DBM FACTS

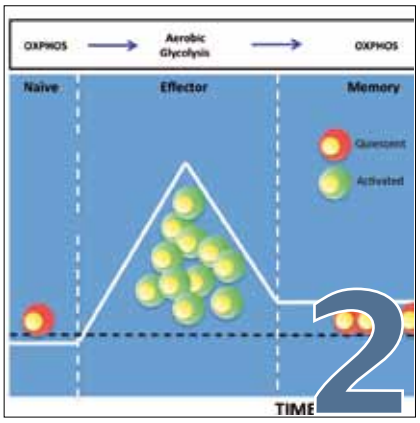
Periodisches Informationsblatt des Departementes Biomedizin
Universität Basel, Universitätsspital Basel und
Universitäts-Kinderspital beider Basel

T-cell metabolism and functionality | Adult neurogenesis in the hippocampus: New players in long-term memory circuit function | Heimat

1 | 13

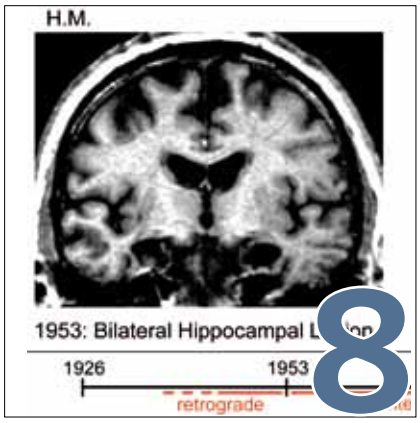
INHALT CONTENTS

Departement Biomedizin Basel



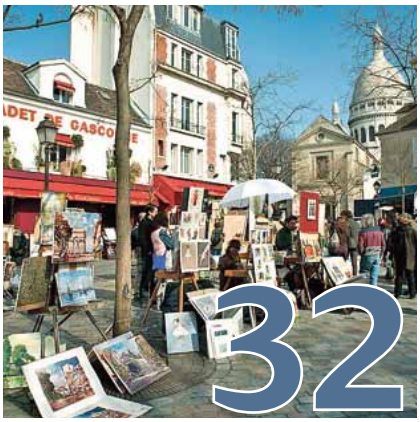
T-cell metabolism and functionality
from Christoph Hess

Impressionen vom Research Day



Adult neurogenesis in the hippocampus: New players in long-term memory circuit function
from Josef Bischofberger

PhD Club – Scientific Winter Retreat



Have a Parisian day
from Elise Dalmas

IMPRESSUM

Redaktion
Heidi Hoyermann

Übersetzungen
Paula Cullen

Layout
Eric Spaety, Morf Bimo Print AG, Binningen

IT-Unterstützung
Niklaus Vogt

Administration
Manuela Bernasconi

Fotos
Mathias Mangold www.sublim.ch
Frank Neumann

Titelfoto
Wooden fencing in village with yellow flowers on it (Ukraine)

Druck
Morf Bimo Print AG, Binningen

Anschrift
Redaktion DBM Facts
Departement Biomedizin
Hebelstrasse 20
4031 Basel
heidi.hoyermann@usb.ch

UKBB Universitäts-Kinderspital beider Basel

Universitätsspital Basel

UNI BASEL

EDITORIAL

Radek Skoda
Leiter DBM

Liebe Leserinnen und Leser

Endlich ist er da, der lang erwartete Frühling. Und mit ihm die neueste Ausgabe der DBM-Facts. Es gibt Gutes zu berichten: Der Research Day war sehr gut besucht und insgesamt ein grosser Erfolg. Nun gilt es, die neuesten Empfehlungen des Scientific Advisory Boards aufzunehmen und umzusetzen (Seite 7).
Die Medizinische Bibliothek ist in die Räumlichkeiten an der Spiegelgasse umgezogen und hat bereits ihren Betrieb wieder aufgenommen. Der Umbau im 2. Stock zu neuen Labors für das DBM kann nun beginnen.
Sven Cichon hat die Professur «Medizinische Genetik» angetreten und die Leitung der Einheit übernommen. Die Medizinische Genetik ist per Januar 2013 vom UKBB an das USB überführt worden. Alfred Zippelius wurde zum Professor für «Translationale Onkologie» an der Medizinischen Fakultät gewählt und zum stv. Chefarzt der Medizinischen Onkologie am USB befördert. Das DBM hat auch zwei neue SNF-Professoren: Mike Recher und Lukas Jeker werden am DBM ihre eigenen Forschungsgruppen gründen. Herzliche Gratulation an Alle und viel Erfolg!
In der vorliegenden Ausgabe erhalten Sie (ab Seite 18) einen Überblick über die neuesten Publikationen des DBM. Christoph Hess und seine Gruppe führen Sie unter dem Titel «T-cell metabolism and functionality» in das Gebiet der «Immunobiology» ein (ab Seite 2) und wir erfahren von Josef Bischofberger mehr über die neuesten Ergebnisse in seiner Forschungsgruppe «Cellular Neurophysiology» (ab Seite 8).
Wo DBM Mitarbeitende sich zu Hause fühlen, Heimat in einer globalisierten Welt finden, lesen Sie ab Seite 30 bis 33. Dass Fussball eine Wissenschaft für sich sein kann, dann ab Seite 34.

Eine spannende Lektüre und geniessen Sie den Frühling!

Dear Readers

The long awaited spring is finally here, and with it comes the latest edition of DBM Facts. There is good news: the Research Day was well attended and was a great success. We are now going to discuss and implement the latest recommendations of the Scientific Advisory Board (page 7). The medical library has moved premises to Spiegelgasse and has already reopened. This now allows constructing laboratory space for the DBM on the 2nd floor.
Sven Cichon has been appointed professor of Medical Genetics and has taken over leadership of the unit.
In January 2013, Medical Genetics has been transferred from the UKBB to the USB. Alfred Zippelius has been chosen as Professor for Translational Oncology by the Medical Faculty and promoted to deputy Head of Medical Oncology at the USB. We also have two new SNF-Professors: Mike Recher and Lukas Jeker will start their own research groups at the DBM. Congratulations to all and best wishes!
In this issue starting on page 18 you will get an overview of the latest publications from the DBM. Christoph Hess and his group will bring you on a journey into the field of "Immunobiology" with their article "T-cell metabolism and functionality" (page 2) and from Josef Bischofberger we learn more about the latest developments in his research group "Cellular Neurophysiology" (page 8).
Between pages 30 and 33 you can find out where DBM workers feel at home and how it is defining home in a globalised world. Then on page 34 you can discover that football (soccer) can be a science of its own.

I hope you all enjoy spring, and I wish you an enjoyable read.

T-cell metabolism and functionality

General Summary

Metabolic pathway usage dictates cellular responsiveness. Our general interest is to assess how the metabolic repertoire of various human lymphocyte subsets defines their functionality. Specifically we aim to characterize metabolic pathway usage and regulation among resting naïve and antigen-experienced CD8+ and CD4+ T cell subsets.

Group picture at CERN/Geneva (from left): Marco Fischer, Anne-Valérie Burgener, Annaïse Jauch, Sarah Dimeloe, Gideon Hönger, Glenn Bantug, Christoph Hess, Patrick Gubser, Christoph Berger, Bojana Durovic, Leyla Razik (Matthias Mehling and Mike Recher were still approaching the speed of light in one of the CERN-accelerators when we took this picture).

Adaptive cellular immunity

During acute infection, pathogen-specific naïve T-cells become activated, followed by their rapid clonal expansion and differentiation into effector cells. Resolution of infection triggers the contraction of effector cells, which is accompanied by formation of a long-lived memory pool (FIGURE 1). Memory T-cells provide a source of quiescent, antigen-experienced cells with high proliferative capacity and a lower activation threshold than naïve cells. These traits allow for a rapid generation of effector cells during secondary infection. The molecular basis defining T-cell memory remains an important unresolved issue in immunology.

CD8+ T-cell metabolism

Naïve CD8+ T-cells are metabolically quiescent and primarily depend on oxidative phosphorylation (OXPHOS) as their energy source (FIGURE 1). Ligation of T-cell receptors (TCR) and co-stimulatory molecules (e.g. CD28) on naïve CD8+ T-cells initiates dramatic changes in cellular metabolic pathway usage. A hallmark of this metabolic remodeling is the upregulation of aerobic glycolysis (Warburg effect), which is a prerequisite for growth, expansion and acquisition of effector function (FIGURE 1). Augmented glycolysis among effector cells is attributable to enhanced glucose uptake and increased expression/activity of glycolytic enzymes, while glucose utilization via OXPHOS is decreased. This 'metabolic switch' satiates higher energy demands ***and*** provides biochemical intermediates utilized in the biosynthesis of macromolecules. Accordingly, aerobic glycolysis is essential for efficacious CD8+ T-cell activation, and blockade of this key metabolic pathway results in the diminution of CD8+ T-cell clonal expansion and maturation. In addition to the Warburg effect, CD8+ T-cell activation has been demonstrated to induce mitochondrial membrane hyper-polarization and to enhance mitochondrial respiration. Inhibition of mitochondrial respiration attenuates CD8+ T-cell responses following activation. During the effector-to-memory transition, a reverse 'metabolic switch' occurs, where aerobic glycolysis is down modulated, again making OXPHOS the primary energy-generating pathway in the cell (FIGURE 1).

Metabolic repertoires of naïve and effector memory CD8+ T-cells

In a coordinated process, distinct subsets of memory CD8+ T-cells enhance host protection upon secondary infection. Immunologic memory relies on memory T-cells that are able to rapidly acquire effector function and re-expand as secondary effector cells. In comparison, naïve CD8+ T-cells acquire effector function only after several rounds of clonal expansion. These important qualitative differences between naïve and memory CD8+ T-cells are potentially linked to imprinted meta-

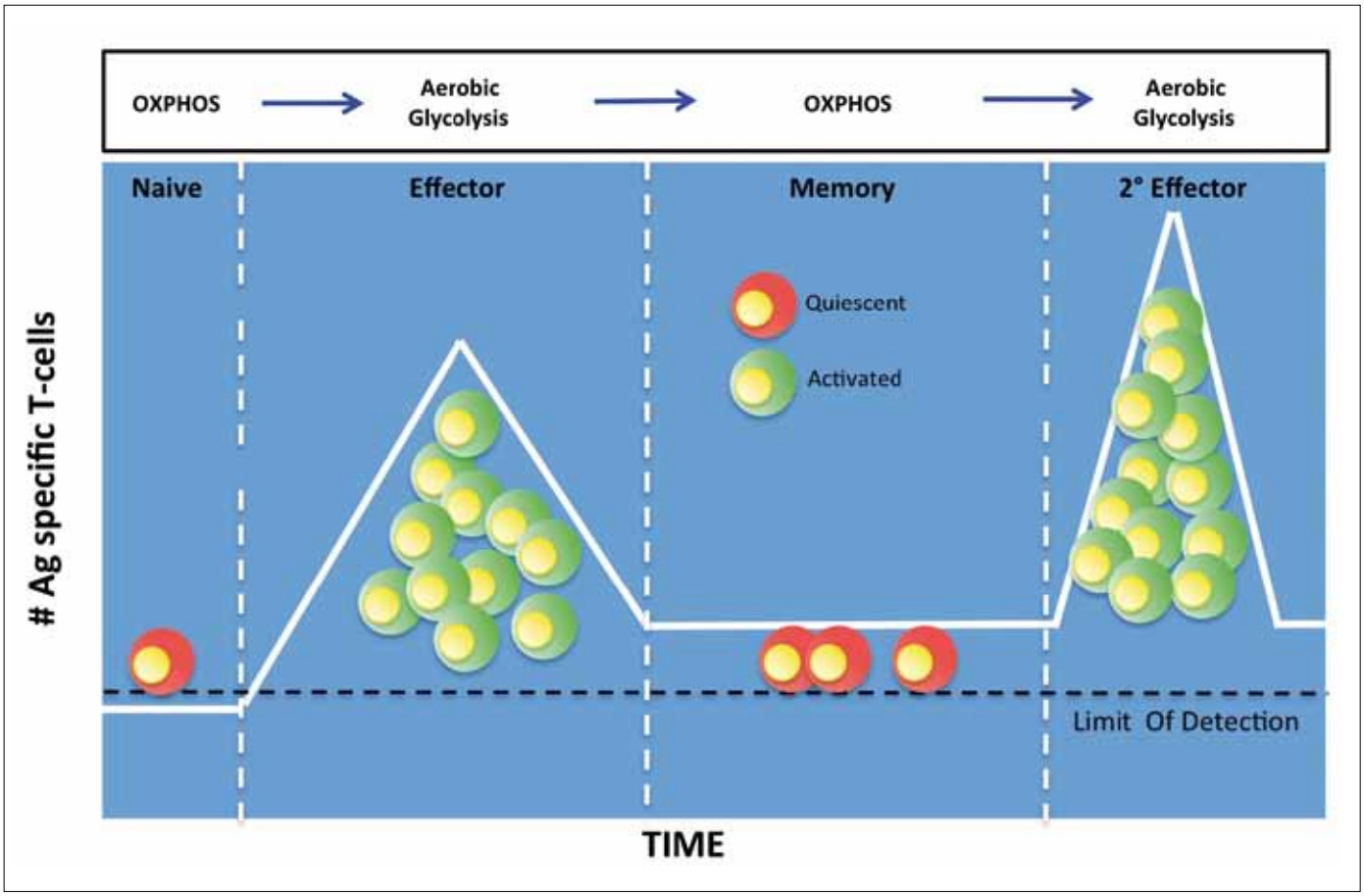


FIGURE 1. General schematic diagram of an antigen-specific T-cell response during primary and secondary infection, and its relation with metabolic pathway usage.

bolic features in memory cells that support their effector maturation.

Effector-memory (EM) CD8+ T-cells are specialized antigen-experienced lymphocytes that traffic between blood and non-lymphoid tissues, whereas, central memory (CM) CD8+ T-cells traffic to lymph nodes. EM CD8+ T-cells are ideally positioned to rapidly respond and execute effector functions at sites of infection. Similar to naïve CD8+ T-cells, the clonal re-expansion of memory cells also requires aerobic glycolysis in order to meet added biosynthetic and energetic demands. The metabolic requirements of EM CD8+ T-cells that rapidly acquire effector function are unknown. Under basal conditions, EM CD8+ T-cells have traditionally been characterized as metabolically quiescent, similar to their naïve counterparts. However, our group has recently determined that naïve and EM CD8+ T-cell subsets possess distinct mitochondrial mass/morphology (FIGURE 2). Moreover, spare respiratory capacity and the glycolytic potential seems to be greater in EM CD8+ T-cells. Against this background, we currently investigate the impact of these imprinted *metabolic repertoires* on the functionality of each CD8+ T-cell population.

