FUTURA

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Data Deluge
How is Big Data transforming science and society?

Projects, Results, MD Fellowships
New PhD projects, completed theses, and MD fellowships

A BIF Fellow's Guide
Discover Aarhus, Denmark's second largest city

The cover illustration alludes to the Big Data development that is expected to have a major impact on society in the years to come. The amount of data collected today is growing exponentially and with it come new ways of interpreting and linking datasets. Read more on how Big Data will transform the scientific world on page 12.

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PAPERS IN THE SPOTLIGHT

PUBLISHING INFORMATION

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Stiftung für medizinische Grundlagenforschung
Schusterstr. 46-48
55116 Mainz
Germany
Tel. +49 6131 27508-0
Fax +49 6131 27508-11
E-mail: secretariat@bifonds.de
www.bifonds.de

Editor-in-Chief Dr Claudia Walther

Editors Kirsten Achenbach (BIF, executive editor), Karsten Fiehe (muehlhausmoers corporate communications gmbh)

Authors in this issue Kirsten Achenbach, Lily Dayton, Timandra Harkness, Dr Claudia Walther

Translating, copy-editing and proofreading Adam Blauhut, Dr Caroline Hadley, Dr Susan Simpson

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Project management Karsten Fiehe
Art direction Britta Siebert

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THE BEST OF BOTH WORLDS

»As a welcome side-effect, the travel grants also foster international cooperation.«

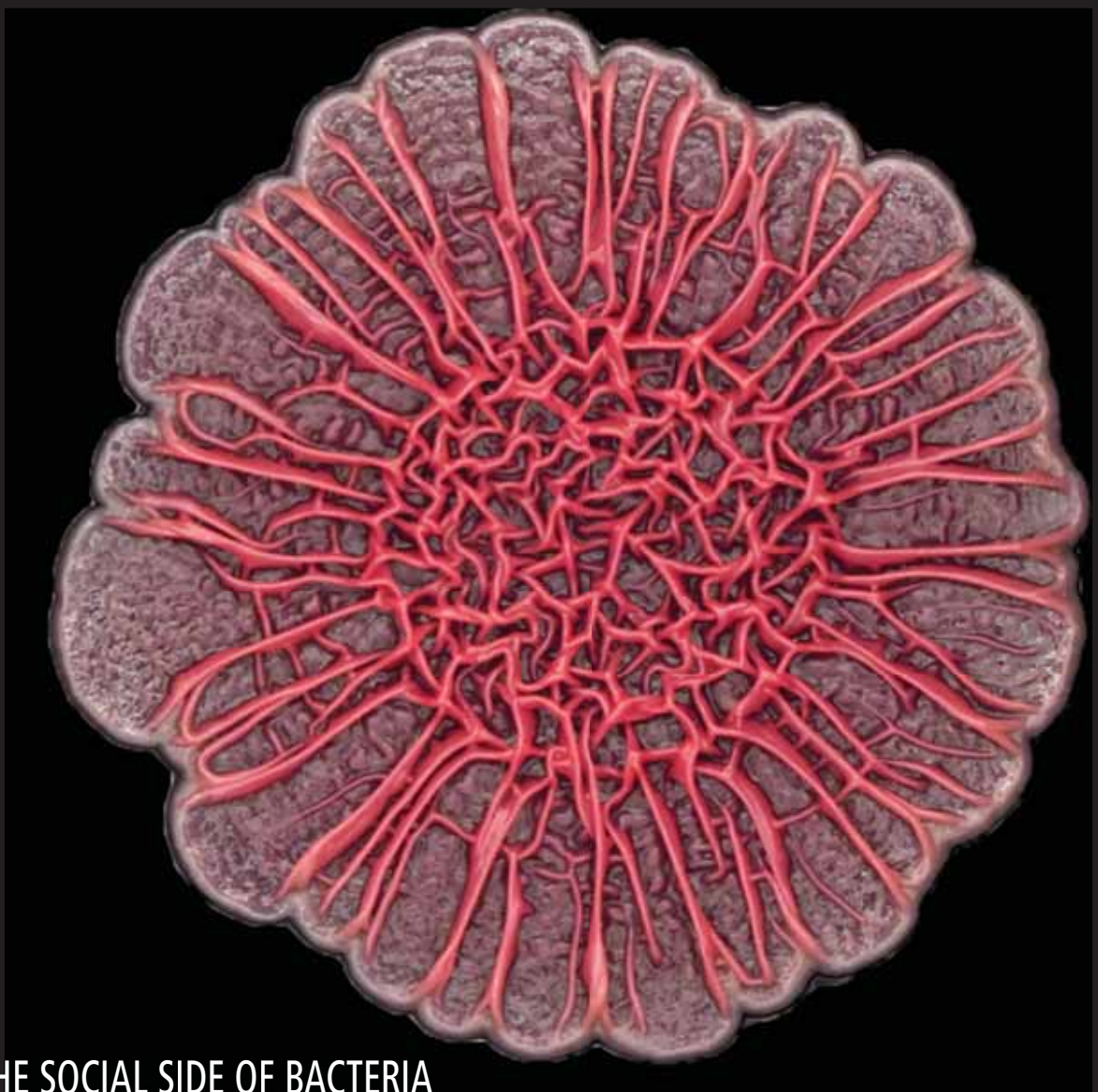
Often enough technical advances drive research and medicine. Take the microscope or Nobel laureate Robert Koch's new staining and culturing methods, for example. They enabled him to identify, among others, the bacterium that causes tuberculosis and eventually led to his four famous postulates and to modern bacteriology. An impressive recent example are Emmanuelle Charpentier and Jennifer A. Doudna, who fundamentally changed gene editing by harnessing a defense trick of bacteria into the so-called CRISPR-Cas systems.

No question: Disseminating such new techniques as quickly as possible aids the progress in science. But even in the age of the Internet some things have to be taught in person. Be it because sometimes research still is a handicraft, not learnable by merely reading a protocol, or because not every piece of equipment is available at one's institute. BIF therefore offers travel grants to postdoctoral fellows, PhD, and MD students for learning methods important to their ongoing research projects. The grants support visits of up to three months in other laboratories or participation in practical courses. Due to its purpose, the programme aims to support not only the top 5% but also many junior scientists. Given the current number and quality of applications, nearly every second applicant can expect to be funded.

With regard to internationality, the travel grants top the list of BIF's funding programmes. Last year, we supported some 120 scientists from 30 different countries. More than 75% of the awardees did not hold a German passport, and nearly all junior researchers changed countries – more than half even changed continents. As a welcome side-effect, the travel grants thus also foster international cooperation.

Awardees usually learn much more than laboratory techniques. The visit may offer the opportunity to work independently for the first time; it may inspire new ways of thinking or may spur fascination for a new subject. On a more practical note, a short stay abroad may tip the scale on deciding whether to do a postdoc outside one's home country, as one PhD student remarked. And a young woman from Germany wrote how very encouraging it was to experience that at an institution like Harvard Medical School in Boston it is deemed perfectly normal and socially acceptable to have young children and work as a researcher.

