

It is now almost exactly 20 years since this episode and sure enough, the Professor, Dr. med. Christoph Moroni, Ordinarius for Medical Microbiology of the University of Basel, has not only managed to transform the historic “Stachelschützenhaus am Petersplatz 10” in Basel into a modern and generously equipped research and diagnostic laboratory called, in short, the “IMM” but he has also generated a personally attractive place for people wanting to work in research and in diagnostics, in one of the nicest locations in the midst of historic Basel. Indeed, going through the Annual Reports of the “IMM” is impressive as it reads a bit like a Who-is-Who of Basel research. Although not primarily involved in routine microbiology, Christoph recognized early on the tremendous potential of the Polymerase Chain Reaction for diagnostic virology and HIV-AIDS. Thus, in 1988, with Hans-Peter Senn, the “IMM” became the first laboratory, other than Geneva, to introduce diagnostic PCR. Together with Professors Peter Erb and Kurt Bienz, Christoph brought together dedicated individuals in research, diagnostics, teaching, administration, and technical staff, side-by-side, all under one roof.
While the scientific success of the “IMM” is well documented by the large list of publications in internationally top-ranked journals such as Nature, Embo, Cell, PNAS, Lancet and the New England Journal of Medicine, the high degree of personal satisfaction becomes obvious in the long-term commitment of many of the more than 50 “IMM” employees. In fact, some have been at the “IMM” for more than 15 years, and others returned after enlightening experiences elsewhere. This positive spirit is remarkable and well portrayed by photographs of the “IMM” annual excursions which typically combined physical activity, cultural exposure, and culinary wellness, fostering casual outside work interactions. Of course, as in every other institution, things did not always just go smoothly. Some vexing trouble would strike, synergizing with disabling back aches that required complex remedies with rocking chairs of futuristic design. In exceptional cases messages came flying like rocks through windows, but altogether, most issues resolved without further repercussions. Clearly, Christoph as “Vorsteher des Instituts” has always been an easily accessible sparring partner for the people around him, whether this was for problems - or for visions.

Christoph also had a tremendous impact as the Head of the former Department Klinische & Biologische Wissenschaften DKBW (today called Department Biomedicine), where he proved to be an efficient, stimulating and innovative science administrator. He was instrumental in attracting and then effectively triaging the appointments of a brilliant new generation of research leaders, with new research facilities at the Mattenstrasse, where the Institute of Biochemistry and Genetics is now located. All of this had signalling effects beyond the borders of the Petersplatz. Christoph also midwifed the “IMM” spin-off company InPheno. Among others honours, Christoph served as the Dean of the Medical Faculty of the University of Basel, was in the advisory board of the Basel Institute of Immunology, godfathered BIG, the Basel Immunology Group meeting of clinicians and basic researchers, functioned as “Forschungsrat” of the Swiss National Science Foundation, and is still in Universitätsrat of the University of Graz, Austria, to name just a few. And it is probably not spilling the beans by suggesting that Christoph’s Mary has been a key to his success. Impressively, Christoph has maintained his major virtues, namely a youthful, optimistic attitude, a great sense of innovation, and scientific curiosity which continues to synergize with good humour, inspiring the people around him. Now, Christoph has been asked to “retire”, but with a quantum of solace. As Guest Professor in the Bio-Pharmazentrum of the University of Basel, he and his team will further explore their most recent research success on mTOR mutants and their pharmacological agonists and antagonist. Thus, in the name of all former and present colleagues and team members, it is my pleasure and honour to congratulate Christoph on his “retirement plans” and express our gratitude for the two creative decades at the “IMM”.

Hans Hirsch

2008 with his wife Mary
“DBM – IT“

Eine gute “IT“ ist eine, die man eigentlich gar nicht wahrnimmt – so die Sicht des «Users». Trotzdem, oder vielleicht gerade deswegen, möchten wir, die IT-Verantwortlichen der einzelnen Häuser und Institute des DBM, uns hier kurz zu Wort melden. Dies um bekannt zu machen, dass wir die immer komplexer werdenden Aufgaben der DBM Informatik in Zukunft nicht mehr als Einzelkämpfer, sondern gemeinsam als Team anpacken möchten.


Seit November 2008 finden nun im Zweimonatsrhythmus IT-Treffen statt, an welchen die IT-Verantwortlichen die Möglichkeit wahrnehmen, sich und ihre Institute in einem informellen Rahmen vorzustellen. Ein erster Nutzen ergab sich sehr schnell durch die Erkenntnis, dass unter den Beteiligten ein sehr breites Fachwissen vorhanden ist, welches durch eine einzelne Person nicht abgedeckt werden könnte. Dadurch, dass die Beteiligten sich nun persönlich kennen gelernt haben, konnten kurze Wege geschaffen werden, über die im Bedarfsfall schnell kompetente Hilfe bei einer Kollegin oder einem Kollegen abgeholt werden kann. Darüber hinaus konnten IT-Bedürfnisse eruiert werden, deren Realisierung ein einzelnes Institut überfordern, welche jedoch im Rahmen des DBM gelöst werden können.