CD4+ T-cell metabolism

CD4+ T-cells are central coordinators of the immune response. Aiming to define the metabolic repertoire of human CD4+ T-cells we currently focus on three phenotypic/functional subsets, namely naïve non-regulatory T-cells, EM non-regulatory T-cells, and regulatory T-cells (Treg). Treg constitute 5-10% of the peripheral blood CD4+ T-cell pool and control innate and adaptive immune cell-activity through a diverse array of mechanisms – thus preventing inappropriate or excessive immune responses. Our aim is to assess whether metabolic pathway usage in human Treg is distinct from that of non-Treg CD4+ T-cell subsets, which in turn would per-

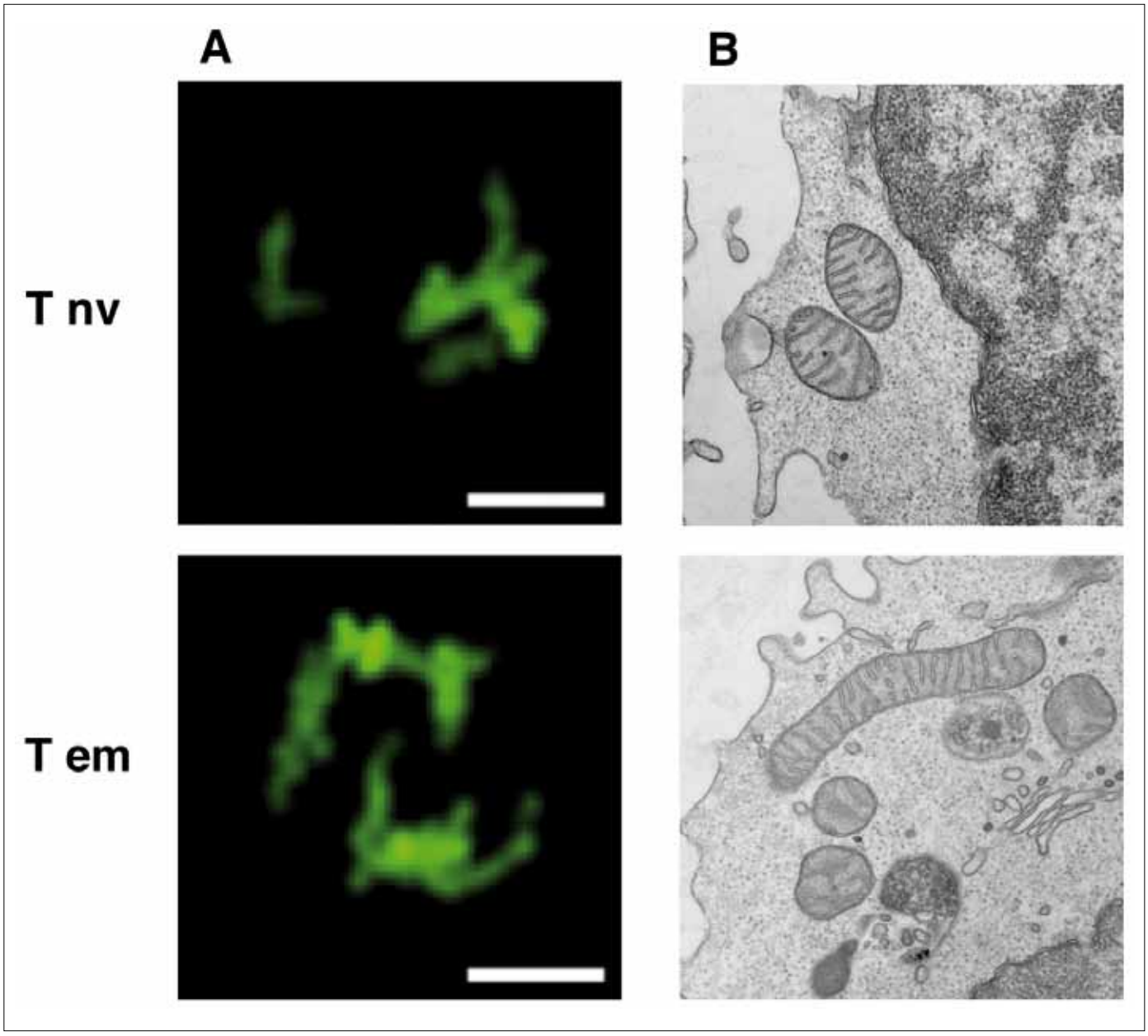


FIGURE 2. Distinct mitochondrial morphology of naïve versus EM CD8+ T-cells.
(A) Live-cell confocal images of mitotracker green-loaded naïve (top; T nv) and EM (bottom; T em) CD8+ T-cells. Magnification = x100, scale bars = 3 µm.
(B) Transmission electron photomicrographs of naïve (top) and EM (bottom) CD8+ T-cells. Magnification = x28'000.

mit to selectively target Treg vs. non-Treg metabolism to impact defined disease situations. In preliminary experiments we observed that EM non-Treg consistently demonstrate higher mitochondrial content, protein expression and functionality than their naïve counterparts, confirming in the CD4+ T-cell compartment what we have already observed in CD8+ T-cells -i.e. that mitochondrial biogenesis occurs during T-cell memory formation. Using functional readouts, the OXPHOS capacity of Treg was the poorest of the three CD4+ T-cell subsets assessed. Testing the capacity of CD4+ T-cell subsets to upregulate aerobic glycolysis upon *in vitro* activation, we observed that both naïve and EM non-Treg subsets, but not Treg, sustainably upregulate aerobic glycolysis. This finding is in keeping with work performed in murine *in vitro* derived cells, agrees with general observations that Treg proliferate poorly *in vitro,* and may suggest a means to target effector CD4+ T-cell activation without affecting Treg functionality.

T-cell migration

As in the differentiation of T-cells, metabolic changes play a central role in defining migration and homing characteristics of distinct T-cell subsets. Homing of T-cells is a highly regulated process mediated by an orchestrated interplay of chemokines/chemokine receptors and adhesion molecules. Interaction of 'peripheral node addressins' (PNAd) with L-selectin on T-cells allows tethering/rolling along high endothelial venules (HEV). Interaction of the chemokine receptor CCR7 with its ligands CCL19/21, and CXCR4 with CXCL12, then mediates firm adhesion to HEV via high-affinity interactions of LFA-1 and ICAM-1, permitting transmigration of T-cells across the HEV cell layer. Within the lymphnode (LN), T-cell migration is directed via T-cell zones towards the cortical sinuses. A sphingosine 1-phosphate (S1P) gradient established across the endothelial cells of the cortical sinuses is directing LN egress of T-cells via efferent lymph back to the peripheral blood circulation. Acting as a functional antagonist on the S1P receptor (S1PR), the pharmacological compound fingolimod – which has shown efficacy in the treatment of multiple sclerosis (MS) – blocks this egress. As a consequence, in fingolimod-treated individuals naïve and CM T-cells are trapped in LN and reduced in the blood circulation. By studying – in close collaboration with the Neurology Department of our Hospital – depletion kinetics of T-cells in the blood of *de novo* fingolimod exposed individuals, we found that CD4+ T-cells diminish earlier than CD8+ T-cells after the first dose of fingolimod. This suggests that CD4+ T-cells enter lymphoid tissue – and thus access dendritic cells – more frequently than CD8+ T-cells. More pronounced, even, were the differences in calculated homing frequencies between phenotypic naïve and antigen-experienced T-cell subsets: naïve T-cells home 2–3x as often to LN as CM T-cells. In line with a higher recirculation-frequency *in vivo,* naïve CD4+ and CD8+ T-cells also *in vitro* migrate more efficiently in gradients of the LN-homing chemokines CCL19/21 and CXCL12, whereas migration towards S1P was comparable in CD4+ and CD8+ T-cells and the respective subsets. We are now following up these findings by further studying migration – also in the context of metabolic characteristics – in different T-cell subsets.

Christoph Hess and team

References

Maciver, N. J., Michalek, R. D. & Rathmell, J. C. (2013). *Metabolic Regulation of T Lymphocytes.* Annu. Rev. Immunology.

Pearce, E. L. et al. (2009). *Enhancing CD8 T-cell memory by modulating fatty acid metabolism.* Nature, 460, 103–107.

Haring, J. S., Badovinac, V. P. & Harty, J. T. (2006). *Inflaming the CD8+ T cell response.* Immunity, 25, 19–29.

Geginat, J., Lanzavecchia, A. & Sallusto, F. (2003). *Proliferation and differentiation potential of human CD8+ memory T-cell subsets in response to antigen or homeostatic cytokines.* Blood, 101, 4260–4266.

Michalek, R. D., Gerriets, V. A., Jacobs, S. R., Macintyre, A. N., MacIver, N. J., Mason, E. F., et al. (2011). *Cutting edge: distinct glycolytic and lipid oxidative metabolic programs are essential for effector and regulatory CD4+ T cell subsets.* J. Immunol., 186, 3299–3303.

Michalek, R. D., Gerriets, V. A., Nichols, A. G., Inoue, M., Kazmin, D., Chang, C.-Y., et al. (2011). *Estrogen-related receptor-α is a metabolic regulator of effector T-cell activation and differentiation.* PNAS, 108, 18348–18353.

Delgoffe, G. M., Kole, T. P., Zheng, Y., Zarek, P. E., Matthews, K. L., Xiao, B., et al. (2009). *The mTOR kinase differentially regulates effector and regulatory T cell lineage commitment.* Immunity, 30, 832–844.

Haxhinasto, S., Mathis, D., & Benoist, C. (2008). *The AKT-mTOR axis regulates de novo differentiation of CD4+Foxp3+ cells.* JEM, 205, 565–574.

Andrian, von, U.H., and Mempel, T.R. (2003). *Homing and cellular traffic in lymph nodes.* Nat. Rev. Immunol., 3, 867–878.

Matloubian, M., Lo, C.G., Cinamon, G., Lesneski, M.J., Xu, Y., Brinkmann, V., Allende, M.L., Proia, R.L., and Cyster, J.G. (2004). *Lymphocyte egress from thymus and peripheral lymphoid organs is dependent on S1P receptor 1.* Nature 427, 355–360.

Mehling, M., Brinkmann, V., Antel, J., Bar-Or, A., Goebels, N., Vedrine, C., Kristofic, C., Kuhle, J., Lindberg, R.L.P., and Kappos, L. (2008). *FTY720 therapy exerts differential effects on T cell subsets in multiple sclerosis.* Neurology, 71, 1261–1267.

Mehling, M., Brinkmann, V., Burgener, A.-V., Gubser, P., Luster, A.D., Kappos, L., and Hess, C. (2013). *Homing frequency of human T cells inferred from peripheral blood depletion kinetics after sphingosine-1-phosphate receptor blockade.* JACI.

Miyasaka, M., and Tanaka, T. (2004). *Lymphocyte trafficking across high endothelial venules: dogmas and enigmas.* Nat. Rev. Immunol., 4, 360–370.

DBM Research Day

Höchste Aufmerksamkeit bei Hans Hirsch …

… Matthias Wymann …

… und Peter Itin am Vormittag des 24.01.2013 am Rednerpult im kleinen Hörsaal ZLF …

… wer nicht vorne steht, hört aufmerksam zu …

… Vieles ist zu diskutieren ..

… Adrian Duek und Bob Löwenberg hören gar nicht mehr auf …

… wer nicht aufs Bild muss, hat gut lachen …

… aber wenn man so freundlich angestrahlt wird … da kann das Foto nur gelingen … Das DBM Advisory Board (von links nach rechts): Prof. Paolo Bianco, Prof. Margaret Frame, Prof. Bob Löwenberg, Prof. Greg Lemke, Prof. Kathryn Wood, Prof. Christian Lüscher, Prof. Brigitta Stockinger und Prof. Karl-Heinz Krause. Fotos: Mathias Mangold und Frank Neumann

Adult neurogenesis in the hippocampus: New players in long-term memory circuit function

Research group from left to right: Mirko Vukcevic, Michael Barz, Liyi Li, Stefanie Heigele, Heidi Ramstein, Jörg Pohle, Selma Becherer, Jan Schulz, Josef Bischofberger. (Missing on photo: Martine Schwager)

Summary

The hippocampal formation within the medial temporal lobe of the cerebral cortex is essential for our conscious memory of facts and events. On the cellular level, learning of new memory items adjusts communication between hippocampal neurons via synaptic plasticity, a major focus of our research. Remarkably, the hippocampus is one of the very few regions in the central nervous system of adult mammals, including humans, where new neurons are continuously generated throughout life. This indicates that the new neurons are involved in learning and formation of new memories. In support of this hypothesis, we have found that newly generated young neurons show enhanced excitability and synaptic plasticity as compared to the neighboring mature cells. Understanding the mechanisms underlying adult neurogenesis and the specific contribution of young and mature cells to hippocampal information processing is the major goal of our current research.

Introduction

Why do we need long-term memory? Since early human history people try to predict the future. By detecting temporal correlations humans try to understand 'why' a specific event happens. This kind of knowledge is obviously to our advantage in order to predict when a certain event may occur and helps to avoid unwelcome surprises. Hence, we try to correlate the occurrence of distinct sequential events, although we know that the accuracy of the prediction is sometimes small. Even the Swiss National Science Foundation forces us to predict the social and economic outcome of research grant applications before we have even started with the experiments! Of course, it is not possible to predict the future. However, the ability to learn from the past presents a big evolutionary advantage. A powerful long-term memory system developed during mammalian evolution within the medial temporal lobe of the cerebral cortex. It enables us to 'learn' from the past and to recognize frequently repeating sequences of events. Memory recall of correlated events allows us to estimate the likelihood that something may happen or not, helping us to decide what to do next.

The medial temporal lobe system

The investigation of the long-term memory system in the medial temporal lobe of the cortex started in the 1950's with the famous patient Henry Molaison (H.M, Scoville and Milner 1957). He experienced a bicycle accident as a 9-year old boy leading to the development of epileptic seizures over the time course of the following years. The epilepsy could not be treated pharmacologically. The major seizure attacks increased in frequency over the years and he was finally unable to work. Thus,

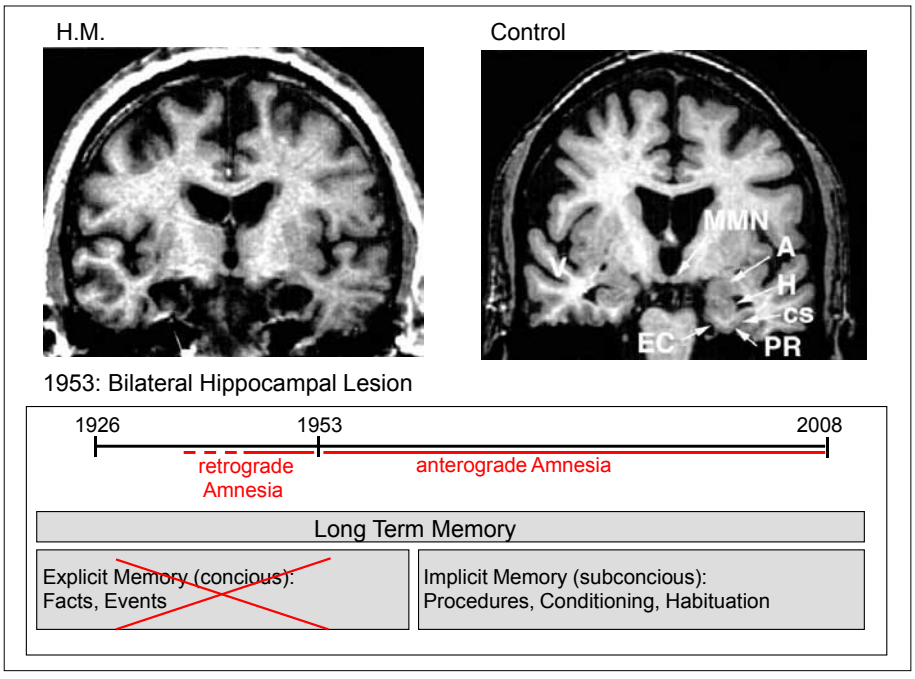


Figure 1
Damage of hippocampus results in memory loss. NMR images from the patient H.M. and an age-matched control person showing the lesioned hippocampus (H) and entorhinal cortex (EC, from Corkin et al. 1997). The time table below indicates the year of birth, surgery and death of patient H.M. as well as his long-term memory deficit.

at the age of 27 it was decided to bilaterally remove part of the entorhinal cortex and the hippocampus within the medial temporal lobe (Figure 1), because electroencephalographic studies had already located focal epileptic activity in this brain region in other patients.

The outcome of the surgery was remarkable. The frequency of seizures was substantially reduced. In addition however, a severe loss of memory for facts and events was recognized. H.M could not remember why he was in hospital or anything that had happened during his half-year hospital stay. He had a complete loss of memory for events happening about 2 years before surgery (retrograde amnesia) and was unable to learn or remember any new information once his attention moved away from a topic (anterograde amnesia). As a consequence, he had to stay at a nursing home for the rest of his life.

Although he lost the ability for explicit (conscious) memory formation and retrieval, many implicit (subconscious) memory functions were intact. For example procedural learning, i.e. the learning of new motor functions like riding a bicycle, was relatively normal. Based on these and related observations, it was concluded that the hippocampal region is essential for learning and memory.

The hippocampal circuit and memory formation

Although we are still far away from understanding the underlying computational processes, some features of the hippocampal neuronal circuit that allow the association of individual memory items in space and time have been identified. The major route of information flow is formed by the so called trisynaptic pathway (Figure 2). The granule cells in the dentate gyrus receive neuronal information from the superficial layers of the entorhinal cortex (1st synapse), which itself is reciprocally connected to all important neocortical association areas. The granule cells project to the CA3 pyramidal cells (2nd synapse), which in turn project to the CA1 region (3rd synapse). The CA1 pyramidal cells project back to the deep layers of the entorhinal cortex, either directly or indirectly via the subiculum. Besides the trisynaptic pathway, there are multiple short cuts with entorhinal fibers directly projecting to CA3 and CA1. All described synapses use glutamate, the major excitatory neurotransmitter of the brain.

Research on the involvement of hippocampal synapses in learning and memory formation was greatly facilitated with the advent of transgenic animal models with specific circuit deficits. Explicit memory formation can be tested in rodents by using animal-relevant memory tasks, e.g. finding the location of an escape platform within a spatial maze. Similar to humans, these learning capabilities are lost after bilateral damage of the hippocampus.