However, the main purpose of BIF's travel grants remains to enable junior researchers to learn and use the most suitable methodology for their projects and to help spread improved or novel techniques that promise advances. We hope that many postdocs and PhD students will share the experience of one awardee: "the input was enormous and I was exhausted […], but it was worth every minute."



THE SOCIAL SIDE OF BACTERIA

By Lars Dietrich, Columbia University, New York, USA

Bacteria are not just autonomous, single-celled organisms. They can communicate with each other via small signalling molecules (quorum sensing) and form multicellular communities (biofilms). This image depicts a two-centimetre colony biofilm of the pathogen *Pseudomonas aeruginosa* PA14, grown on the surface of an agar plate. Each colony contains roughly five billion cells that are embedded in a matrix. Under low oxygen conditions the colonies form flower-like structures by spreading on the surface of the agar plate and forming pronounced wrinkles. This morphogenesis may be an adaptation to increase access to oxygen.

We are always looking for exciting scientific photos and illustrations! If you would like to have your image published, contact Kirsten at kirsten.achenbach@bifonds.de.

FOREIGNERS NOT WELCOME HERE

Wasps may have tiny brains, but they sure do amazing things with them. As a recent study discovered, a species of tropical wasps, *Liostenogaster flavolineata,* can memorize the faces of members of their colony and will attack any individual with an unfamiliar face. Each nest of this tiny South East Asian wasp contains a family of related individuals. Hundreds of these nests can be clustered together to form a kind of city. Close proximity to so many other families means each colony faces persistent landing attempts by intruders from the neighbourhood, who might steal resources or theoretically lay cuckoo eggs. *Liostenogaster flavolineata* can also tell family members by the colony-specific scent they bear, but the new study reveals a sophisticated interplay between sight and scent in the insects. When the wasps had only visual information, they were more likely to accidentally attack a friend, and when they had only odours, they were more likely to misidentify an enemy as a friend. They appeared to prioritize whether or not they recognized the face of another wasp when deciding whether to attack. By examining both visual and odour recognition in wasps, the study adds to the understanding of how animals prioritize different senses.

REFERENCE

Baracchi D, Petrocelli I, Chittka L, Ricciardi G, Turillazzi S (2014) Speed and accuracy in nest-mate recognition: A hover wasp prioritizes face recognition over colony odour cues to minimize intrusion by outsiders. *Proc R Soc Lond B Biol Sci,* doi: http://dx.doi.org/10.1098/rspb.2014.2750

ONCE MORE, WITH FEELING!

Pressing or touching plants is a centuries-old practice in Japanese agriculture. As a kind of mechanical stress treatment, it strengthens the plants' resistance and thus improves yield. However, it also leads to stunted growth and delayed flowering. Known as thigmomorphogenesis, the process is regulated by a gene that was recently discovered by Professor Theo Lange of the Institute of Plant Biology at the Technical University in Braunschweig, Germany. Working with Dr Maria Pimenta Lange, he conducted experiments with wild-type *Arabidopsis* plants and was able to identify the gene responsible for the touch-induced response in plants. The *AtGA2ox7* gene encodes a protein involved in the catabolism of a specific plant hormone, gibberellin. *Arabidopsis* ga2ox7 loss-of-function mutants do not respond to touch by changing morphologically but grow in the same way as uninfluenced plants in the comparison group. Given the fact that AtGA2ox7 helps to confer resistance to salt stress and that touch can increase plant resistance to pathogens, the Braunschweig researchers now intend to study how gibberellin catabolism can be targeted to improve plant resistance to abiotic and biotic stress.

If plants are repeatedly touched, they react with morphological change.

REFERENCE

Lange MJP, Lange T (2015) Touch-induced changes in *Arabidopsis* morphology dependent on gibberellin breakdown. *Nat Plants*, doi: 10.1038/nplants.2014.25

Chimpanzees can learn and actively change grunt calls for objects such as apples.

840

MILLION

barrels of oil are thought to lie under one area of Yasuní National Park in Ecuador. Too much, unfortunately, to allow nature conversation to prevail over commercial interests. Drilling could start in 2016, endangering one of the most biodiverse regions on the planet – home to more tree and frog species than the USA and Canada combined.

Source: The Guardian, yasunimovie.wordpress.com

ASKING FOR IT

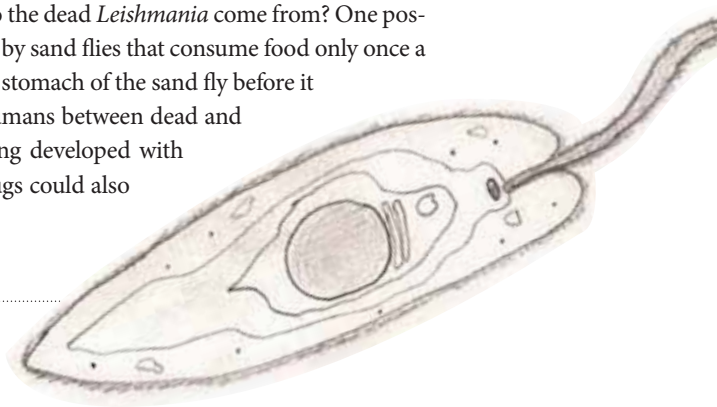
That chimpanzees and other great apes are capable of learning a form of sign language is well known. The ability to speak, however, is still thought to be exclusive to humans. Non-human primates are capable of producing alarm and food calls that refer to objects in their environment. However, researchers have so far assumed that the acoustic structures of these calls are an expression of their excitement and cannot be controlled by the chimpanzees. Now Dr Simon Townsend, evolutionary biologist from the University of Zurich, has shown with his colleagues that chimpanzees can also learn calls that refer to specific objects. In 2010, a group of adult chimpanzees from Beekse Bergen Safari Park in the Netherlands were introduced to a group of chimpanzees in Edinburgh Zoo. The researchers observed that before the integration the two groups of chimpanzees produced acoustically different grunt calls for apple. Three years and some close friendships later, the new group members had switched their calls to the ones produced by the chimpanzees already living there. According to the researchers, this is evidence that chimpanzees are capable of actively changing and socially learning the structure of meaningful, object-specific calls. These findings could also shed some light on the evolutionary origins of this basic ability.

REFERENCE

Watson SK, Townsend SW, Schel AM, Wilke C, Wallace EK, Cheng L *et al* (2015) Vocal learning in the functionally referential food grunts of chimpanzees. *Curr Biol,* doi: http://dx.doi.org/10.1016/j.cub.2014.12.032

JOINING FORCES WITH THE DEAD

The human body has a highly efficient immune system, but *Leishmania*, the parasite causing Leishmaniasis, often escapes it. Why this is so has been unclear. A research team at the Paul-Ehrlich-Institut (PEI) in Langen, Germany, has now discovered that living *Leishmania* parasites can only survive within human cells if there are also dead *Leishmania* present. In this context, a process called autophagy plays an important role. Among other things, autophagy serves to degrade viruses, bacteria, and foreign proteins. The PEI researchers discovered that dead *Leishmania* are digested by the autophagy pathway. But this process has a downside – the defence mechanism of the adaptive immune system against the living *Leishmania* is deactivated. If only living *Leishmania* enter the cell, the adaptive immune system is not inhibited. In this case, strikingly more specific immune cells of the blood, so-called T cells, are formed which ascertain that the *Leishmania* are killed. But where do the dead *Leishmania* come from? One possible explanation could be that the parasites are transmitted by sand flies that consume food only once a week. Only half the parasites may survive this period in the stomach of the sand fly before it bites a human, also explaining why the ratio in infected humans between dead and living parasites is 1:1. Active substances are currently being developed with which autophagy can be triggered or deactivated. Such drugs could also be effective in the treatment of Leishmaniasis infections.