Das Team kurz vorgestellt:

**Daniel Fröhlich**

**Jose Girau**
Claudia Haupt

Claude A. Levy

Reto Schaub

Ismerai Steiner-Lobo

Primo Schär
Viele von Euch werden mich kennen – oder vielleicht doch nicht?


Ashwini and I decided to get married last monsoon in an Indian traditional style which dates back to a 5000 years old system, and which is still performed by chanting mantras in Sanskrit language. According to Vedic civilization, marriage is the ritual to enter Grahasthram (2nd phase of life devoted for family life). As it is traditional for the Hindu family to look for an auspicious date, our parents, upon consulting astrologers, decided our wedding should be on 8th July, the Hindu month of Ashada Shukla Panchami (fifth day after new moon day). Once the date was decided, both families geared up for the series of ceremonies.
The rituals started with worshipping Lord Ganesh (elephant God) installed in a Mantapa (a construction of coconut and mango leaves) in front of the house and prayers were made to inhibit obstacles. The bride and groom were showered with holy water containing turmeric, sandalwood paste, and perfumes.

On the evening of the second day we left for the marriage hall where Ashwini and her family members were ready to welcome us. I was escorted to the marriage hall on a decorated horse, in a procession with music, dance, and crackers, in which my friends and family actively participated. We received a warm welcome from the bride’s parents and relatives, who applied tilak (vermilion) on our foreheads. The next ritual was “Milap”, where family members of both sides greeted each other with flowers and coconuts. Later, a delicious dinner was served on banana leaves. The first dinner is usually devoted to the groom’s mother. The night was fun-filled with music and dance and preparations for main occasion of the wedding the next day. Decorators were busy decorating marriage hall with colourful flowers and mango leaves.
The wedding started on the third day at Brahmi mahurat (5 AM) when the bride carries out Gauri Pooja (offering prayers and flowers to the Goddess Gauri). Later, the bride and groom were separately brought to the northeast part of hall, each accompanied by their parents. (We were not allowed to see each other until this time!!). Everyone was given Akshata, mixture of raw rice, vermillion and turmeric (A-not, kshata-perishable). Priests started chanting “Mangalastaka” (8 divine blessings) ending with kuryat sada mangalam exactly when Sun entered the zodiac sign of cancer on July 8th, and once these chants had ended, everyone showered us with Akshata, representing their imperishable blessings to us for our happy married life! Nadaswaram (an instrument blown during auspicious occasions) added more divinity to the ceremony. It was followed by Hasta Milap and Kanyadaan, where my in-laws placed Ashwini’s palm on my palm (denoting handing her over to me and to share our lives together) in presence of various Gods and rivers. On the day of the wedding the bride is considered to be as the Goddess Lakshmi and groom as the Lord Vishnu (celestial couple). Later we exchanged garlands.

The next ritual was Mangalasutra Dharaña. Mangalasutra is a necklace made of black and golden beads. The groom ties this necklace to bride for her happy life and promises that he will protect her all during his life. The groom and his mother apply sindhoor (vermilion) on bride’s forehead and gift a variety of jewellery, including bangles. Later we did Saptapadi, where we made seven rounds of the sacred Agni (fire) vowing to share love, duty, respect, happiness and sorrow together and be companions forever.

Guests were served with a grand lunch, consisting of various dishes and we had nearly 1000 guests wishing us “Kuryat Sada Mangalam”.

Manjunath B. Joshi
In der nächsten Ausgabe . . .

... erfahren wir mehr über die Forschungsaktivitäten im Labor Neurooncology

... nimmt uns Andreas Jehle mit auf Entdeckungstour in die Welt der Molecular Nephrology

... stellt sich die Uni-Personalabteilung vor

... berichtet Anna Marsano von ihrem Forschungsaufenthalt in New York

... erleben wir den Advent im USB mit Pfarrer Jürg Merz
Dämmern liegt der Sommerabend...
Dämmern liegt der Sommerabend
Über Wald und grünen Wiesen;
Goldner Mond, im blauen Himmel,
Strahlt herunter, duftig labend.

An dem Bache zirpt die Grille,
Und es regt sich in dem Wasser,
Und der Wanderer hört ein Plätschern
Und ein Atmen in der Stille.

Dorten an dem Bach alleine,
Badet sich die schöne Elfe;
Arm und Nacken, weiß und lieblich,
Schimmern in dem Mondenschein.

Heinrich Heine (1797-1856)