How can hippocampal synapses become adjusted in a meaningful way? The currently available data indicate that synaptic plasticity, regulating synaptic strength and growth, is induced by sequential neuronal activity. One of the major sequence detectors at excitatory synapses

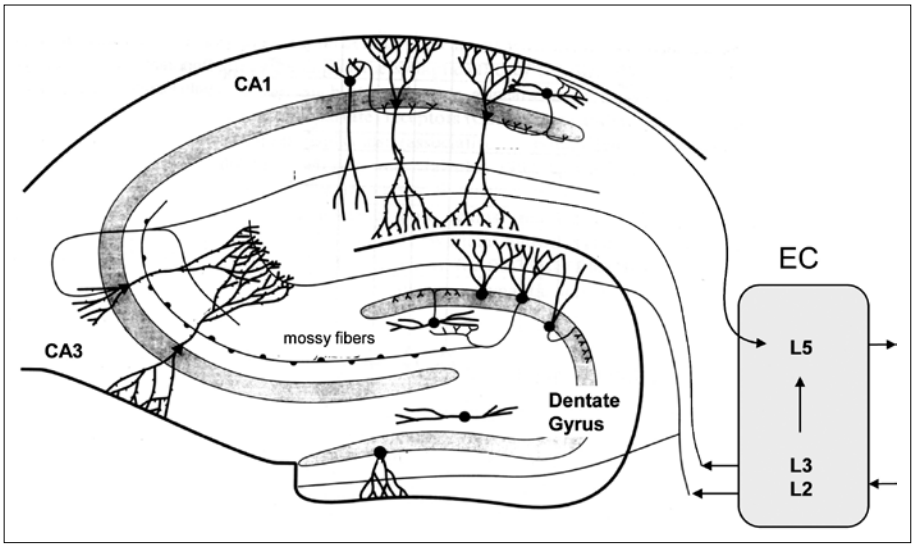


Figure 2
Synaptic organization of the hippocampal circuit (from Bischofberger et al. 2006b).

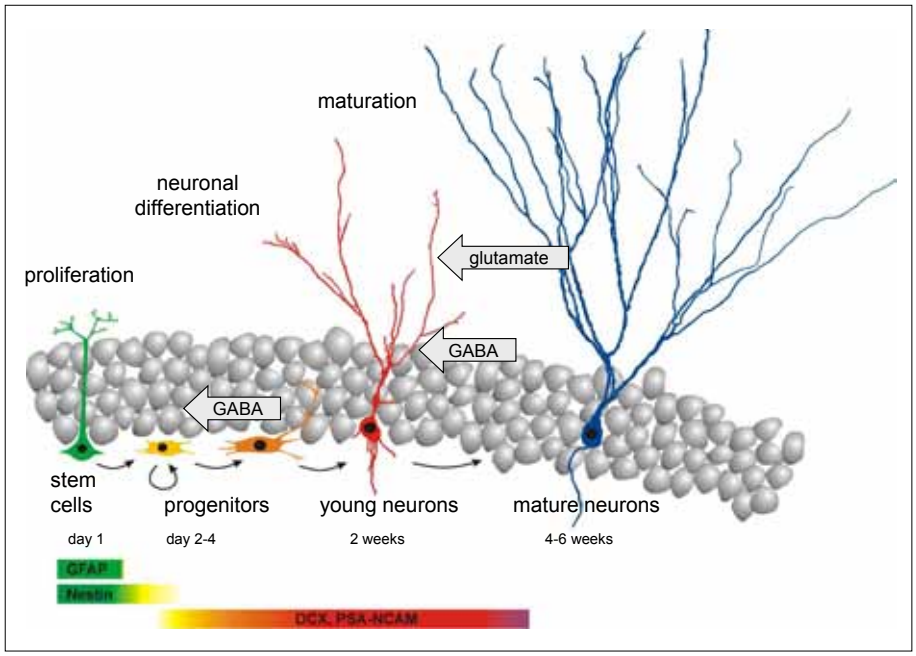


Figure 3
Adult neurogenesis in the hippocampus. The young neurons express specific proteins like DCX (doublecortin) and PSA-NCAM (polysialylated neural cell adhesion molecule) which can be used for identification of the young cells within the adult brain (from Bischofberger 2007).

is the NMDA-type glutamate receptor that is most active when an action potential in a postsynaptic neuron B repetitively follows presynaptic glutamate release from an ensemble of synchronously active presynaptic neurons (A1, A2, A3,.....) and not otherwise. NMDA-receptors are highly Ca^{2+}-permeable inducing Ca^{2+}-dependent growth of synapses. Therefore, temporal correlations in the neuronal input patterns, evoked by sensory input or by intrinsic neocortical computations, are well suited to shape hippocampal synaptic communication.

After spatial learning, pyramidal cells show action potential firing selective at a specific location within the maze, the so-called place field. Sequences of place field firing can be consistently reactivated not only during spatial navigation but also during post-training periods important for memory consolidation (Davidson et al. 2009). Importantly, blocking synaptic plasticity by genetic deletion of NMDA-type glutamate receptors in CA1 or CA3 pyramidal cells, not only disturbed synaptic plasticity and place field firing, but also spatial memory formation as well as sequence learning (Huerta et al. 2000, Nakazawa et al. 2004). The excitatory recurrent network in CA3 appears to be specifically important for the rapid formation of auto-associative cell assemblies, which allow memory retrieval after the presentation of partially incomplete cues, a function that is called 'pattern completion'. Taken together, current research shows that temporal correlations in neuronal input patterns shape hippocampal connectivity during learning. After learning, a partial cue reactivates not only a small neuronal subset but also correlated neuronal activity in large cell assemblies. Activity in these hippocampal cell assemblies may in turn reactivate, via back projection to the entorhinal cortex, cell assemblies in the neocortex which code for the various details associated with the memory trace.

What is the dentate gyrus good for?

When I started as a young postdoc in the lab of Peter Jonas at the University of Freiburg in 1996, the function of the dentate gyrus was basically unknown. Interestingly, dentate gyrus granule cells constitute the largest population of hippocampal principal cells. In the human hippocampus, there are about 20 million granule cells compared to ~3 million CA3 and ~15 million CA1 pyramidal cells (West et al. 1994). Furthermore, granule cells are the only type of neurons in the mammalian cortex that are not only generated during development but also in the adult nervous system throughout life. Finally, they are known to have relatively large presynaptic terminals (Ø ≈ 2–5 μm), the so called mossy fiber boutons forming large synapses with CA3 pyramidal cells. Back then, we decided to employ a bottom-up approach starting with the functional analysis of these boutons. After im-

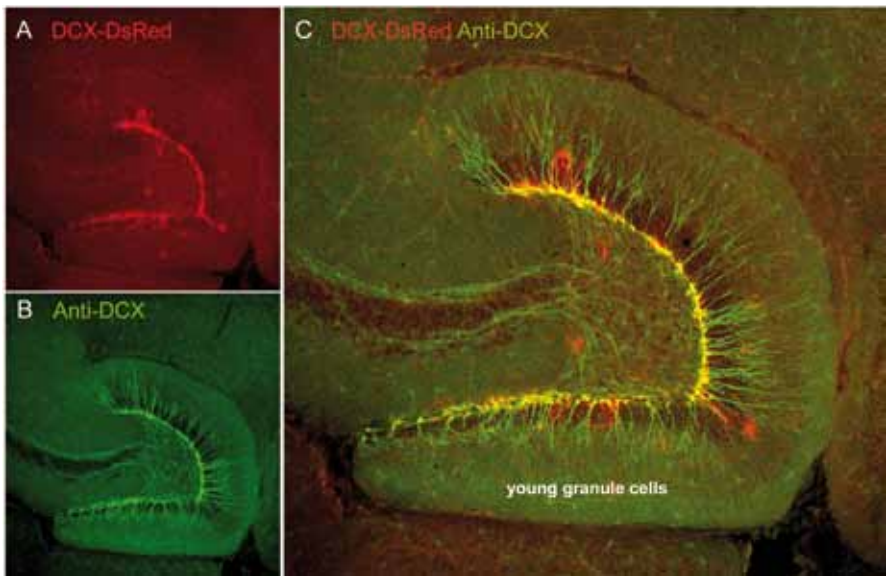


Figure 4
Newly generated young granule cells in the hippocampus of a 2-month-old transgenic mouse expressing DsRed under the control of the DCX promoter.

proving electrophysiological recording techniques, we obtained for the first time direct patch-clamp recordings from these cortical presynaptic terminals (Bischofberger et al. 2006a). We could show that the mossy fiber boutons form extremely powerful synapses. They have an extraordinarily large pool of presynaptic vesicles and show precise presynaptic Ca^{2+} signalling to generate synchronous release of a large number of vesicles (Bischofberger et al. 2006b). As a result, the mossy fiber boutons form probably the only synapses in the mammalian forebrain where a single presynaptic neuron can induce action potential firing in postsynaptically connected neurons (Henze et al. 2002). These results suggest that granule cells powerfully shape the activity patterns in CA3.

Other groups focusing on behavioral analysis in rodents and humans suggested that the dentate gyrus contributes to the separation of similar memory items (Leutgeb et al. 2007, McHugh et al. 2007, Bakker et al. 2008), a function called 'pattern separation'. Hence, similar experiences or events are transformed into distinct non-overlapping neuronal representations in the dentate as well as in the CA3. In support of this idea, optogenetic reactivation of a small ensemble of granule cells that were active during learning was recently shown to be sufficient for the initiation of context-specific memory recall (Liu et al. 2012).

Adult neurogenesis in the hippocampus

When starting my own research group in 2004, I decided to focus on another unique feature of the dentate gyrus, the continuous generation of new neurons throughout life (Bischofberger 2007, Bischofberger and Schinder 2008, Figure 3). Neural progenitors are generated from slowly dividing radial-glia type neural stem cells with cell bodies in the subgranular zone. The progenitors continue to divide several times about once per day. The postmitotic neurons rapidly grow during the next weeks, form dendrites towards the molecular layer where entorhinal input fibers terminate, and send axons into the CA3 region. Proliferation and survival is highly regulated in an activity dependent manner, ensuring that only 'useful' new neurons will be permanently added. During the first 3–4 weeks of maturation, the young granule cells express a number of typical embryonic proteins like PSA-NCAM and doublecortin, which can be used for identification of the young cells within the adult brain (Figure 4).

When studying the functional properties of newly generated granule cells, we found some striking differences compared to the neighboring mature cells. Young neurons show enhanced electrical excitability and are much more sensitive to excitatory inputs than mature cells (Figure 5). Furthermore, they show a lower threshold for synaptic plasticity within a time period of about 2–4 weeks after mitosis (Schmidt-Hieber et al. 2004).

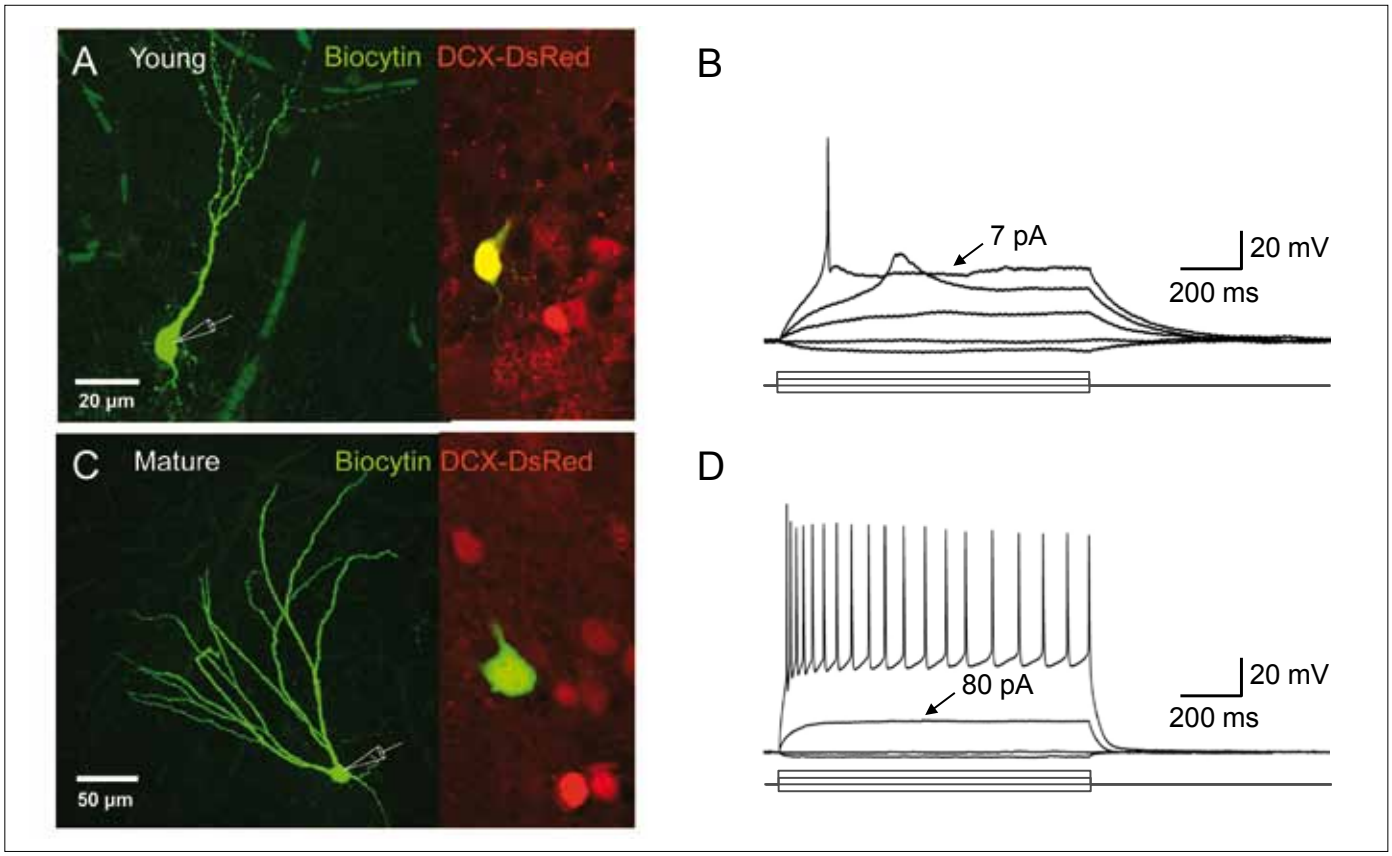


Figure 5 Enhanced excitability of newly generated young granule cells. A Young granule cell filled with biocytin in a hippocampal brain slice, identified on the basis of DCX-DsRed expression. B Action potential firing can be elicited with much smaller current injection in young as compared to mature granule cells shown in C and D. (from Couillard-Despres et al. 2006)

Most interestingly, the newly generated young granule cells were shown to contribute specifically to pattern separation (Clelland, 2009; Sahay et al., 2011; Nakashiba et al., 2012). Based on these results, we hypothesize that synapses in young cells are efficiently adjusted during learning in order to detect 'characteristic features' in neuronal input patterns during memory recall (Bischofberger, 2007). Together with the powerful output synapses, this may help to decorrelate CA3-activity patterns that are associated with different memory items.

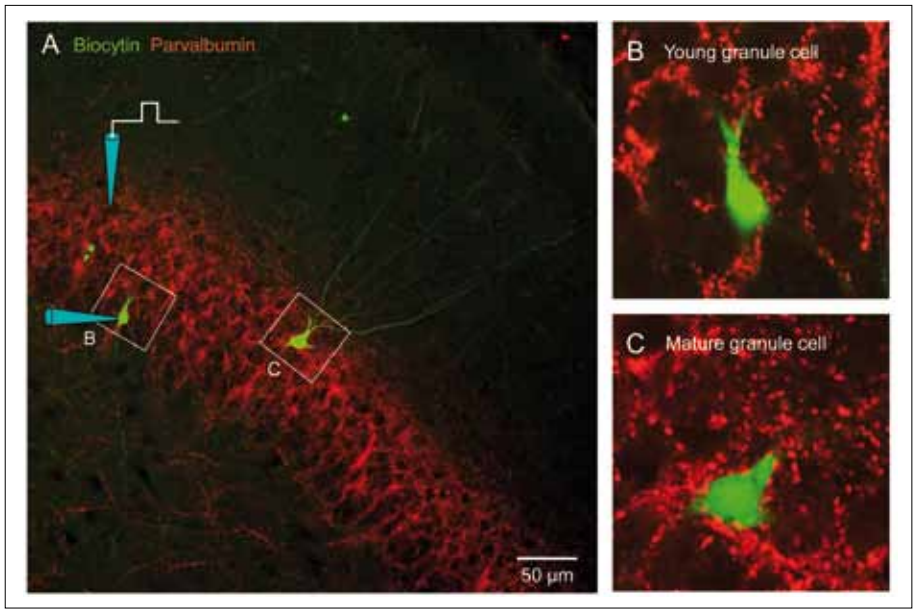


Figure 6
GABAergic synapses in the granule cell layer. A Immunohistochmical staining of parvalbumin-positive presynaptic boutons showing labeled basket cell terminals near a recorded young (B) and mature granule cell (C) filled with biocytin during whole-cell patch clamp recording.