REFERENCE

Crauwels P, Bohn R, Thomas M, Gottwalt S, Jäckel F, Krämer S *et al* (2015) Apoptotic-like *Leishmania* exploit the host's autophagy machinery to reduce T-cell 1 mediated parasite elimination. *Autophagy* **11:** 285-297

Photos: Tony Campbell/shutterstock (top left); paratryp.org (top right); wrobel27/iStock (bottom right)

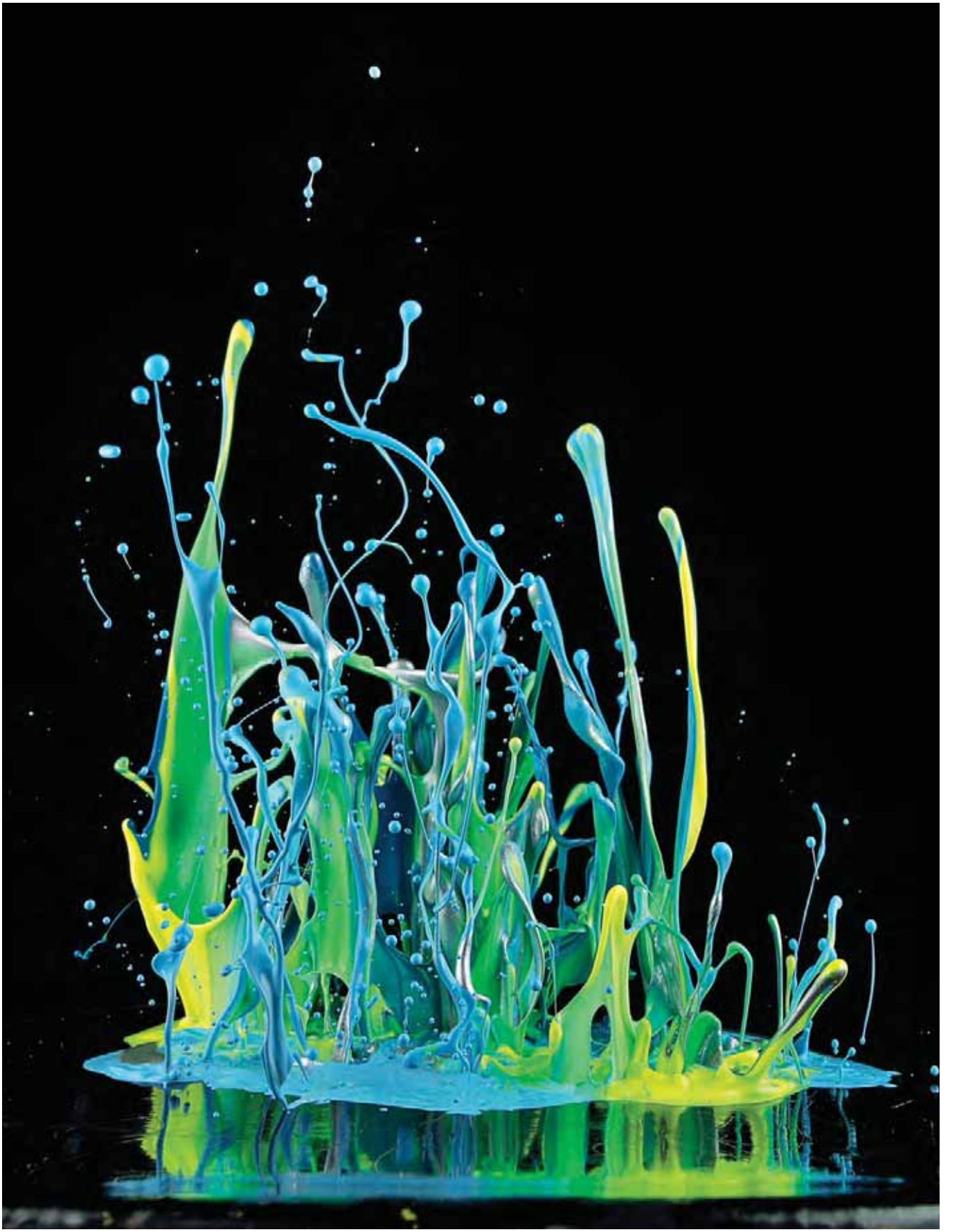
Dry lands with termite mounds survive on less rain.

A PEST IN THE HOUSE, A BLESSING OUTSIDE

House owners all over the world fear them: termites. The tiny insects come in large numbers and are capable of destroying whole houses. But termites are not pests everywhere – new research suggests that their large dirt mounds are crucial to stopping deserts from spreading into semi-arid ecosystems. In the parched grasslands and savannas of Africa, South America, and Asia, termite mounds store nutrients and moisture and, via internal tunnels, allow water to better penetrate the soil. As a result, vegetation flourishes on and near these bizarre hills in drylands that are otherwise vulnerable to desertification. The vegetation persists longer and declines slower. Ecosystems with termite mounds can survive on significantly less rain than those without them. Thus, termite mounds could make these areas more resilient to climate change. The research was inspired by the fungus-growing termite species *Odontotermes*, but the results apply to all types of termites that increase resource availability on or around their mounds.

REFERENCE

Bonachela, Juan, *et al* (2015) Termite mounds can increase the robustness of dry land ecosystems to climatic change. *Science* **347:** 651-655



Artist and synaesthete Martin Klimas pours paint on loudspeakers to make it dance in an approximation of his synaesthetic experience.

WHAT WE **CAN LEARN** FROM **SYNAESTHESIA**

By Lily Dayton

Synaesthesia is a neurological phenomenon where a stimulus to one sense, like touch, also elicits a response in another sense, for example colour. As neurologists begin to unravel the mystery of these cross-sensory perceptions, they are discovering an in-road to understanding perception in general.

Like many people, some of Joel Salinas's earliest childhood memories involve a crayon box. But Salinas didn't simply draw pictures with the crayons inside – he interacted with each crayon on a personal level. "Each crayon had an emotion and a personality based on its colour," says Salinas, today chief resident for the Harvard Neurology Residency Program in Boston. "If I couldn't find a crayon in the box, I'd think it was because I'd hurt its feelings and it was pouting." To Salinas, the world is coloured in a spectrum of feelings, emotions, and personality traits. He associates numbers with particular colours – and the personalities of the colours also transfer to them. Even sounds and motions evoke the perception of colour. Salinas also ascribes colours and numbers to people he encounters.