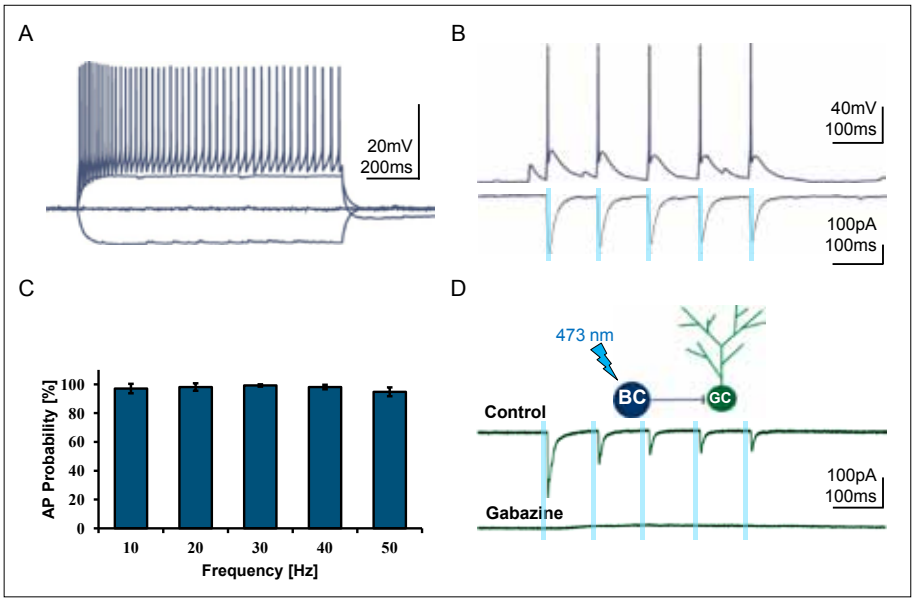


Figure 7
Optogenetic stimulation of GABAergic synapses onto hippocampal granule cells.
A Action potential (AP) firing pattern of a putative GABAergic basket cell evoked by current injection. The interneuron was identified on the basis of its VGAT-YFP-ChR2 expression and the localization within the granule cell layer.
B Single APs evoked by brief 5-ms pulses of blue laser light (upper trace) and Chr2-mediated inward currents (lower trace).
C AP probability at different light stimulation rate.
D GABAergic synaptic currents in a granule cell evoked by light stimulation are mediated by $GABA_A$-receptors sensitive to gabazine.

In consequence, our ability to precisely distinguish similar but different memory items may specifically depend on young neurons in the adult brain.

To better understand the underlying cellular signal processing, we used Ca^{2+} imaging and studied dendritic Ca^{2+} signals in young and mature cells during synaptic activation (Stocca et al. 2008). Young neurons showed relatively large dendritic Ca^{2+} transients with slow decay time courses compared to mature neurons. We could show that the slow decay of Ca^{2+} signals was due to low expression levels of various Ca^{2+} pumps. Furthermore, young neurons showed a surprisingly small Ca^{2+} buffer capacity. Together with the slow Ca^{2+} extrusion rate, this facilitates the generation of large Ca^{2+} signals important for synaptic plasticity and dendritic growth. By contrast, mature granule cells have fast extrusion rates and a high Ca^{2+} buffer capacity. This probably restricts the activation of Ca^{2+} dependent plasticity processes and supports stability of synaptic connections in mature cells.

Current research and outlook

A potential mechanism helping to distinguish and decorrelate different neuronal activity patterns is contrast enhancement. Therefore, we have extended our research on young granule cells to synaptic interactions with GABAergic interneurons forming inhibitory synapses (Figure 6). For functional analysis, we use Ca^{2+}-imaging, patch-clamp recordings and optogenetic stimulation of synaptically connected neurons in acute hippocampal brain slices from transgenic animals where the light-sensitive bacterial protein channelrhodopsin (ChR2) is expressed under the control of interneuron-specific promoters like parvalbumin. In these mice, we can apply brief pulses of blue laser light (5 ms) to selectively generate action potentials in interneurons as well as synaptic response in postsynaptic granule cells (Figure 7).

Pattern separation deficits may contribute to many pathophysiological conditions including mental retardation, anxiety disorders and major depression. Therefore, disturbed adult neurogenesis has been suggested to contribute to these cognitive brain disorders (Kheirbeck et al. 2012, Eisch and Petrik 2012). By investigating mechanisms of synaptic integration and survival of newly generated granule cells in mouse models of mental retardation or anxiety disorders, we hope to contribute to the development of new treatment strategies for memory dysfunction and cognitive disorders in the future.

Josef Bischofberger

References

Bakker A, Kirwan CB, Miller M, Stark CE (2008) *Pattern separation in the human hippocampal CA3 and dentate gyrus.* Science 319:1640–2.

Bischofberger J, Engel D, Liyi L, Geiger JR, Jonas P (2006a) *Patch-clamp recording from mossy fiber terminals in hippocampal slices.* Nature Protocols 1: 2075–2080.

Bischofberger J, Engel D, Frotscher M and Jonas P (2006b) *Mechanisms underlying the efficacy of transmitter release at the mossy fiber synapses in the hippocampal network.* Eur J Physiol 453: 361–372.

Bischofberger J (2007) *Young and excitable: New neurons in memory networks.* Nature Neurosci, 10: 273–275.

Bischofberger J, Schinder A (2008) *Maturation and functional integration of newly generated granule cells into the adult hippocampus.* In Gage FH, Song H, Kempermann G (eds): Adult Neurogenesis. Cold Spring Harbor Press.

Clelland CD, Choi M, Romberg C, Clemenson GD Jr, Fragniere A, Tyers P, Jessberger S, Saksida LM, Barker RA, Gage FH, Bussey TJ. (2009) *A functional role for adult hippocampal neurogenesis in spatial pattern separation.* Science 325:210–213.

Corkin S, Amaral DG, González RG, Johnson KA, Hyman BT (1997) *H. M.'s medial temporal lobe lesion: findings from magnetic resonance imaging.* J Neurosci. 17:3964–79.

Couillard-Despres S, Winner B, Karl C, Lindemann G, Schmid P, Munding M, Aigner R, Wachs F, Laemke J, Kunz-Schughart L, Bogdahn U, Winkler J, Bischofberger J, Aigner L (2006) *Targeted transgene expression in neuronal precursors: Watching young neurons in the old brain.* Eur J Neurosci 24: 1535–45.

Davidson TJ, Kloosterman F, Wilson MA (2009) *Hippocampal replay of extended experience.* Neuron 63:497–507.

Eisch AJ, Petrik D (2012) *Depression and hippocampal neurogenesis: a road to remission?* Science 338:72–5

Henze DA, Wittner L, Buzsáki G (2002) *Single granule cells reliably discharge targets in the hippocampal CA3 network in vivo.* Nat Neurosci 5:790–5.

Huerta PT, Sun LD, Wilson MA, Tonegawa S (2000) *Formation of temporal memory requires NMDA receptors within CA1 pyramidal neurons.* Neuron 25:473–80.

Kheirbek MA, Klemenhagen KC, Sahay A, Hen R (2012) *Neurogenesis and generalization: a new approach to stratify and treat anxiety disorders.* Nat Neurosci 15:1613–20.

Leutgeb JK, Leutgeb S, Moser MB, Moser EI (2007) *Pattern separation in the dentate gyrus and CA3 of the hippocampus.* Science 315:961–6.

Liu X, Ramirez S, Pang PT, Puryear CB, Govindarajan A, Deisseroth K, Tonegawa S (2012) *Optogenetic stimulation of a hippocampal engram activates fear memory recall.* Nature 484:381–5.

McHugh TJ, Jones MW, Quinn JJ, Balthasar N, Coppari R, Elmquist JK, Lowell BB, Fanselow MS, Wilson MA, Tonegawa S (2007) *Dentate gyrus NMDA receptors mediate rapid pattern separation in the hippocampal network.* Science 317:94–9.

Nakashiba T, Cushman JD, Pelkey KA, Renaudineau S, Buhl DL, McHugh TJ, Rodriguez Barrera V, Chittajallu R, Iwamoto KS, McBain CJ, Fanselow MS, Tonegawa S (2012) *Young dentate granule cells mediate pattern separation, whereas old granule cells facilitate pattern completion.* Cell 149:188–201.

Nakazawa K, McHugh TJ, Wilson MA, Tonegawa S. (2004) *NMDA receptors, place cells and hippocampal spatial memory.* Nat Rev Neurosci. 5:361–72.

Sahay A, Wilson DA, Hen R (2011) *Pattern separation: a common function for new neurons in hippocampus and olfactory bulb.* Neuron 70:582–8.

Scoville WB, Milner B (1957) *Loss of recent memory after bilateral hippocampal lesions.* J Neurol Neurosurg Psychiatry. 20:11–21.

Schmidt–Hieber C, Jonas P, Bischofberger J (2004) *Enhanced synaptic plasticity in newly generated granule cells of the adult hippocampus.* Nature 429: 184–187.

Stocca G, Schmidt-Hieber C, Bischofberger J (2008) *Differential dendritic Ca^{2+} signalling in young and mature hippocampal granule cells.* J Physiol 586: 3795–3811.

West MJ, Coleman PD, Flood DG, Troncoso JC (1994) *Differences in the pattern of hippocampal neuronal loss in normal ageing and Alzheimer's disease.* Lancet 344:769–72.

SCIENTIFIC WINTER RETREAT 2013

UNIVERSITÄT BASEL
Vizerektorat Forschung und Nachwuchsförderung
Nachwuchsförderung

This year marked the inaugural DBM PhD winter retreat. Even though this was no small undertaking, the weekend went off without a hitch, running seamlessly thanks to months of planning by the PhD club committee.

Nestled amongst the snow-capped Swiss Alps, Hotel Viktoria in Hasliberg was chosen as the host location this year due to its excellent facilities capable of accommodating 64 students from 34 labs for three days. For students new to the region, there could not have been a better display of the truly awe inspiring nature that Switzerland has to offer.

The retreat was divided into scientific presentations and poster sessions organized, chaired and delivered by the students. 19 talks and over 40 posters covered all four focal areas of the department. Keynote speakers Prof. Nicole Schaeren-Wiemers and Prof. Alfred Zippelius gave enlightening talks regarding their current research, which further elevated the breadth of scientific discussion. Recipients of best posters (Frédéric Laurent and Robert Beattie) and presentation (Sabrina Di Fulvio) were awarded the highly coveted golden pipette and golden pipetboy. When asked how Sumit Jaiswal, a participant in the retreat, felt in regards to the quality of the talks he had this to say. “I walked away more informed and with a greater understanding of the scientific questions my colleagues are tackling, not only within the building [Mattenstrasse], but across our entire department.”

Over the course of the retreat, organizers planned a variety of social activities to encourage discussion amongst the students. There was a traditional fondue dinner and an apéro to close off the poster and presentation sessions. On the final day, there was some time set-aside for skiing, sledging and hiking. Although much of the snow had already left the lower slopes, the top half of the mountain was still fresh with powder.

One of the greatest benefits of a retreat like this is being able to step back from the bench and look at how our work fits into the larger scope. Creating this open scientific forum allowed for stimulating intradepartmental discussions fostering creativity and future collaborations. It gives us a primary role in influencing our development as scientists, further enhancing the pioneering status of the department. With students expectations exceeded, it will be exciting to see how the momentum from this winter retreat will continue to have cumulative effects throughout the Department of Biomedicine.

Robert Beattie, PhD student in the lab of Prof. Verdon Taylor.

The organizers (Carlos Mayer, Frédéric Laurent, Fabrizio Botindari, Maren Diepenbruck, Yvonne Fink and Kea Martin) would like to thank the DBM and all the sponsors that made this event possible.
More info: phdclub-dbm@unibas.ch

Dissertationen

Seit dem 18. Dezember 2012 darf sich **Bei Zhang** von der Forschungsgruppe Gyn. Oncology (Departement Biomedizin Hebelstrasse) Frau Dr. nennen. Sie befasste sich in ihrer Doktorarbeit mit dem Thema: "Salinomycin as a potential chemotherapeutic compound in cisplatin-resistant ovarian cancer: effects and mechanisms".

Mit der Doktorprüfung ebenfalls am 18. Dezember 2012 schloss **Stefan Sladecek** von der Forschungsgruppe Molecular Neurobiology Synaptic Formation (Departement Biomedizin Klingelbergstrasse) erfolgreich seine Dissertationszeit ab. Das Thema seiner Doktorarbeit lautete: "Agrin promotes acetylcholine receptor clustering at the mammalian neuro-muscular junction by PI3K/GSK3β-mediated regulation of +TIPs and microtubule capture".

Am 27. Februar 2013 stellte sich **Gaia Trincucci** von der Forschungsgruppe Hepatology (Departement Biomedizin Hebelstrasse) dem Dissertationskomitee. Der Titel ihrer Dissertation hiess: "Interferon signaling in viral hepatitis".

Am 1. März 2013 konnte **Maria Broggi** von der Forschungsgruppe Immunoregulation (Departement Biomedizin Hebelstrasse) ihre Dissertation mit Erfolg beenden. Sie befasste sich in ihrer Dissertation mit dem Thema "Characterization of lymph node stromal cells during Treg-mediated tolerance".

Mit der Doktorprüfung am 26. März 2013 beendete **Linda Simmler** von der Forschungsgruppe Psychopharmacology Research (Departement Biomedizin Hebelstrasse) erfolgreich ihre Dissertationszeit. Das Thema ihrer Doktorarbeit lautete: "Pharmacology of amphetamine-type designer drugs".

Venia docendi an Jean-Louis Boulay und Peter Hruz

In ihrer Sitzung am 13. März 2013 hat die Regenz der Universität Basel **Jean-Louis Boulay** von der Forschungsgruppe Brain Tumor Biology (Departement Biomedizin Klingelbergstrasse) die Venia docendi für Experimentelle Medizin, speziell Molekulare Onkologie, erteilt. **Petr Hruz** von der Forschungsgruppe Exp. Immunology (Departement Biomedizin Hebelstrasse) erhielt die Venia docendi für Innere Medizin, speziell für Gastroenterologie und Hepatologie. Beide dürfen nun den Titel eines Privatdozenten führen.

Alfred Zippelius zum Professor für Translationale Onkologie gewählt

Der Universitätsrat hat in seiner Sitzung am 21. März 2013 **Alfred Zippelius** von der Forschungsgruppe Cancer Immunology (Departement Biomedizin Hebelstrasse) zum neuen Professor für Translationale Onkologie gewählt. Alfred Zippelius ist am USB stv. Chefarzt der Abteilung Onkologie.

Herzliche Gratulation an alle!

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 May 31, 2013.

Lancet Oncology THE LANCET 2012; 13: 1234–41 IF 22,5

Tolerability, safety, pharmacokinetics, and efficacy of doxorubicin-loaded anti-EGFR immunoliposomes in advanced solid tumours: a phase 1 dose-escalation study

Christoph Mamot, Reto Ritschard, Andreas Wicki, Gregor Stehle, Thomas Dieterle, Lukas Bubendorf, Christoph Hilker, Stefanie Deuster, Richard Herrmann, Christoph Rochlitz

Summary

Background Results of preclinical studies have shown that EGFR immunoliposomes have substantial antitumour effects. We aimed to assess the tolerability, safety, pharmokinetics, and efficacy of anti-EGFR immunoliposomes loaded with doxorubicin (anti-EGFR ILs-dox) in patients with solid tumours.

Methods In this first-in-man, open-label, phase 1 clinical study, we enrolled patients at University Hospital of Basel, Switzerland, who had EGFR-overexpressing advanced solid tumours no longer amenable to standard treatment. Anti-EGFR ILs-dox nanoparticles were constructed by covalently linking pegylated liposomes containing doxorubicin to antigen-binding fragments (Fabc) of cetuximab. We intravenously infused the nanoparticle at escalating doses (doxorubicin 5 mg/m^2, 10 mg/m^2, 20 mg/m^2, 30 mg/m^2, 40 mg/m^2, 50 mg/m^2, and 60 mg/m^2) once every 4 weeks for a maximum of six cycles. The primary endpoint was to establish the maximum tolerated dose. We analysed patients who received at least one dose of study drug. This study is registered with ClinicalTrials.gov, number NCT01702129.

Findings Between Jan 30, 2007, and March 4, 2010, we gave the drug to 29 patients, three of whom were withdrawn from the study because we could not complete a safety assessment. Of the 26 patients assessed for the primary endpoint, two who received a dose of 60 mg/m^2 had dose-limiting toxicities (one had neutropenia and the other had anaemia); therefore, the maximum tolerated dose was defi ned as 50 mg/m^2. At all lower doses, anti-EGFR ILs-dox was well tolerated; grade 1 skin toxicity occurred in two patients only. We recorded 22 serious adverse events (SAEs) in 17 patients, mostly due to tumour progression. Three SAEs were fatal. Only three SAEs (febrile neutropenia, septicaemia, and a fatal massive oral bleed) were probably or possibly related to study drug. No patients had palmar-plantar erythrodysaesthesia, alopecia, cardiotoxicity, or cumulative toxicity.

Division of Oncology
Department of Biomedicine
Division of Cardiology
Institute of Pathology and Hospital Pharmacy
University Hospital of Basel, Switzerland; and Division of Haematology/Oncology, Cantonal Hospital of Aarau, Switzerland

PLOS Biology | PLOS BIOLOGY | 2012 | Vol. 10 | Issue 12 | e1001439 IF 11,4

Growth Cone *MKK7* mRNA Targeting Regulates MAP1b-Dependent Microtubule Bundling to Control Neurite Elongation

Daniel Feltrin[1], Ludovico Fusco[1], Harald Witte[2], Francesca Moretti[1], Katrin Martin[1], Michel Letzelter[1], Erika Fluri[1], Peter Scheiffele[2], Olivier Pertz[1]

Abstract

Local mRNA translation in neurons has been mostly studied during axon guidance and synapse formation but not during initial neurite outgrowth. We performed a genome-wide screen for neurite-enriched mRNAs and identified an mRNA that encodes mitogen-activated protein kinase kinase 7 (MKK7), a MAP kinase kinase (MAPKK) for Jun kinase (JNK). We show that *MKK7* mRNA localizes to the growth cone where it has the potential to be translated. MKK7 is then specifically phosphorylated in the neurite shaft, where it is part of a MAP kinase signaling module consisting of dual leucine zipper kinase (DLK), MKK7, and JNK1. This triggers Map1b phosphorylation to regulate microtubule bundling leading to neurite elongation. We propose a model in which *MKK7* mRNA localization and translation in the growth cone allows for a mechanism to position JNK signaling in the neurite shaft and to specifically link it to regulation of microtubule bundling. At the same time, this uncouples activated JNK from its functions relevant to nuclear translocation and transcriptional activation.

[1] Institute for Biochemistry and Genetics, Department of Biomedicine, University of Basel, Switzerland
[2] Biozentrum, University of Basel, Switzerland

Arthritis & Rheumatism | American College of Rheumatology | Vol. 64, No. 11, Nov. 2012, pp 3706–3714 IF 7,8

Identification of a Major Linear C1q Epitope Allows Detection of Systemic Lupus Erythematosus Anti-C1q Antibodies by a Specific Peptide-Based Enzyme-Linked Immunosorbent Assay

Dominique Vanhecke[1], Lubka T. Roumenina[2], Hui Wan[1], Michael Osthoff[1], Monica Schaller[3], Marten Trendelenburg[1]

Objective

Autoantibodies against C1q strongly correlate with the occurrence of severe nephritis in patients with systemic lupus erythematosus (SLE). We undertook this study to determine whether identification of the C1q epitope(s) recognized by these autoantibodies might lead to a better diagnostic assay and help elucidate the putative role of C1q and anti-C1q in SLE.

Methods

SLE patient–derived anti-C1q Fab were used in a microarray-based peptide scan to identify the peptide sequence recognized by anti-C1q. Anti-C1q Fab binding to the target peptide was further analyzed using real-time interaction measurements (surface plasmon resonance) and peptide-based enzyme-linked immunosorbent assays (ELISAs).

Results

A peptide scan of the collagen-like region of C1q identified 2 regions, 1 on the A chain and 1 on the B chain, that were the targets of the anti-C1q Fab. Binding was confirmed by surface plasmon resonance and showed nanomolar affinity. The A chain–derived peptide could specifically be detected in a peptide-based ELISA by SLE patient sera. Competition experiments suggested that this peptide represented one of the major linear epitopes of C1q that is the target of anti-C1q in SLE. Serum antibodies from most SLE patients but not from healthy individuals specifically bound to this epitope. Binding to the peptide correlated with binding of the same sera to native C1q but was found to be more sensitive for the detection of lupus nephritis.

Conclusion

We identified a major linear epitope of C1q that is the target of anti-C1q in SLE. The ELISA using this peptide was more specific and more sensitive than a conventional anti-C1q assay for the detection of active nephritis in SLE patients.

[1] University Hospital Basel, Switzerland
[2] Centre de Recherche des Cordeliers, INSERM, U872, Université Pierre et Marie Curie – Paris 6, UMR S 872, and Université Paris Descartes, UMR S 872, Paris, France
[3] University Hospital Basel, Switzerland, and Inselspital – Bern University Hospital and University of Bern, Switzerland.

Biomaterials 34 (2013) 393–401 IF 7,4

The effect of controlled expression of VEGF by transduced myoblasts in a cardiac patch on vascularization in a mouse model of myocardial infarction

Anna Marsano[a,b,c], Robert Maidhof[a], Jianwen Luo[a], Kana Fujikara[a], Elisa E. Konofagou[a], Andrea Banfi[b,c,*], Gordana Vunjak-Novakovic[a,*]

Abstract

Key requirements for cardiac tissue engineering include the maintenance of cell viability and function and the establishment of a perfusable vascular network in millimeters thick and compact cardiac constructs upon implantation. We investigated if these requirements can be met by providing an intrinsic vascularization stimulus (via sustained action of VEGF secreted at a controlled rate by transduced myoblasts) to a cardiac patch engineered under conditions of effective oxygen supply (via medium flow through channeled elastomeric scaffolds seeded with neonatal cardiomyocytes). We demonstrate that this combined approach resulted in increased viability, vascularization and functionality of the cardiac patch. After implantation in a mouse model of myocardial infarction, VEGF-expressing patches displayed significantly improved engraftment, survival and differentiation of cardiomyocytes, leading to greatly enhanced contractility as compared to controls not expressing VEGF. Controlled VEGF expression also mediated the formation of mature vascular networks, both within the engineered patches and in the underlying ischemic myocardium. We propose that this combined cell-biomaterial approach can be a promising strategy to engineer cardiac patches with intrinsic and extrinsic vascularization potential.

[a] Columbia University, Department of Biomedical Engineering, New York, USA
[b] Basel University Hospital, Department of Biomedicine, Switzerland
[c] Basel University Hospital, Department of Surgery, Switzerland
[*] Equally contributing authors.

Angiogenesis Springer (2013) 16:123–136 IF 6,0

VEGF over-expression in skeletal muscle induces angiogenesis by intussusception rather than sprouting

Roberto Gianni-Barrera[1], Marianna Trani[1], Christian Fontanellaz[2], Michael Heberer[1], Valentin Djonov[2], Ruslan Hlushchuk[2], Andrea Banfi[1]

Abstract

Therapeutic over-expression of vascular endothelial growth factor (VEGF) can be used to treat ischemic conditions. However, VEGF can induce either normal or aberrant angiogenesis depending on its dose in the microenvironment around each producing cell in vivo, which limits its clinical usefulness. The goal herein was to determine the cellular mechanisms by which physiologic and aberrant vessels are induced by over-expression of different VEGF doses in adult skeletal muscle. We took advantage of a well-characterized cell-based platform for controlled gene expression in skeletal muscle. Clonal populations of retrovirally transduced myoblasts were implanted in limb muscles of immunodeficient mice to homogeneously over-express two specific $VEGF_{164}$ levels, previously shown to induce physiologic and therapeutic or aberrant angiogenesis, respectively. Three independent and complementary methods (confocal microscopy, vascular casting and 3D-reconstruction of serial semi-thin sections) showed that, at both VEGF doses, angiogenesis took place without sprouting, but rather by intussusception, or vascular splitting. VEGF-induced endothelial proliferation without tip-cell formation caused an initial homogeneous enlargement of pre-existing microvessels, followed by the formation of intravascular transluminal pillars, hallmarks of intussusception. This was associated with increased flow and shear stress, which are potent triggers of intussusception. A similar process of enlargement without sprouting, followed by intussusception, was also induced by VEGF over-expression through a clinically relevant adenoviral gene therapy vector, without the use of transduced cells. Our findings indicate that VEGF over-expression, at doses that have been shown to induce functional benefit, induces vascular growth in skeletal muscle by intussusception rather than sprouting.

[1] Cell and Gene Therapy, Department of Biomedicine and Department of Surgery, Basel University Hospital, Switzerland
[2] Institute of Anatomy, University of Bern, Switzerland

European Respiratory Journal 2013; 41: 195–202 IF 5,8

Dimethylfumarate inhibits CXCL10 *via* haem oxygenase-1 in airway smooth muscle

Petra Seidel[1,2], Katrin E. Hostettler[1], J. Margaret Hughes[2], Michael Tamm[1], Michael Roth[1]

Abstract

CXCL10 stimulates mast cell infiltration into airway smooth muscle bundles and, thus, activate cytokine secretion and airwaysmoothmuscle cell (ASMC) proliferation. Dimethylfumarate (DMF) reduces cytokine secretion by lymphocytes and ASMC proliferation through haem oxygenase (HO)-1. Therefore, we investigated the potency of DMF to inhibit tumour necrosis factor (TNF)-α-and interferon (IFN)-γ-induced CXCL10 secretion by human ASMCs.

Human primary ASMCs were pre-incubated with DMF and/or fluticasone and/or glutathione ethylester before cells were stimulated with IFN-γ and/or TNF-α.

DMF inhibited CXCL10 secretion and increased HO-1 levels, and p38 mitogen-activated protein kinase (MAPK) inhibition reduced DMF-dependent HO-1 expression. The DMF effect on CXCL10 secretion was abrogated by pre-treatment with HO-1 small interfering RNA (siRNA). Glutathione supplementation reversed all DMF effects on CXCL10 secretion and p38 MAPK phosphorylation. Importantly, combining DMF with fluticasone further reduced CXCL10 secretion. In addition, DMF inhibited IFN-γ-induced CXCL10 secretion. This effect was compensated by glutathione supplementation or by pre-treatment with HO-1 siRNA. In addition, DMF reduced TNF-α-induced granulocyte colony-stimulating factor (G-CSF) secretion but had no effect on INF-γ-induced G-CSF secretion.

In human primary ASMCs, DMF inhibits CXCL10 secretion by reducing the cellular glutathione level and by activation of p38 MAPK and HO-1. Therefore, DMF may reduce airway inflammation in asthma by a glucocorticoid-independent pathway.

Keywords

Airway smooth muscle, cell inflammation, dimethylfumarate, glutathione, haem oxygenase-1, interferon-γ, tumour necrosis factor-α

[1] Pulmonary Cell Research, Dept of Biomedicine and Pneumology, Dept of Internal Medicine, University Hospital Basel, Switzerland.
[2] Faculty of Pharmacy, University of Sydney, Australia.

Human Mutation HGVS HUMAN GENOME VARIATION SOCIETY DOI: 10.1002/humu.22209 IF 5,686

JP-45/*JSRP1* Variants Affect Skeletal Muscle Excitation-Contraction Coupling by Decreasing the Sensitivity of the Dihydropyridine Receptor

Toshimichi Yasuda[1], Osvaldo Delbono[2], Zhong-Min Wang[2], Maria L. Messi[2], Thierry Girard[3], Albert Urwyler[3], Susan Treves[3,4], Francesco Zorzato[3,4]

Abstract

JP-45 (also JP45; encoded by *JSRP1*) is an integral protein constituent of the skeletal muscle sarcoplasmic reticulum junctional face membrane interacting with Cav1.1 (the α.1 subunit of the voltagesensing dihydropyridine receptor, DHPR) and the luminal calcium-binding protein calsequestrin. Two *JSRP1* variants have been found in the human population: c.323C>T (p.P108L) in exon 5 and c.449G>C (p.G150A) in exon 6, but nothing is known concerning the incidence of these polymorphisms in the general population or in patients with neuromuscular diseases nor the impact of the polymorphisms on excitation–contraction (EC) coupling. In the present report, we investigated the frequencies of these two JSRP1 polymorphisms in the Swiss malignant hyperthermia population and studied the functional impact of the variants on EC coupling. Our results show that the polymorphisms are equally distributed among malignant hyperthermia negative, malignant hyperthermia equivocal, and malignant hyperthermia susceptible individuals. Interestingly, however, the presence of either one of these JP-45 variants decreased the sensitivity of the DHPR to activation. The presence of a *JSRP1* variant may explain the variable phenotype seen in patients with malignant hyperthermia carrying the same mutation and, more importantly, may counteract the hypersensitivity of EC coupling caused by mutations in the *RYR1* gene.

[1] Department of Anesthesiology and Critical Care, Hiroshima University, Manami-ku, Japan
[2] Department of Internal Medicine, Gerontology and Geriatric Medicine, Wake Forest School of Medicine, Medical Center Boulevard, Winston-Salem, North Carolina
[3] Departments of Anesthesia and Biomedizin, Basel University Hospital, Switzerland
[4] Department of Experimental and Diagnostic Medicine, General Pathology Section, University of Ferrara, Italy

Neurobiology of Disease 49 (2013) 221–231 IF 5,4

An essential role of MAG in mediating axon–myelin attachment in Charcot–Marie–Tooth 1A disease

Jochen Kinter[1], Thomas Lazzati[1], Daniela Schmid[1], Thomas Zeis[1], Beat Erne[1], Roland Lützelschwab[1,2], Andreas J. Steck[1,2], Davide Pareyson[3], Elior Peles[4], Nicole Schaeren-Wiemers[1,2]

Abstract:
Charcot–Marie–Tooth disease type 1A (CMT1A) is a hereditary demyelinating peripheral neuropathy caused by the duplication of the PMP22 gene. Demyelination precedes the occurrence of clinical symptoms that correlate with axonal degeneration. It was postulated that a disturbed axon–glia interface contributes to altered myelination consequently leading to axonal degeneration. In this study, we examined the expression of MAG and Necl4, two critical adhesion molecules that are present at the axon–glia interface, in sural nerve biopsies of CMT1A patients and in peripheral nerves of mice overexpressing human PMP22, an animal model for CMT1A. We show an increase in the expression of MAG and a strong decrease of Necl4 in biopsies of CMT1A patients as well as in CMT1A mice. Expression analysis revealed that MAG is strongly upregulated during peripheral nerve maturation, whereas Necl4 expression remains very low. Ablating MAG in CMT1A mice results in separation of axons from their myelin sheath. Our data show that MAG is important for axon–glia contact in a model for CMT1A, and suggest that its increased expression in CMT1A disease has a compensatory role in the pathology of the disease. Thus, we demonstrate that MAG together with other adhesion molecules such as Necl4 is important in sustaining axonal integrity.

[1] Neurobiology, Department of Biomedicine, University Hospital Basel, University of Basel, Switzerland
[2] Department of Neurology, University Hospital Basel, University of Basel, Switzerland
[3] IRCCS Foundation, C. Besta Neurological Institute, Milan, Italy
[4] Department of Molecular Cell Biology, The Weizmann Institute of Science, Rehovot 76100, Israel

Am J Respir Cell Mol Biol AMERICAN JOURNAL OF Respiratory Cell and Molecular Biology® Vol 47, Iss. 6, pp 759–767, 2012 IF5, 1

Steroids and β_2-Agonists Regulate Hyaluronan Metabolism in Asthmatic Airway Smooth Muscle Cells

Eleni Papakonstantinou[1], Ioannis Klagas[1], George Karakiulakis[1], Katrin Hostettler[1], Chong Teck S'ng[3], Vassiliki Kotoula[2], Spasenija Savic[4], Michael Tamm[3], Michael Roth[3]

Glycosaminoglycans (GAGs), especially hyaluronic acid (HA), regulate tissue flexibility, cell motility, and inflammation. Airway smooth muscle cells (ASMCs) of patients with asthma exhibit abnormal HA metabolism, which contributes to inflammation and remodeling. Here, we investigated the effects of glucocorticoids and long-acting β_2-agonists(LABAs)onGAGsynthesisandHAmetabolismbyhuman primary ASMCs. ASMCs were isolated from airway specimens of 10 patients without asthma and 11 patients with asthma. ASMCs were incubated with glucocorticoids, LABAs, or their combination, as well as with their specific receptor antagonists. Secreted and deposited total GAGs were measured by [^{3}H]-glucosamine incorporation. The expression of specific GAGs was determined by ELISA and electrophoresis. The expression of HA synthases (HAS), of hyaluronidases (HYALs), and of the HA receptor CD44 was determined by RT-PCR, immunoblotting in cell cultures, and immunohistochemistry in tissue sections of asthmaticlungs.Inserum-activatedasthmatic ASMCs, glucocorticoids and LABAs significantly inhibited the increased secretion and deposition of total GAGs, but they stimulated secreted and deposited HA of high molecular mass. This effect was attributed to increased mRNA and protein expression of HAS-1 and to the reduced expression of HYAL-1. Furthermore, drug treatment stimulated the expression of CD44 receptors in asthmatic ASMCs. Theseeffects of the drugs were eliminated by their respectivereceptor inhibitors. Our findings indicate that the combination of glucocorticoids with LABAs counteracts the pathologic degradation of HA, and thereby may reduce the proinflammatory potential of asthmatic ASMCs.