It wasn't until Salinas was in medical school that he realized he perceived the world in an extraordinary way. While listening to a woman give a talk about the health benefits of meditation, a fellow student with a PhD in neuroscience commented that people who hear colours and see sounds can more easily reach a meditative state. Salinas recollects asking his friend later that night, "What do you mean? Doesn't everyone see colours with sound?" That's when he learned that he had a rare, inherited condition known as synaesthesia. Derived from Greek roots meaning "joined sensation", synaesthesia is a neurological phenomenon where a stimulus to one sensory modality kindles a percept in another sensory modality. To a synaesthete, these intersensory perceptions are immediate and automatic. But within the synaesthetic brain, there is an unusual cross-wiring of neuronal traffic, where normally separate brain areas are connected by sensory highways.

The number of sensory combinations that different synaesthetes describe is virtually unlimited – to some people, spoken words elicit different tastes on the tongue; to others, a texture on the fingertips may evoke a particular emotion. Overall, about 4% of the population experiences synaesthesia, with the most common form being coloured days of the week, followed by grapheme–colour synaesthesia, where people "see" each grapheme (letter or number) in a distinct colour. As neurologists begin to unravel the mystery of these cross-sensory perceptions, they are discovering an in-road to understanding perception in general.

Vilayanur Ramachandran, professor of neuroscience at the University of California at San Diego, was studying the phenomenon of phantom limbs in amputees in the early 1990s when he began to apply his findings to the nascent field of synaesthesia research. When Ramachandran touched the faces of people who had had their arm amputated, the amputees experienced a phantom sensation in their missing hand. "We found there was cross-wiring going on in the brain," says Ramachandran, explaining that within the somatosensory cortex, the part of the brain involved in tactile perception, lies a complete body map. The area corresponding to sensory input from the face is adjacent to the area for sensory input from the hand. Through magnetoencephalography, a technique that measures localized electrical activity in the brain, he validated a sensory re-mapping of brain areas. "If you remove →

Photo: Martin Klimas, o.T. (Paul Hindemith, "Ludus Tonalis") 2011

This alphabet shows how letters appear to the wife of graphic designer Jesse Jaren. For her, letters not only have a certain colour, but also a personality. This is a combination of grapheme–colour synesthesia and ordinal–linguistic personification. The full alphabet can be viewed in Jesse Jaren's blog "Cornfed in Seattle" (http://goo.gl/umHVNT).

the hand, no sensory input is going to the hand anymore, so the hand area hungers for sensory input. The face starts cross-activating the hand area. I started thinking about cross-activation in synaesthesia." Ramachandran and Edward Hubbard were the first to demonstrate that synaesthesia was a real perceptual phenomenon, rather than a memory association or fabrication. They devised a series of experiments in which they presented grapheme–colour synaesthetes and normal controls with a one-second view of a matrix of small black graphemes (for example, the letters P and F) in which additional graphemes (for example, the letter H) were arranged in a pattern that formed a geometrical shape, such as a triangle or square. To most people, the matrix would look like a jumble of small black letters. But to grapheme–colour synaesthetes, who perceived each grapheme in a distinct colour, the shape formed by the pattern of H-letters would "pop out" of the matrix and they could readily identify it. The researchers hypothesized that grapheme–colour synaesthesia arose from cross-activation between the areas of the brain that process graphemes and those that perceive colour. Today it's generally accepted that synaesthesia results from increased cross-talk between different brain areas, though there has been long-standing debate about whether this increased communication results from a structural increase in synaptic connections between brain regions or from diminished chemical inhibition that allows neurotransmission in one region to kindle excitation in another region.

Perhaps it's a bit of both, says Jamie Ward, professor of cognitive neuroscience at the University of Sussex. "It's been shown over the past few years that there are structural changes in the brain, but the question is how these differences develop. Synaesthesia could start as a chemical change that leads to a structural change."

In grapheme–colour synaesthetes, there are structural differences in parts of the brain involved in visual processing, such as the fusiform gyrus. But differences also extend to other regions of the brain that don't seem to be involved in either the visual processing of graphemes or colour. For example, compared with controls, grapheme–colour synaesthetes appear to have more grey matter on the lateral surface of the parietal lobes in a region called the intraparietal sulcus (IPS), an area thought to be involved with attention, working memory, and spatial representation of the external world. When grapheme–colour synaesthetes are presented with letters and numbers, they have greater fMRI activation in this region.

Comparison studies of grapheme–colour synaesthetes and non-synaesthetes show that grapheme–colour synaesthetes have a hyperactive visual cortex. If the visual cortex is stimulated with a magnet, synaesthetes show an enhanced EEG response and are more likely to report seeing visual imagery. Grapheme–colour synaesthetes are also better able to discriminate between high spatial frequency stimuli, such as textures with fine lines and colour gradations, says Nicolas Rothen, visiting research fellow at the University of Sussex. This gives them a memory advantage for words, abstract fractals, and geometric shapes (though not necessarily for autobiographical events). Recent research has shown brain differences in another kind of synaesthesia – mirror–touch

synaesthesia, a condition in which a person observing touch to another person experiences a tactile sensation on the corresponding part of his or her body. "Functionally, when we observe people being touched, we all activate certain parts of our brain within the somatosensory cortex," says Michael Banissy, professor of psychology at Goldsmiths, University of London. "This is known as our mirror–touch system. When you show mirror–touch synaesthetes videos of people being touched, they activate the same system – but they have an overactive system."

Mirror–touch synaesthetes seem to have more grey matter in the somatosensory cortex, which is involved in empathy as well as touch perception. They also appear to have less grey matter volume at the temporal parietal junction, which is thought to be involved in the ability to distinguish between the self and others. Banissy explains that it may be because mirror–touch synaesthetes have an atypical self–other representation that they blur the boundaries between themselves and others.

When Salinas took part in a synaesthesia test in Ramachandran's lab, he was surprised to find out that he could add mirror–touch to the list of synaesthesia forms he experienced. "Up until that point, it was just part of my normal subjectivity," he says. He describes the feeling as being close to real, like a ghost of touch. Fortunately, he doesn't feel actual pain if he watches someone getting hurt – but he will feel a sensation. "It can certainly catch me off guard," he says. "I was doing a trauma rotation and I remember seeing someone's arm being amputated. I didn't feel pain, but I felt distressed. The thing that I felt most was the positioning of the hand and the parts that were macerated. It felt uncomfortable."

Just as grapheme–touch synaesthetes are more sensitive to certain visual stimuli, mirror–touch synaesthetes are more sensitive to tactile stimuli on their own bodies. "When we push little pressure pads with horizontal or vertical lines against their skin, mirror–touch synesthetes are better at discriminating the different textures," explains Ward. There is also evidence that mirror–touch synaesthetes are more socially sensitive than others. They are better able to recognize facial expressions and they rank high on measures of emotional empathy. Salinas says this skill helps him a great deal in his interactions with patients. "In certain situations with patients, I'm able to understand that they might be in distress, even if they're not really communicating that through words. I'll see their facial expression and pick up on their feelings because I'm feeling it in my body."