[1] Department of Pharmacology, School of Medicine, Aristotle University, Thessaloniki, Greece
[2] Department of Pathology, School of Medicine, Aristotle University, Thessaloniki, Greece
[3] Division of Pulmonary Cell Research–Pneumology, Department of Biomedicine and Internal Medicine, University Hospital Basel, Switzerland
[4] Department of Pathology, University Hospital Basel, Switzerland

The Journal of Biological Chemistry | THE JOURNAL OF BIOLOGICAL CHEMISTRY | Vol. 287, No. 47, pp. 39869–39877, 2012 IF 4,7

Opposite Effects of KCTD Subunit Domains on $GABA_B$ Receptor-mediated Desensitization*

Riad Seddik[1,•], Stefan P. Jungblut[1,•], Olin K. Silander[2], Mathieu Rajalu[1], Thorsten Fritzius[1], Valérie Besseyrias[1], Valérie Jacquier[1], Bernd Fakler[3,4], Martin Gassmann[1], Bernhard Bettler[1]

$GABA_B$ receptors assemble from principle and auxiliary subunits. The principle subunits $GABA_{B1}$ and $GABA_{B2}$ form functional heteromeric $GABA_{B(1,2)}$ receptors that associate with homotetramers of auxiliary KCTD8, -12, -12b, or -16 (named after their K^+ channel tetramerization domain) subunits. These auxiliary subunits constitute receptor subtypes with distinct functional properties. KCTD12 and- 12b generate desensitizing receptor responses while KCTD8 and -16 generate largely non-desensitizing receptor responses. The structural elements of the KCTDs underlying these differences in desensitization are unknown. KCTDs are modular proteins comprising a T1 tetramerization domain, which binds to $GABA_{B2}$, and a H1 homology domain. KCTD8 and -16 contain an additionalC-terminal H2 homology domain that is not sequence-related to the H1 domains. No functions are known for the H1 and H2 domains. Here we addressed which domains and sequencemotifs in KCTD proteins regulate desensitization of the receptor response. We found that the H1 domains in KCTD12 and-12b mediate desensitization through a particular sequence motif, T/NFLEQ, which is not present in the H1 domains of KCTD8 and -16. In addition, the H2 domains in KCTD8 and -16 inhibit desensitization when expressed C-terminal to the H1domains but not when expressed as a separate protein *in trans*. Intriguingly, the inhibitory effect of the H2 domain is sequence-independent, suggesting that the H2 domain sterically hinders desensitization by the H1 domain. Evolutionary analysis supports that KCTD12 and -12b evolved desensitizing properties by liberating their H1 domains from antagonistic H2 domains and acquisition of the T/NFLEQ motif.

[1] Department of Biomedicine, University of Basel, 4056 Basel, Switzerland
[2] Core Program Computational and Systems Biology, Biozentrum, University of Basel, Switzerland
[3] Institute of Physiology II, University of Freiburg, Germany
[4] Center for Biological Signaling Studies (bioss), 79104 Freiburg, Germany
• Both authors contributed equally to this work.
* This work was supported by grants from the Swiss National Science Foundation (3100A0-117816), the National Center for Competence in Research (NCCR) "Synapsy, Synaptic Bases of Mental Health Disease" (to B. B.), the European Community's Seventh Framework Programme (FP7/2007-2013) under Grant Agreement 201714 (to B. B.), an Ambizione fellowship from the Swiss National Science Foundation (to O. K. S.), and a Marie Heim-Vögtlin fellowship from the Swiss National Science Foundation (to V. J.).

Toxicological Sciences | TOXICOLOGICAL SCIENCES | 131(2), 480–490 2013 IF 4,6

Mechanisms of Hepatocellular Toxicity Associated with Dronedarone—A Comparison to Amiodarone

Andrea Felser[1,2], Kim Blum[1,2], Peter W. Lindinger[1,2,3], Jamal Bouitbir[1,2,3], Stephan Krähenbühl[1,2,3]

Dronedarone is a new antiarrhythmic drug with an amiodaronelike benzofuran structure. Shortly after its introduction, dronedarone became implicated in causing severe liver injury. Amiodarone is a well-known mitochondrial toxicant. The aim of our study was to investigate mechanisms of hepatotoxicity of dronedarone in vitro and to compare them with amiodarone. We used isolated rat liver mitochondria, primary human hepatocytes, and the human hepatoma cell line HepG2, which were exposed acutely or up to 24h. After exposure of primary hepatocytes or HepG2 cells for 24h, dronedarone and amiodarone caused cytotoxicity and apoptosis starting at 20 and 50μM, respectively. The cellular ATP content started to decrease at 20μM for both drugs, suggesting mitochondrial toxicity. Inhibition of the respiratory chain required concentrations of ~10μM and was caused by an impairment of complexes I and II for both drugs. In parallel, mitochondrial accumulation of reactive oxygen species (ROS) was observed. In isolated rat liver mitochondria, acute treatment with dronedarone decreased the mitochondrial membrane potential, inhibited complex I, and uncoupled the respiratory chain. Furthermore, in acutely treated rat liver mitochondria and in HepG2 cells exposed for 24 h, dronedarone started to inhibit mitochondrial β-oxidation at 10μM and amiodarone at 20μM. Similar to amiodarone, dronedarone is an uncoupler and an inhibitor of the mitochondrial respiratory chain and of β-oxidation both acutely and after exposure for 24h. Inhibition of mitochondrial function leads to accumulation of ROS and fatty acids, eventually leading to apoptosis and/or necrosis of hepatocytes. Mitochondrial toxicity may be an explanation for hepatotoxicity of dronedarone in vivo.

[1] Department of Clinical Pharmacology & Toxicology, University Hospital, Basel, Switzerland
[2] Department of Biomedicine, University of Basel, Switzerland
[3] Swiss Center of Applied Human Toxicology (SCAHT), University of Basel, Switzerland

British Journal of Pharmacology | BJP British Journal of Pharmacology | (2013) 168 458–470 | IF 4,409

Pharmacological characterization of designer cathinones *in vitro*

LD Simmler[1], TA Buser[1], M Donzelli[1], Y Schramm[2], L-H Dieu[3], J Huwyler[3], S Chaboz[4], MC Hoener[4], ME Liechti[1]

Background and purpose
Designer β-keto amphetamines (e.g. cathinones, 'bath salts' and 'research chemicals') have become popular recreational drugs, but their pharmacology is poorly characterized.

Experimental approach
We determined the potencies of cathinones to inhibit DA, NA and 5-HT transport into transporter-transfected HEK 293 cells, DA and 5-HT efflux from monoamine-preloaded cells, and monoamine receptor binding affinity.

Key results
Mephedrone, methylone, ethylone, butylone and naphyrone acted as non-selective monoamine uptake inhibitors, similar to cocaine. Mephedrone, methylone, ethylone and butylone also induced the release of 5-HT, similar to 3,4-methylenedioxymethamphetamine (MDMA, ecstasy) and other entactogens. Cathinone, methcathinone and flephedrone, similar to amphetamine and methamphetamine, acted as preferential DA and NA uptake inhibitors and induced the release of DA. Pyrovalerone and 3,4-methylenedioxypyrovalerone (MDPV) were highly potent and selective DA and NA transporter inhibitors but unlike amphetamines did not evoke the release of monoamines. The non-β-keto amphetamines are trace amine-associated receptor 1 ligands, whereas the cathinones are not. All the cathinones showed high blood–brain barrier permeability in an *in vitro* model; mephedrone and MDPV exhibited particularly high permeability.

Conclusions and implications
Cathinones have considerable pharmacological differences that form the basis of their suggested classification into three groups. The predominant action of all cathinones on the DA transporter is probably associated with a considerable risk of addiction.

[1] Division of Clinical Pharmacology and Toxicology, Departments of Biomedicine and Internal Medicine, University Hospital and University of Basel, Switzerland
[2] Department of Chemistry, University of Basel, Switzerland
[3] Department of Pharmaceutical Sciences, University of Basel, Switzerland
[4] Pharmaceuticals Division, Neuroscience Research, F. Hoffmann-La Roche Ltd, Basel, Switzerland

Human Gene Therapy Methods | Mary Ann Liebert, Inc. publishers | 23:346–356 (October 2012) | IF 4,2

Controlled Angiogenesis in the Heart by Cell-Based Expression of Specific Vascular Endothelial Growth Factor Levels

Ludovic F. Melly[1,2,*], Anna Marsano[1,*], Aurelien Frobert[3,4], Stefano Boccardo[5], Uta Helmrich[1], Michael Heberer[1], Friedrich S. Eckstein[2], Thierry P. Carrel[3], Marie-Noëlle Giraud[3,4], Hendrik T. Tevaearai[3], Andrea Banfi[1]

Abstract
Vascular endothelial growth factor (VEGF) can induce normal angiogenesis or the growth of angioma-like vascular tumors depending on the amount secreted by each producing cell because it remains localized in the microenvironment. In order to control the distribution of VEGF expression levels *in vivo,* we recently developed a high-throughput fluorescence-activated cell sorting (FACS)-based technique to rapidly purify transduced progenitors that homogeneously express a specific VEGF dose from a heterogeneous primary population. Here we tested the hypothesis that cell-based delivery of a controlled VEGF level could induce normal angiogenesis in the heart, while preventing the development of angiomas. Freshly isolated human adipose tissue–derived stem cells (ASC) were transduced with retroviral vectors expressing either rat VEGF linked to a FACS-quantifiable cell-surface marker (a truncated form of CD8) or CD8 alone as control (CTR). VEGF-expressing cells were FACS-purified to generate populations producing either a specific VEGF level (SPEC) or uncontrolled heterogeneous levels (ALL). Fifteen nude rats underwent intramyocardial injection of 10^7 cells. Histology was performed after 4 weeks. Both the SPEC and ALL cells produced a similar total amount of VEGF, and both cell types induced a 50%–60% increase in both total and perfused vessel density compared to CTR cells, despite very limited stable engraftment. However, homogeneous VEGF expression by SPEC cells induced only normal and stable angiogenesis. Conversely, heterogeneous expression of a similar total amount by the ALL cells caused the growth of numerous angioma-like structures. These results suggest that controlled VEGF delivery by FACS-purified ASC may be a promising strategy to achieve safe therapeutic angiogenesis in the heart.

[1] Cell and Gene Therapy, Department of Biomedicine and Department of Surgery
[2] Clinic for Cardiac Surgery, Basel University Hospital and University of Basel, Switzerland
[3] Department of Cardiovascular Surgery, Inselspital, Bern University Hospital and University of Bern, Switzerland
[4] Cardiology, University of Fribourg, Switzerland
[5] Istituto Italiano di Tecnologia, Robotics, Brain and Cognitive Sciences Department, Genova, Italy
[*] These authors contributed equally.

Plos One PLOS ONE Nov. 2012, Vol. 7, Issue 11, e48651 IF 4,0

Py2T Murine Breast Cancer Cells, a Versatile Model of TGFβ-Induced EMT *In Vitro* and *In Vivo*

Lorenz Waldmeier, Nathalie Meyer-Schaller, Maren Diepenbruck, Gerhard Christofori

Abstract

Introduction: Increasing evidence supports a role of an epithelial to mesenchymal transition (EMT) process in endowing subsets of tumor cells with properties driving malignant tumor progression and resistance to cancer therapy. To advance our understanding of the underlying mechanisms, we sought to generate a transplantable cellular model system that allows defined experimental manipulation and analysis of EMT *in vitro* and at the same time recapitulates oncogenic EMT *in vivo.*

Methodology/Results: We have established a stable murine breast cancer cell line (Py2T) from a breast tumor of an MMTV-PyMT transgenic mouse. Py2T cells display a metastable epithelial phenotype characterized by concomitant expression of luminal and basal cytokeratins and sheet migration. Exposure of Py2T cells to transforming growth factor β (TGF β) *in vitro* induces reversible EMT accompanied by downregulation of E-cadherin and upregulation of mesenchymal markers, including EMT transcription factors, and a gain in single cell motility and invasiveness. Py2T cells give rise to tumors after orthotopic injection into syngeneic FVB/N mice. Notably, transplantation of epithelial Py2T cells results in the formation of invasive primary tumors with low to absent E-cadherin expression, indicating that the cells undergo EMT-like changes in vivo. This process appears to at least in part depend on TGFb signaling, since tumors formed by Py2T cells expressing a dominant-negative version of TGFβ receptor widely maintain their epithelial differentiation status.

Conclusions/Significance: Together, the data demonstrate that the Py2T cell line represents a versatile model system to study the EMT process *in vitro* and *in vivo.* The observation that Py2T cells give rise to tumors and collectively undergo EMT-like changes in vivo highlights the suitability of the Py2T model system as a tool to study tumor-related EMT. In particular, Py2T cells may serve to corroborate recent findings relating EMT to cancer cell stemness, to therapy resistance and to tumor recurrence.

Institute of Biochemistry and Genetics, Department of Biomedicine, University of Basel, Switzerland

Plos One PLOS ONE Dec. 2012, Vol. 7, Issue 12, e52637 IF 4,0

Inactivation of MARCH5 Prevents Mitochondrial Fragmentation and Interferes with Cell Death in a Neuronal Cell Model

Lei Fang[1], Charles Hemion[1], David Goldblum[2], Peter Meyer[1,2], Selim Orgül[2], Stephan Frank[3], Josef Flammer[2], Albert Neutzner[1,2]

Abstract

Purpose: To study the impact of the mitochondrial ubiquitin ligase MARCH5 on mitochondrial morphology and induction of apoptosis using an in vitro model of neuronal precursor cells exposed to glaucoma-relevant stress conditions.

Methods: RGC5 cells transfected with expression constructs for MARCH5, MARCH5^{H43W}, Dpr1^{K38A} or vector control were exposed to either elevated pressure of 30 mmHg, oxidative stress caused by mitochondrial electron transport chain (ETC) inhibition, or hypoxia-reoxygenation conditions. Mitochondrial morphology of RGC5 cells was analyzed following staining of the mitochondrial marker cytochrome c and photoactivatable GFP (PAGFP) diffusion assay. Induction of apoptotic cell death in these cells was determined by analyzing the release of cytochrome c from mitochondria into the cytosol and flow cytometry.

Results: Exposure of RGC5 cells to oxidative stress conditions as well as to elevated pressure resulted in the fragmentation of the mitochondrial network in control cells as well as in cells expressing MARCH5. In cells expressing inactive MARCH5^{H43W} or inactive Drp^{K38A}, mitochondrial fragmentation was significantly blocked and mitochondrial morphology was comparable to that of control cells under normal conditions. Exposure of RGC5 cells to elevated pressure or oxidative stress conditions induced apoptotic cell death as assessed by cytochrome c release and DNA staining, while expression of dominant-negative MARCH5^{H43W} or Drp1^{K38A} did significantly delay cell death.

Conclusion: Preventing mitochondrial fragmentation through interference with the mitochondrial fission machinery protects neuronal cells from programmed cell death following exposure to stressors physiologically relevant to the pathogenesis of glaucoma.

[1] Department of Biomedicine, University Basel, Switzerland
[2] Department of Ophthalmology, University Basel, Switzerland
[3] Division of Neuropathology, Institute of Pathology, University Basel, Switzerland


The Editorial Team of DBM Facts wishes
all its readers a swinging spring time!

DEPARTEMENT BIOMEDIZIN HEBELSTRASSE

Lorenz Joerin
Clinical Pharmacology

Henriette Kurth
Medical Genetics

Sonia Lebboukh
Cardiobiology

Riccardo Mancuso
Clinical Pharmacology

Jana Orellana
Molecular Nephrology

Ausserdem haben angefangen:

DEPARTEMENT BIOMEDIZIN HEBELSTRASSE

Martine Meichsner
Med. Genetik
Clarisse Wermuth
Med. Genetik
Friedel Wenzel
Med. Genetik
Sven Cichon
Med. Genetik
Stefan Herms
Med. Genetik
Per Hoffmann
Med. Genetik
Peter Miny
Med. Genetik
Marianne Häusler
Med. Genetik
Margrit Bernoulli
Med. Genetik
Joëlle Jenni
Med. Genetik
Daniela Thommen
Cancer Immunology
Urs Fisch
Brain Ischemia and Regeneration
Matthias Kreutz
Tissue Engineering
Christoph Renner
Cancer Immunology
Frank Stenner
Cancer Immunology
Adeline Stiefvater
Neuromuscular Research
Thierry Nordmann
Diabetes Research
Tatiana Pochechueva
Gyn. Research
Bernhard Rieder
Tissue Engineering
Valentina Strusi
Tissue Engineering
Merete Bock
Clin. Immunology
Sarah Brunner
Neurobiology
Elizaveta Fasler-Kan
Gastroenterology
Magdalena Filipowicz Sinnreich
Hepatology
Emanuela Janner
Clin. Pharmacology
Dominik Schnell
Clin. Pharmacology
Kerstin Wagner
Immunobiology
Reto Kohler
Gyn. Research
Andreas Schötzau
Gyn. Research

DEPARTEMENT BIOMEDIZIN MATTENSTRASSE

Stefania Fidele
Neuronal Stem Cells
Chiara Rolando
Neuronal Stem Cells
Zahra Ehsaei
Neuronal Stem Cells

DEPARTEMENT BIOMEDIZIN PETERSPLATZ

Aline Burchardt
Infektionsdiagnostik
Bettina Oettli
Infektionsdiagnostik

WENN EIN MENSCH GÜTIG UND HÖFLICH IST, BEWEIST ER, DASS ER EIN WELTBÜRGER IST.

WHEN SOMEONE IS KIND-HEARTED AND POLITE HE SHOWS THAT HE IS A COSMOPOLITAN.

CHI È BENEVOLO E BEN EDUCATO DIMOSTRA DI ESSERE COSMOPOLITA.

(SIR FRANCIS VON VERULAM BACON)

Congratulations

Ewen Sesia
Geboren am 31.03.2013

Das DBM gratuliert ganz herzlich!