Another quality often associated with synaesthesia in general is creativity, and Ramachandran says there may be a link. "Synaesthesia is more common in poets, artists, and novelists. It may be that the hyperconnectivity [trait is] expressed more diffusely throughout their brains, which creates the propensity to connect brain regions and may create the propensity to be more creative." In a recent study, Rothen found the prevalence of synaesthesia among art students was 7%, which is higher than the 4% prevalence for the general population. "This suggests that either synaesthetes are more creative so they tend to study the arts, or their way of thinking in the arts helps to develop the synaesthetic experience …. The question is really what causes synaesthesia."

It's been known for over a century that synaesthesia tends to run in families, and 40% of synaesthetes report a family member who also has the condition. Yet inheritance patterns are elusive, with the trait often skipping a generation or occurring in a single family in a variety of different phenotypes. "A few years ago, people thought there was a single gene for synaesthesia," says Ward. "We currently believe there are several genes that convey susceptibility, maybe multiple genes, and the genes don't specify exactly what the outcome will be."

It's been known for over a century that synaesthesia tends to run in families, and 40% of synaesthetes report a family member who also has the condition. Yet inheritence patterns are elusive.

SYNAESTHESIA AND THE ARTS

Like many other artists, the Russian painter and art theorist Wassily Kandinsky is strongly suspected to have been a synaesthete who could hear colours and see music. He reports on a performance of Wagner's opera Lohengrin in Moscow: "I saw all my colours in spirit, before my eyes. Wild, almost crazy lines were sketched in front of me."

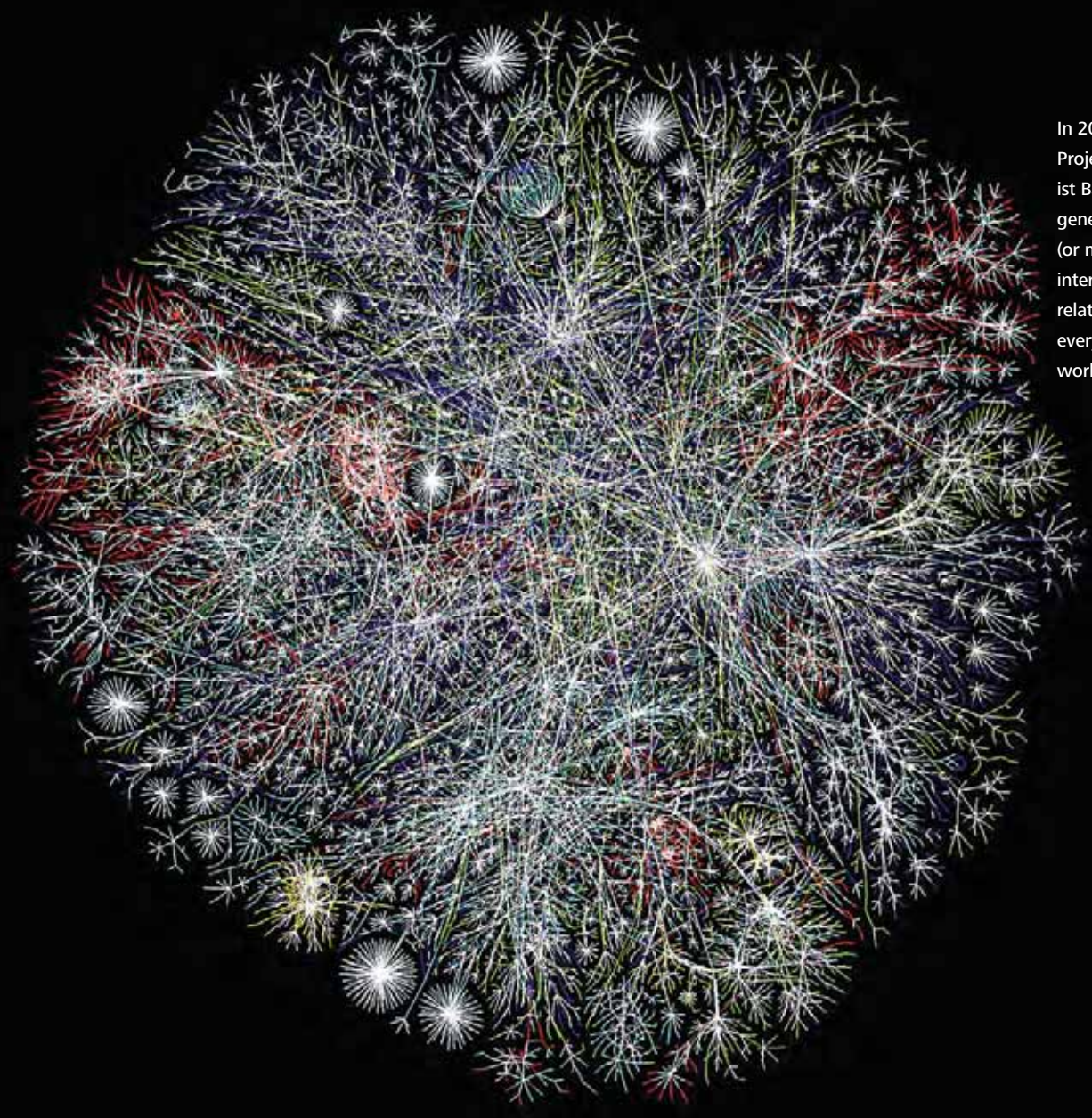
Katerina Kucera, postdoctoral researcher in the Language and Genetics Department at the Max Planck Institute for Psycholinguistics in Nijmegen, is taking two complementary approaches to searching for the genes involved in synaesthesia, in hopes that each will result in convergence on the same biological pathways. To look for the role of common genetic variants in synaesthesia, Kucera and colleagues are conducting an ongoing genome-wide association study: They are sampling DNA from a large number of unrelated synaesthetes to search for variants that are more common in synaesthetes than in the general population. Because they don't know if different types of synaesthesia share a common genetic mechanism, they are only looking at grapheme–colour synaesthetes in order to reduce genetic noise (information for potential participants can be found online at http://www.mpi.nl/synaesthesia). To look for rare genetic variants, Kucera is also currently working on an exome sequencing study, where she is sequencing the protein-encoding DNA sequences in three families with several members who have auditory–visual synaesthesia, in which sounds elicit visual experiences of colour and shape. "Right now we have some candidate variants that are shared by the synaesthetes in these families," she says, adding that they are currently validating whether the variants contribute to synaesthesia or whether they are simply shared because the family members are related. "Once we find the genes involved in synaesthesia, we will look at how variants alter proteins and the structure and function of neurons – and whether this has an effect on connectivity and signalling in the brain," says Kucera.

By learning about the physiological mechanisms and genetic underpinnings of synaesthesia, scientists are elucidating brain function in general. Isolating the genes for synaesthesia may give clues as to how genes influence brain networks – and what happens when brain circuitry goes awry. Atypical connectivity between brain regions has been implicated in a number of disorders, ranging from ADHD and autism spectrum disorder to schizophrenia, major depression, bipolar disorder, Alzheimer's disease, and post-traumatic stress disorder. Gaining an understanding of cross-talk within an otherwise healthy synaesthetic brain may someday lead researchers towards a cure for disorders that involve brain circuitry. But synaesthesia also offers a glimpse into different ways of thinking and being.

"For me, the interesting bit is using synaesthesia as a model to understand individual differences in perception," says Rothen. "Is your yellow the same as my yellow? Or is your yellow my blue? Or is it something different entirely? People understand the world in different ways. If we group them together – for example, synaesthetes and non-synaesthetes – we can begin to understand how individual differences in perception affect higher cognitive functions, such as memory and attention."

As a neurologist and a synaesthete, Salinas continues to be a scientific observer of his own life. Being constantly flooded with sensory stimulation can be overwhelming, so he's learned to experience his sensations – even uncomfortable ones – then process them and let them go. Salinas compares living with multiple forms of synaesthesia to "a practice of compulsory mindfulness." Empathizing with others and engaging with the people and objects he encounters on a multi-sensory level gives him the opportunity to be truly present with his world – and perhaps that's one thing we can all learn from synaesthesia.

Photo: Vassily Kandinsky/Composition 7/Wikimedia



In 2003, the Opte Project by artist Barrett Lyon generated a picture (or map) of the internet, showing the relationship between every routable network on the internet.

THE DATA **DELUGE**

By Timandra Harkness

Today, digital information is being created, analysed, and stored at an astonishing rate. The enormous amount of data has the power not only to transform the economy, but also science. But is Big Data also a qualitative advance on previous research methods?

Photo: iStock

If you're a researcher, you have surely noticed that the volume of data is increasing exponentially. To use the popular analogy of data as water, a dribble has become a deluge. What was a precious resource, extracted with toil and care, can now feel more like a tsunami. But in spite of the cynical definition of Big Data as "slightly more than your current computer can handle", there's more to it than size. Business analyst Doug Laney coined the "three Vs" of Big Data – volume, velocity, and variety – in 2001. The volume has increased by orders of magnitude since then, and the other two still apply. Velocity means that data is collected in real-time, automatically, from digital processes or interactions. Variety means that datasets from diverse sources, in diverse forms, can be linked. A fourth "V" is veracity – how valid is the data, how good is the quality?

But is Big Data a qualitative advance on previous research methods? Professor John D. Van Horn runs a neuroscience lab at the University of Southern California, Los Angeles, USA. When he started as a postdoctoral fellow at America's National Institutes for Health in the late 1990s, he was sent out to buy the biggest hard disc his lab could afford, to store all the data their research would generate. "People would come to our lab to look at the hard disk in awe," he says, "because it was four gigabytes! Wow! Amazing! At that time, I thought four gigabytes was infinity." Today, most cellphones have more than twice as much memory, and Professor Van Horn's lab stores "many, many petabytes of data".

In neuroscience, imaging technology progresses alongside what researchers want to ask. For instance, blood flow measurements have evolved from one full-volume brain image every eight minutes with PET scans to three or four images per second with functional MRI. But improved spatial and temporal resolution is only part of the promise of Big Data techniques, says Van Horn. He's now looking for rare genetic variants associated with brain diseases such as autism and Parkinsons by linking human genome data to his database of brain images. Finding such correlations is one of the strengths of Big Data techniques.

There's something for everybody in this field, whether you're a clinician, a researcher, or a neuroscientist. It's an exciting time to be thinking about Big Data, what it means in science, and how we can utilize it to make progress in understanding and curing diseases.

"We're looking at an object which, in the past, you had to wait for somebody to die to look at. Now we're doing that routinely, in living people. We're able to model brain form, brain function, and brain connectivity in the living person, correlate that with lifestyle, genetic, phenomic variables, and try and make associations between brain health and other things which potentially lead to disease. In order to do that, we end up with a Big Data problem." Professor Paul Matthews, head of Brain Sciences in Imperial College London's Department of Medicine, is equally excited about Big Data: "Large data is data in which we have collected lots of a single type, or a limited type, of information, and we are using it within the context that was originally envisioned. Rather than studying the size of the brain in 20 subjects, we're going to study it in a thousand. So we're scaling up a simple idea. Big Data is where we're not just collecting one or a few types of data, like brain scan, age, and gender: we're collecting all the things we can possibly think about and more, and we're looking at relationships between them that we might not have anticipated. Moreover, we're starting to link different datasets." And Professor Matthews looks far beyond just linking an individual's health records with his own research data. For example, "because you have their postal code, you can go back to meteorological records and say something about exposure to sun, or to particulates in the air, that these individuals had. So using bits of data from one dataset to link into other datasets, you suddenly have a rapidly expanding picture of a person's life."

This kind of population-scale research is relatively easy in developed countries, where so much of human life is conducted through computers. But Dr Catherine L. Moyes at the University of Oxford works on diseases that are big killers in developing countries: dengue, yellow fever, and malaria, for example. How can Big Data help here? Moyes and her colleagues aren't researching medical

WORLD CHAMPION THANKS TO BIG DATA

Big Data lurks behind Amazon's recommendations, Barack Obama's presidential re-election campaign, and even Germany's victory in the World Cup in 2014. Together with a large software company, the German Football Federation used Big Data techniques to analyse feeds from eight on-field cameras and measure key performance indicators such as the number of touches, average possession time, distance travelled, movement speeds, and directional changes. In just ten minutes, ten players with three balls produce over seven million data points. The real-time analysis enabled fast and highly individual feedback, which helped the team to cut down the average time for ball possession from 3.4 seconds in 2010 to 1.1 seconds in 2014 – an improvement that may have tipped the scale in close games such as the final match against Argentina.

cures. They model variations in disease risk, building multi-dimensional maps that update with changing knowledge. Traditionally, they used public health data, gathered from clinicians and government bodies, to track outbreaks of disease. "It's the gold standard," says Moyes, "People are carefully checking their data. But that system is very slow. So timeliness is one issue with the traditional data sources. And they rely on a country having public health infrastructure." So today they start with online news sources like Google News or its Chinese version Baidu. Reports about the target diseases are collected automatically from the internet and mapped onto a spatial grid that divides the world into five-kilometre squares. A quality check finds misleading data, such as a report tagged "Germany" because a German traveller caught leishmaniasis in Malaysia, or refines location data for greater precision, wherever possible. Validation of this data is carried out by machines that weigh it by how likely the system believes each point →

Photo: wavebreakmedia/shutterstock

to be valid. This process uses machine learning guided by human experts. It refers the most doubtful entries to humans for judgment and learns from the results.

The resulting spatial model is updated weekly, and the maps are released online. Not only the model, but all the data, including measures of uncertainty, are made available for anyone to use. In the short term, this detailed knowledge allows organizations to predict upsurges in, for example, dengue fever and target health interventions where they will be most useful. In the longer term, it's a warning system for changes in how diseases spread that might reflect a new type of insect vector or a change in local environmental conditions. This sharing of entire datasets, not just results, is one change Big Data brings to research.