Anaé Mollé-Grisouard
Geboren am 10.02.2013

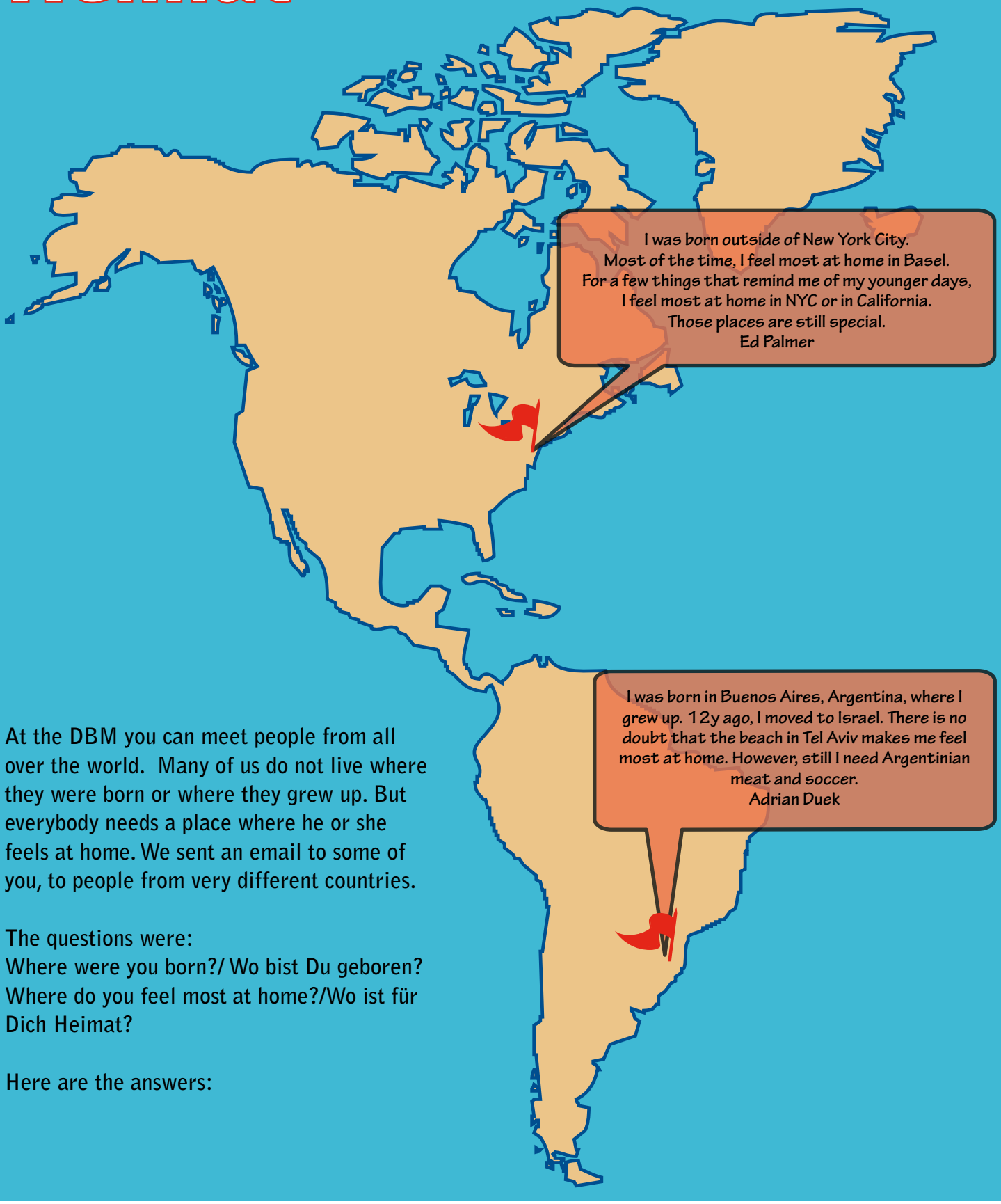
Derin Eken Bolkan
Geboren am 31.03.2013

Herzlich willkommen, allerseits!

Lucie Meira Bokalot
Geboren am 25.11.2012

Michal Stepanek
Geboren am 27.02.2013

Heimat



At the DBM you can meet people from all over the world. Many of us do not live where they were born or where they grew up. But everybody needs a place where he or she feels at home. We sent an email to some of you, to people from very different countries.

The questions were:
Where were you born?/ Wo bist Du geboren?
Where do you feel most at home?/Wo ist für Dich Heimat?

Here are the answers:


I was born in Kalinin (now Tver, Moscow region); grown up in Izhevsk (native town of Kalashnikov).
For me, It is not an exact place, but the weather and nature, that make me feel at home. When I see white, snowy landscape and/or birch trees I feel at home.
Artem Kalinichenko
Mein Geburtsort ist Bratislava.
Heimat ist da, wo ich mich wohl fühle.
Radek Skoda
I come from Nanjing, PR China.
When I sit together with a group of Chinese friends to have a traditional Chinese Cuisine, and to chat about what going on in my hometown, I feel at home.
Lei Fang
Ich bin in Zürich geboren.
Die Heimat ist für mich die Region Basel, wo ich aufgewachsen bin und heute noch mit meiner Familie lebe.
Nicole Kessler-Uehlinger
Where I am born: Togane city in Japan
Where I feel most at home: When I arrive at my parent's home in Japan and when I come back to my home in Rümmingen (Germany)
Noriko Shikama
Place of birth: Karaman (Turkey), grown up in Kaysersberg (Alsace / France).
When I drive trough the vineyard in Alsace valley towards the hills of the Vosges mountains, I feel at home.
In french:
Née à Karaman (Turquie), j'ai grandi à Kaysersberg (Alsace / France).
Sur la route des vins d'Alsace, en ballade vers la ligne bleue des Vosges, je me sens chez moi.
Leyla Razik
I was born in Zimbabwe, Africa
I carry my home in my mind. When I shut my eyes and imagine a peaceful concoction of endless horizons, warm sunshine, smells of clean dust, tastes of falling rain and the sounds of birds and crickets, that is when I feel most at home.
Thérèse Resink

Have a Parisian day

Paris is one of the first world tourist destinations. Why is that? Probably because Paris has the great reputation of being the most beautiful and romantic of all cities, and I believe Woody Allen's movies partly built up this popularity. Paris is also filled with history and has numerous famous landmarks that everybody is familiar with. Dreaming of Paris, one will think of the Eiffel Tower, The Louvre Museum or Notre Dame Cathedral, and every good guidebook will tell you everything you need to know. But beyond those "clichés", Paris is a big city that is full of secret places and each of its districts shows different Parisian social scenes. The city is highly influential in the fields of culture and art and you will find countless museums from antique to modern arts. Paris is also the capital of "French cuisine", with fine restaurants and wine-tasting bars that are just waiting for you! The city is divided into 20 districts or "arrondissements" (arr.) forming a snail pattern beginning from the first in the centre. As Paris was built up on many small hills along the River Seine, you will often hear the word "Butte", meaning hillock. Now, I will tell you a little bit about my favorite spots in the city.

If you visit Paris, you cannot miss the old Montmartre district that spreads at the back of the "Sacré Cœur" (Sacred Heart) basilica in the 18th arr., that is the highest place in Paris. There, you have one of the most stunning panoramic views of Paris, with all its main monuments easily distinguishable in front of you. It is a beautiful area but it is also one of the most touristic places of the capital. The "place du Tertre" is the heart of Montmartre, with its lively square terraces and open-air restaurants along with a permanent exposition of many artists. Here, a village ambiance unfolds within the big city. A few streets away, you can discover the "rue des Saules" that borders the last vineyard of the "Butte Montmartre". In addition to these famous places, take some time to stroll along the lovely village streets, until you reach the "rue des Abbesses", which is a very nice neighborhood to do a little bit of shopping.

Further to the South, the Marais is one of Paris' oldest and "so Parisian" districts (3rd and 4th arr.). There, you will go down some narrow streets that typically recall architectural styles of the Middle Age and Renaissance eras. It is also one of the most surprising areas gathering different populations.

At first, you will find lots of small design, vintage and fashion stores as well as art galleries. Then, you will notice that the Marais is also the gay-friendly district in Paris and you may enjoy the hustle and bustle of its bars and typical cafés. And finally, you will observe that this neighborhood is also hosting one of Paris' oldest Jewish communities. If you are hungry, I will advise you to go to the "rue des Rosiers" where you can take away some tasty Falafel! Or you might walk up to the "Marché des enfants rouges", the oldest food market in Paris. It is a pleas-

ant indoor market that sells ready-to-eat cuisine from all over the world. Close to the Marais, the Pompidou Centre is the largest museum of modern art in Paris, designed by the famous architect Renzo Piano and presenting amazing art exhibitions that you might want to discover! From there, you can easily reach the "canal Saint Martin". There, walking along the river, you will find nice cafés where you can have a Sunday morning brunch! At night, you can relax enjoying a glass of wine by the water's edge at the typical Parisian bar "Chez Prune"; the outdoor terrace is perfect!

Enjoying a sunny day in Paris, you can relax in one of the beautiful public parks within the city. The two largest areas are the "parc Montsouris" (14th arr.) and the "parc des Buttes Chaumonts" (20th arr) where all Parisians love to spend their weekends during summer time. A nice walk to do is the "Promenade Plantée", also known as "Coulée Verte", literary "green course", that stretches for 4,7 km long through the 12th arr. up to the astonished wood of Vincennes. It actually follows a railway dating back from the 19th Century, which was recently transformed into a flowery pedestrian zone. From 7 meters up, you will enjoy walking alongside hazel trees, weeping willows and bushes of roses. Another idea for a sweet picnic is the "Port de l'Arsenal" garden in the 11th arr.. It is a unique place in Paris. It houses the only marina of the city where pleasure boats are moored all year round and it is surrounded by beautiful pergolas decorated with flowers. This garden is a delightful place to rest after a visit in the crowded Bastille neighborhood nearby.

Although less famous, la "Butte aux Cailles" is another village-like area in Paris (13th arr.). Nearly all of the action in this little residential neighborhood can be found on the "rue de la Butte aux Cailles". In the spring and summer times its bars overflow onto the sidewalk with Parisians sharing drinks. It also gathers a high concentration of restaurants proposing regional specialties from all over France. Notice on the walls, the funny street art graffiti from the local artist MissTic. Last but not least, one cannot go to Paris without visiting one of the countless bakeries. The choice will be hard! But if you want to taste the best macaroons in town, the typical French almond-based airy pastries, you must go to Pierre Hermé's boutique in the swanky neighborhood of "Saint Germain des Près". It is delicious, it is heaven!

Paris is a huge city and there are too many things to see and to do to be able to talk about everything! And during summer, there are a lot of special events or music festivals (rockenseine.com or welovegreen.fr) inside the city that you might want to check for. One word of warning however: compared to Basel, Paris is definitely not the safest place to visit. Be careful whenever you visit crowded places or take the metro. Finally, I will let you decide whether French people live up to their reputation of rudeness!

Elise Dalmas

Le Marche des Enfants Rouges:
39 rue de Bretagne – 3rd arr.

Chez Prune:
36 Rue Beaurepaire – 10th arr.

Promenade plantée's starting point:
44 rue de Lyon – 12th arr.

Port de l'Arsenal garden:
53 boulevard de la Bastille – 12th arr.

Pierre Hermé's bakery:
72 Rue Bonaparte – 6th arr.

Inglorious Club of Football addicted Sscientists 2011

The ICFS 2011 publishes this report aimed at i) recruitment of further study volunteers and ii) briefly reporting on recent study results based on investigations during the last two years.

Introduction

Football refers to a number of sports that involve kicking a ball with the foot to score a goal. The most popular of these sports worldwide is association football, more commonly known as just "football" or "soccer". Unqualified, the word *football* applies to whichever form of football is the most popular in the regional context in which the word appears, including association football, as well as American football, Australian rules football, Canadian football, Gaelic football, rugby league, rugby union[1] and other related games. These variations of football are known as football codes.

Various forms of football can be identified in history, often as popular farmer games. Contemporary codes of football can be traced back to the codification of these games at English public schools in the eighteenth and nineteenth century.[2] The influence and power of the British Empire allowed these rules of football to spread to areas of British influence outside of the directly controlled Empire,[3] though by the end of the nineteenth century, distinct regional codes were already developing: Gaelic Football, for example, deliberately incorporated the rules of local traditional football games in order to maintain their heritage.[4] In 1888, The Football League was founded in England, becoming the first of many professional football competitions. During the twentieth century, the various codes of football became amongst the most popular team sports in the world.[5]

Though several codes and adaptions developed over time, those various codes of football share common elements. Generally two teams of usually 11 to 18 players play the game on a clearly defined area (field). The aim is to score goals or points by means of moving the ball to the opponents' goal area or over a defined line. Goals are achieved by moving the ball between two goalposts – depending on the code – by kicking, carrying, or hand-passing the ball.

In most codes, there are rules restricting the movement of players *offside*, and players scoring a goal must put the ball either under or over a *crossbar* between the goalposts. Other features common to several football codes include: points being mostly scored by players carrying the ball across the goal line; and players receiving a free *kick* after they *take a mark or make a fair catch*.

Peoples from around the world have played games which involved kicking or carrying a ball, since ancient times. However, most of the modern codes of football have their origins in England.

This article aims at the detailed investigation of the influence of several types of football – focusing mainly on indoor and outdoor football- on the behavior and health of scientists and the impact on their daily work.

Materials and Methods:

leather ball

Ball:

A ball is a spherical, round object intended for many purposes. It might consist of solely one material, e.g. rubber or might be composed of several materials adding special properties to the ball. A football in general consists of an inflatable inner rubber ball and an outer shell commonly made of leather or polymeric materials. This multi-layered design enables the ball to withstand forces exerted onto it (shooting) and at the same time avoids injuries of the players.

Players:
The players are the main figures in this game. They are usually human and are not supposed to be modified in any way (i.e. genetically or mechanically). Players of one team should be easily distinguishable from the players of the other team. Here the study group consists of males with the age of mid-20s to 30s working in the broad field of science.

Field:
The field size and nature are almost arbitrary. Usually a rectangular shape is preferred. Depending on the season, fields can be located outdoors (grass or hard court) or indoors (gyms). The existence of goals, goal areas or goal lines is indispensable.

Injury assessments:
Injury assessment and if needed first aid is carried out by trained personnel, i.e. MD's being members of the ICFS 2011.

Communications skills:
The alteration of communication skills is assessed through loudness measurements by scoring inner ear pain.

Behavioral alteration assessments:
Behavioral changes were assessed by interrogating all players available at each day of practice. As behavioral baseline, the day-to-day behavior in a familiar environment was used (i.e. laboratories in the DBM).

Results

This study reveals outstanding insight into behavioral changes based of emotions occurring during attending a football match. An obvious observation was the place of birth-depending metamorphism of players during the matches. In particular, specimens originating from the southern territories of Europe, tend to morph into loud screaming, unceasingly arguing players who lose their focus. In contrast, specimens originating from the middle to northern parts of Europe – though northern European specimens are missing – tend to silently enjoy their time spent among friends. With regards to injuries, the specimens seemed to be outstandingly lucky. Other than some smaller injuries including hematomas, bruises and strain traumas only one slightly more severe injury occurred, thatbeing splintering of the shinbone.

Conclusion

Football helps scientists in several categories to improve their health, attitude, reactivity and communication as well as anticipation skills. These improvements will not only help advancing their skills related to football, but will also undoubtedly impact their daily working lives. The margin of improvement spans from increased ability to concentrate due to the total exhaustion leading to better and deeper sleep, to an extended stay in their laboratories due to the inability of moving based on severe muscle hangovers as well as the formation of friendships improving team-work attitude.