Not everybody welcomes it. Scientists who have worked hard to gather data may guard it jealously, and commercial companies have a vested interest in keeping their results hidden from competitors.

Those working with individual patient records have issues of confidentiality, too. The capacity to link datasets, however anonymized, makes it possible to identify individuals from just a few pieces of information. A recent MIT study published in *Science* showed that by correlating anonymized credit card data with, for instance, time-stamped photos on Facebook, it took only four data points to identify 90% of the subjects. This brings a new obstacle to the research imperative to share data. Professor Paul Matthews talks about a fundamental dialectic from the individual's point of view, "the two interests, first in preserving your privacy, but secondly a sense of altruism, a sense of public interest: in general you would like to help further medical research." Legal systems around the world tackle this dilemma in different ways. In the USA, data tends to be regulated according to its intended use, with severe penalties for misleading practices or misuse. The EU is developing a new regulatory regime that is more uniform across countries and more powerfully enforced, but the European Parliament's call for more explicit consent at every stage, which could be enshrined in law by 2016, has researchers worried. "We need new ways of thinking about consent," says Professor Matthews. "We can't really give informed consent to these sorts of things, because the use of data will change over time." Matthews joined a working party of the UK's Nuffield Council on Bioethics, which produced a report in early 2015 entitled "The Collection, Linking and Use of Data in Biomedical Research and Health Care: Ethical Issues". Although the Nuffield report recommends changes to the law, it also calls for engagement with the people whose data is being used, at every stage from study design to changing use.

Early career researchers may have their own worries about Big Data, wondering how it will change the way research is conducted. What is the future for human researchers when computers can work so much faster? "For me, it started with simple statistics on ventricular size in schizophrenic patients," says Professor Van Horn. "That took me my whole PhD. Now that's how we spend our morning." Indeed, with machines learning to find novel associations, does the whole scientific process of designing experiments to test hypotheses become redundant?

Professor Matthews thinks not. For him the beauty of Big Data is that it allows both: starting with the hypothesis or with the data. "I might hypothesize that people on statins are less likely to show progression in multiple sclerosis (MS), which recent trials suggest. I can go into the dataset of people with MS and identify those who have used statins and those who have not, balance them for other factors, and ask the question, which population did better? That would be a hypothesis-led association study. The alternative is to use sophisticated data-mining approaches to allow clusters of features to be identified. It's always back and forth."

Professor Van Horn certainly thinks there is plenty of work for early-career researchers. "There's something for everybody in this field. Whether you're a clinician, a researcher, a neuroscientist, a computer scientist, an engineer, or a physicist, you can make a contribution. It's an exciting time to be thinking about Big Data, what it means in science, and how we can utilize it to make progress in understanding and curing diseases."

Please understand that in the interest of our fellows, we publish only results online, not descriptions of ongoing projects.

Therefore, this pdf continues with the section Results.

RESULTS The Boehringer Ingelheim Fonds funds excellent PhD students who are selected as much for their academic record as for their ambitious projects. Here they present a synopsis of their findings, which aim to push the boundaries of our knowledge of the fundamental phenomena of human life.

IN-VITRO RECONSTITUTION OF CONFINED MICROTUBULE CYTOSKELETON SELF-ORGANIZATION

cf. BIF FUTURA, VOL. 25 | 3.2010

HELLA BAUMANN
Discipline: Biochemist, Diploma
Institute: London Research Institute, Cancer Research UK, London, UK
Supervisor: Dr Thomas Surrey

The microtubule cytoskeleton determines the internal architecture of cells, which is crucial for proper cell function. In many vertebrate interphase cells, the microtubule cytoskeleton has an astral organization with stable microtubule ends focused near the cell centre and dynamic microtubule ends emanating out towards the plasma membrane. This arrangement is determined by a complex self-organization process involving the cell boundary and several proteins within and at the periphery of the cell. The aim of my PhD project was to elucidate the minimal set of protein activities required for the self-organization of microtubule arrays within a boundary. Using fluorescence microscopy, I systematically studied this process with self-organizing microtubule arrays reconstituted from purified components inside lipid monolayer-surrounded droplets in oil. I found that in the presence of a surrounding lipid monolayer, microtubule nucleation and polymerization can be achieved using a lipid composition close to that of a physiological plasma membrane and microtubule-nucleation enhancers, such as glycerol and paclitaxel, in the buffer. If no other proteins are incorporated into the droplets, the arrangement of microtubules depends on the droplet diameter and the efficiency of microtubule nucleation. Upon the addition of the minus-end directed microtubule-crosslinking motor kinesin 14, I detected single microtubule asters in larger droplets and bundles of microtubules in the smaller droplets. My results therefore indicate that the ratio of microtubule length to droplet diameter influences aster formation within a confined volume. In the future, these *in-vitro* reconstitution experiments could be extended by, for example, including regulators of microtubule dynamics and an additional actin cortex and by performing them in liposomes. This would provide further insights into the molecular mechanism of cytoskeleton self-organization inside a pliable boundary, which more closely reflects the situation *in vivo*.

PUBLICATIONS

Baumann H, Surrey T (2015) Self-organization of motors and microtubules in lipid-monolayered droplets. *Methods Cell Biol* **128:** (in press)

Baumann H, Surrey T (2014) Motor-mediated cortical versus astral microtubule organization in lipid-monolayered droplets. *J Biol Chem* **289:** 22524–22535

Gropeanu RA, Baumann H, Ritz S, Mailander V, Surrey T, Del Campo A (2012) Phototriggerable 2',7-caged Paclitaxel. *PLoS One* **7:** e43657

BST2 RESTRICTS HIV TRANSMISSION ACROSS MACROPHAGE–T CELL VIROLOGICAL SYNAPSES

cf. BIF FUTURA, VOL. 25 | 3.2010

SEBASTIAN GIESE
Discipline: Biologist, MSc
Institute: MRC Laboratory for Molecular Cell Biology, University College London, London, UK
Supervisor: Prof. Mark Marsh

Macrophages are key targets for human immunodeficiency virus (HIV) infection, but their role in pathogenesis is poorly understood. When HIV replicates in macrophages, the virus assembles in intracellular plasma membrane-connected compartments (IPMCs). IPMCs contain proteins that are also found at the cell surface, such as the focal adhesion protein CD18 and the restriction factor Bst2. CD18 is associated with coat structures on the membranes of IPMCs and may play a structural role in IPMC formation, whereas Bst2 inhibits HIV release from cells by physically tethering nascent virus particles at their assembly sites. The goal of my PhD was to investigate the function of CD18-containing coats in IPMC formation and the impact of IPMC-localized Bst2 on HIV transmission from macrophages. Using immunostaining and confocal microscopy, I found that IPMC formation coincides with increased cellular CD18 levels. When CD18 is depleted, the coat structures disappear, and the cells have fewer morphologically aberrant IPMCs. Using biochemical and imaging approaches, I showed that Bst2 retains HIV in IPMCs and reduces virus release and cell-free transmission, which relies on diffusion of virus to its target cell. HIV can also spread via direct cell–cell transmission across intercellular contacts termed virological synapses (VSs). I visualized VSs between macrophages and T cells by confocal microscopy, and used a co-culture assay to show that Bst2 also inhibits direct HIV transmission. When I used an HIV clone encoding the viral Vpu protein, the virus was efficiently released from macrophages and also spread via cell–cell transmission to T cells. Thus, Vpu efficiently overcomes Bst2-mediated inhibition of HIV propagation from macrophages. My results help to explain why the efficient viral counteraction of Bst2 was required for the global spread of HIV.