References

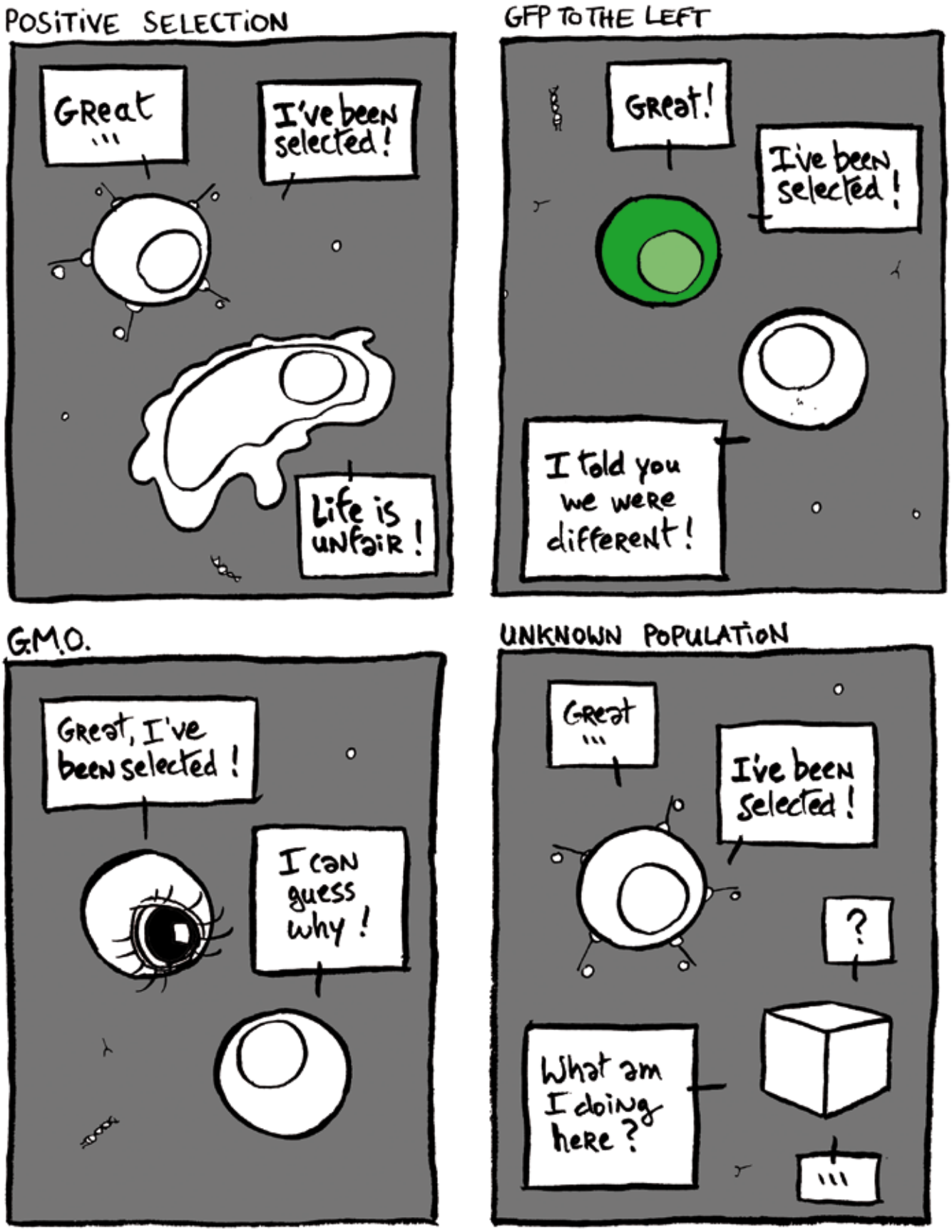
1. Reilly, Thomas; Gilbourne, D. (2003). "Science and football: a review of applied research in the football code". Journal of Sports Science 21: 693-705.
2. Bailey, Steven (1995). "Living Sports History: Football at Winchester, Eton and Harrow". The Sports Historian 15 (1): 34-53.
3. Perkin, Harold (1989). "Teaching the nations how to play: sport and society in the British empire and commonwealth". The International Journal of the History of Sport 6(2): 145–155.
4. Reilly, Thomas; Doran, D. (2001). "Science and Gaelic football: A review". Journal of Sports Sciences 19 (3): 181–193.

ICFS 2011 was founded on 11th November 2011 in order to create a community within the DBM that shares some free-time (once a week every Tuesday) in order to join the wonderful game called football. If you are interested in joining the club as a study volunteer please contact one of the following persons:

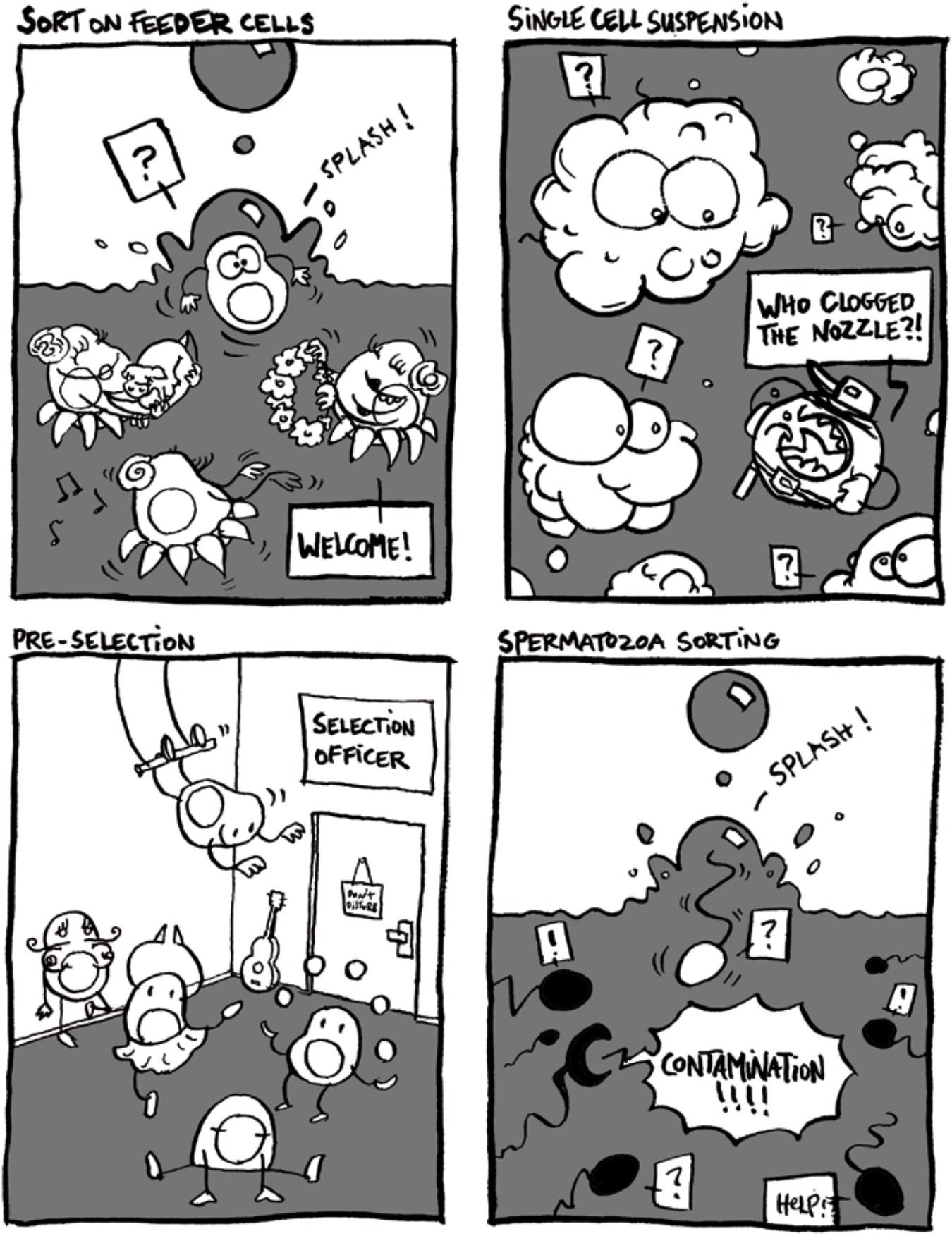
Waldemar Hoffmann, Evangelos Panopoulos, Emanuele Trella

Waldemar Hoffmann

FACS & FICTION



from Emmanuel Traunecker



+ IT News +++ IT News +++ IT News ++

Would you like your own version of Windows 8? If so then you can download it for free at http://e5.onthehub.com (Windows Server for Educational Institutes). Choose the option “Download Windows 8”. After that select country “Switzerland” and then “Universität Basel”. You can also download other operating systems. This is only possible if you have an email address ending with @unibas.ch.

Do you need your own copy of Microsoft office? For home use Mac 2011 and/or Win 2013 can be downloaded from http://hup.microsoft.com/. Enter your email adress ending in @unibas.ch and code 7E0C151BDB. Both the Windows Plus 2013 and the Mac versions cost €19.45.

If you require help then please visit our support server http://gadget.dfusb.unibas.ch. You will find lots of guidance and news on many topics there.

At the moment we cannot offer installation of Windows 8. There is no problem with someone using a preinstalled version of Windows 8 on a PC. However, we do not as yet have a licence server in service that would allow us to install new copies of Windows 8 with the University licence.

Christian Hirt, Oncology Surgery

Running after caps and hats...

I can describe my life in one sentence; running after caps and hats. That may sound strange – what does a PhD student have to do with hats or caps? The official PhD one is still ahead, but I would like to tell you about the other ones that once covered my head...

The first one I suppose was a **straw** one – I grew up in the canton of Aargau, where straw hats have been produced in the Freiamt for the last 300 years. The Aargau village, Wohlen, was even called "little-Paris" in the 19th century. Today, Aargau is the largest industrial region in Switzerland with its main industries being engineering and life sciences (ABB, Alstom, Roche, Johnson & Johnson etc.). Aargau has a very interesting history with parts belonging once to Bern, Zurich, and even Austria. The famous Habsburg Dynasty, which ruled Eastern Europe for centuries, had their castle near Brugg. As for myself, I grew up in the hamlet of Rütihof in Gränichen, which is a village near Aarau and is one of the largest villages in the Aargau. By living in the countryside, with forest on one side and straw, grass, and cornfields on the other, I can describe it as a place where foxes and rabbits still say goodnight to each other.

After my obligatory school time with first **baseball caps,** I moved to the high school or "Kantonsschule" in Aarau (which as an aside was the first capital of Switzerland). My school, the "Alte Kantonsschule Aarau", or AKSA for short, was where Albert Einstein did some of his early schooling at the end of the 18th century, before he entered the ETH in Zurich. His final exams are still in the archives with his best marks being in physics and maths and his worst in French.

At the AKSA I got my second hat by joining the Kantonsschülerturnverein or KTV Aarau, a former gymnastics group, which is now a fraternity. Fraternities here in Switzerland are not secret organizations, nor are they political, but rather a place for socializing and drinking good beer! Each fraternity has it's own hat, called the **"Couleur",** and a specific two-to-three-color-band. My KTV-time was followed by the "Rhenania" fraternity at the University of Bern. This fraternity is the oldest in Switzerland and dates back to 1816. It was formerly a gymnastics organization, but today is a fencing society. An in-depth description of all the rules and costumes of this fraternity is beyond the scope of this article; however what I can tell you is that fraternities are not outdated, archaic institutions, but rather places where people of all ages come together for camaraderie and social activities.

Coming from a family with a pharmacologist, (my grandfather was able to develop his own creams and pills!) I was always interested in research that could benefit people. This desire led me to study medicine in Bern, where I received a comprehensive overview in many different specialties. Bern is a UNESCO World Heritage Centre that has a unique charm

KTV Maienzug 2000 – AKSA, Aarau

Emergency Station St. Cruz 2009 – Galapagos, Ecuador (Christian Hirt back row 3rd from the left).

and relaxed atmosphere. During this time I was able to combine studying with travelling and had several small internships in clinics throughout Switzerland such as Visp in the southwest, Basel in the north, Lucerne in the center, and Mendrisio in the south.

I did not accumulate as many hats during my internships in Switzerland; however during my internship in the Ural mountains and Arkhangelsk (in Northern Russia) I was introduced to another cap: the **surgical cap** of Russia (with its cylindrical form it is unique to the East). It seemed to me that the higher the cap, the higher the person was in the medical hierarchy, but this was just speculation. Another hat that I came across during my internship was in Ecuador; where I spent time doing clinical rotations in pneumology, traumatology, and emergency medicine. Ecuador is a beautiful, small Andean country situated between Peru and Colombia to which the Galapagos Islands belong. The iconical **Panama hat** is still produced there and was used as sun protection for the thousands of workers who built the Panama Canal.

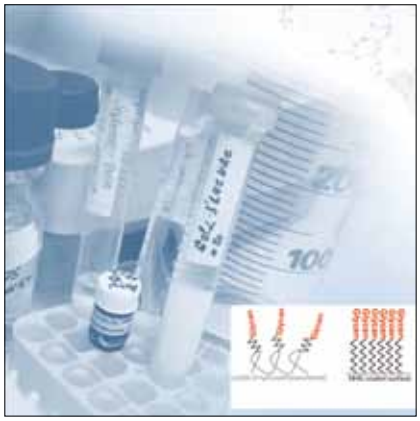
Besides my medical studies, I had to spend some time in the army. It was here that I was introduced to another form of head cover, called the **"Beret".** Switzerland has a militia system for its troops, meaning that everybody is conscripted to the military. Upon joining, a cadet will normally do intensive training, also known as the "Rekrutenschule" and then afterwards, an annual monthly training session until the person reaches 30–35 years. Interested in technicals and tactics, I joined the tank forces and also decided to do officer training. For some years now I have been a captain and have led a tank company of over one hundred men for one month every year. This has helped me develop skills in leadership and organization.

The direct and methodological approach of surgery brought me to the Emergency Department of Basel's University Hospital. The Emergency Department is the largest in the German-speaking part of Switzerland with an annual admission of approximately 45,000 patients. After my initial training, I received a residency in visceral surgery at the St. Claraspital. This hospital, which is situated behind the Badischer Bahnhof, is a private hospital belonging to the Ingebohl Monastery where few nuns still work to this very day! It is smaller than the University Hospital of Basel and has many different specialties, but with a strong focus on gastrointestinal diseases and their treatments. Under the **surgical mask, or cap,** I spent several hours sweating and learning the basic surgical skills required for a surgeon.

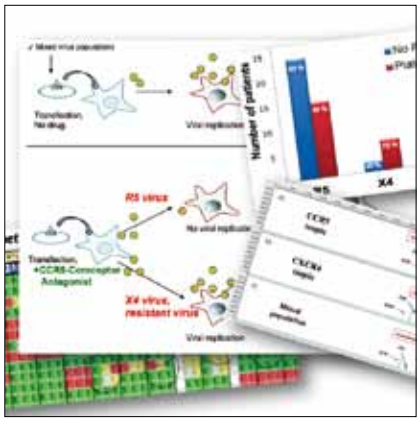
Although I enjoyed working in surgery and seeing patients, I soon realized that if I wanted to dedicate time and training to research, a deeper understanding in that area would be required. Specifically, I wanted to do translational research that linked the "bench to the bedside". My main interest was, and still is, cancer immunotherapy and fortunately, I started my PhD with the oncology surgery group. Of course the patient contact is still missing, but the world of science is full of challenges. The international atmosphere both within the group and department is also a great experience, and has sparked my interest to one day work abroad. Hopefully, I will be able to continue along this path and maybe pick up a few other hats along the way... stay tuned.

VORSCHAU PREVIEW

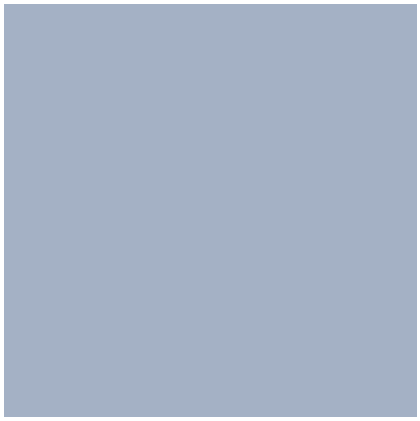
In der nächsten Ausgabe . . .



. . . stellt das Labor Gyn. Research seine Forschung vor

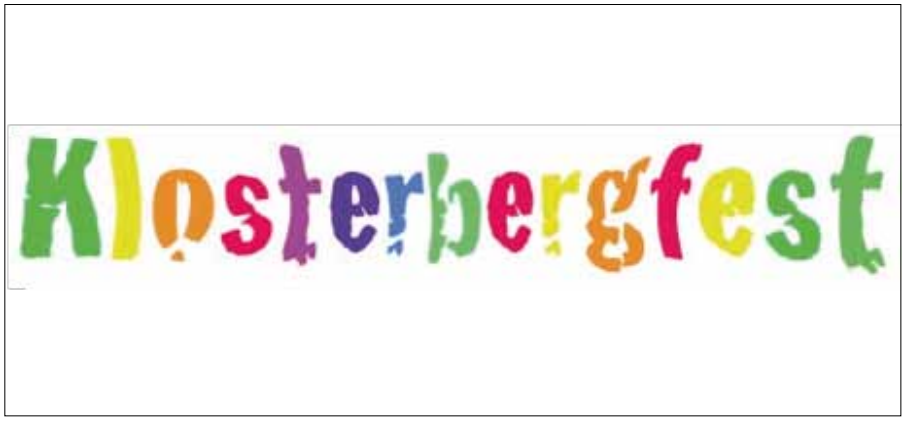


. . . erfahren wir von Thomas Klimkait mehr über Molecular Virology



. . . lernen wir mit Jana Orellana Sevilla kennen

. . . treten wir mit Marc Bichsel in die Fussstapfen von Roger Federer



. . . stellen wir die schönsten Feste im Umland vor

Magie

Aus unbeschreiblicher Verwandlung stammen
solche Gebilde-: Fühl! und glaub!
Wir leidens oft: zu Asche werden Flammen;
doch: in der Kunst: zur Flamme wird der Staub.

Hier ist Magie. In das Bereich des Zaubers
scheint das gemeine Wort hinaufgestuft…
und ist doch wirklich wie der Ruf des Taubers,
der nach der unsichtbaren Taube ruft.

Rainer Maria Rilke
(Muzot, August 1924)