PUBLICATIONS

Giese S, Marsh M (2014) Tetherin can restrict cell-free and cell-cell transmission of HIV from primary macrophages to T cells. *PLoS Pathog* **10:** e1004189

Giese S, Marsh M (2013) Cellular trafficking mechanisms in the assembly and release of HIV. In Advances in HIV-1 Assembly and Release pp 23–53. Springer, New York, NY, USA:

Pelchen-Matthews A, Giese S, Mlcochova P, Turner J, Marsh M (2012) β2 integrin adhesion complexes maintain the integrity of HIV-1 assembly compartments in primary macrophages. *Traffic* **13:** 273–291

AFFINITY PROTEIN SWITCHES TO CONTROL PROTEIN FUNCTION IN LIVING CELLS

cf. BIF FUTURA, VOL. 26 | 3.2011

RUDOLF GRISS
Discipline: Chemical Biologist, MSc
Institute: École Polytechnique Fédérale de Lausanne (EPFL), Lausanne, Switzerland
Supervisor: Prof. Kai Johnsson

Research in cell biology relies on methods to modulate protein function in a specific and time-resolved manner. Many tools can be used to inactivate proteins, but they each have limitations. Gene knockouts and RNA interference, for example, suffer from limited temporal resolution. Although small molecule inhibitors excel in this regard, they can be difficult to generate with sufficient potency and specificity. This is particularly true when they are used to disrupt protein–protein interactions or inactivate members of highly conserved families of enzymes. Engineered antibody mimetics are protein-based inhibitors that provide the large interaction surface required for such targets; however, they are not cell permeable, which prevents them from being used as reversible inhibitors in time-resolved studies inside the cell. The goal of my PhD project was to develop a universal design for protein-based inhibitors that can be switched on and off using small molecules. This design would combine the high temporal resolution and reversibility of small molecule inhibitors with the high specificity and simple generation of protein-based inhibitors. Such affinity protein switches would thus serve as adaptors that transform unrelated small molecules into specific inhibitors of complex targets. Using protein engineering in combination with synthetic molecules, I constructed a protein that can exist in either of two distinct conformational states *in vitro* depending on the presence of a cell-permeable small molecule. By incorporating an affinity protein into the switchable system, I was able to translate the conformational change into a change in the potency of the protein-based inhibitor. I optimized the system further by altering the protein's geometry and modifying the synthetic molecule. I confirmed that the resulting switches could specifically inhibit a protease *in vitro* and could be controlled using a small molecule that is not related to the protease. Efforts to generalize the affinity protein switches for other targets and to study their behaviour in living cells are ongoing. If successful, they could become a powerful research tool for the elucidation of biological mechanisms.

PUBLICATIONS

The results of this project have not yet been published.

CONFORMATIONAL DYNAMICS OF LARGE PROTEINS AND PROTEIN COMPLEXES

cf. BIF FUTURA, VOL. 26 | 2.2011

DAVID HASELBACH
Discipline: Biophysicist, MSc
Institute: Max Planck Institute for Biophysical Chemistry, Göttingen, Germany
Supervisor: Prof. Holger Stark

Life is only possible through the enormous capabilities of proteins and their assemblies. A protein's function is encoded in its amino-acid sequence, which gives rise to a defined structure. This structure is not static; the thermal energy of the surrounding medium forces the molecule into different conformations. These movements often have an important role in protein function, so understanding function relies on high-resolution three-dimensional information about those movements. Single-particle electron cryo-microscopy (cryo-EM) is the preferred method for obtaining structural data for large proteins and protein complexes in different conformations. Since analysing the complete conformational landscape is not yet routine due to technical and computational limitations, the goal of my PhD was to develop the necessary methodology. The most important prerequisite for any structural analysis is an intact and homogeneous sample, so I developed a new method called ProteoPlex to systematically search for stabilizing conditions for a protein or complex of interest. ProteoPlex modifies an existing fluorescence-based stability screen called ThermoFluor to enable data obtained from large multidomain proteins and protein complexes to be analysed. I used ProteoPlex to find stabilizing conditions for more than 80 complexes from all branches of life. After implementing these conditions for a protein of interest, I could then perform conformational analyses. I focused on chromosome region maintenance 1 (CRM1), which is rather small (120 kDa) for cryo-EM analysis. I showed that the apo protein in the thermophilic eukaryote *Chaetomium thermophilum* cycles freely between an open superhelical and a closed ring-shaped conformation, and that only a short C-terminal helix is responsible for the enthalpic stabilization of the closed state. My work demonstrates that with a method such as ProteoPlex as a prerequisite, the routine analysis of conformational dynamics is possible even for challenging proteins.

PUBLICATIONS

Dölker N, Blanchet CE, Voß B, Haselbach D, Kappel C, Monecke T *et al* (2013) Structural determinants and mechanism of mammalian CRM1 allostery. *Structure* **21:** 1350–1360

Monecke T, Haselbach D, Voß B, Russek A, Neumann P, Thomson E *et al* (2013) Structural basis for cooperativity of CRM1 export complex formation. *Proc Natl Acad Sci USA* **110:** 960–965

INVESTIGATION OF POST-TRANSCRIPTIONAL GENE REGULATION IN T HELPER CELLS

cf. BIF FUTURA, VOL. 26 | 2.2011

GITTA HEINZ
Discipline: Molecular Medic, MSc
Institute: Helmholtz Zentrum München, Munich, Germany
Supervisor: Prof. Vigo Heissmeyer

Post-transcriptional gene regulation controls the translation efficiency and stability of mRNAs and has an important role in immune cells. The RNA-binding proteins Roquin-1 and Roquin-2 are required to prevent autoimmunity and excessive T cell activation in mice. One known target of Roquin-mediated post-transcriptional regulation in T cells is the inducible co-stimulator Icos. Roquin proteins bind to the *Icos* mRNA and thereby mediate mRNA degradation. However, the identity of other targeted mRNAs and the molecular requirements for Roquin/RNA interactions were unknown. In my PhD work, I deleted Roquin-encoding genes in primary murine T helper cells and compared those to wild-type controls in mRNA expression arrays. I identified a set of more than 50 potential Roquin target mRNAs. In cell-based functional experiments and *in-vitro* binding studies, I confirmed that the target candidate *Tnfrsf4,* another co-stimulatory receptor of T cells, was directly targeted by Roquin. Using crystal structures of the RNA-binding ROQ domain, I identified amino acids within the ROQ/RNA interface and tested their requirement for RNA binding. I also proved that reduced RNA-binding affinity resulted in impaired post-transcriptional repression of reporter genes upon overexpression of mutant Roquin-1 proteins in murine cells. My investigation of the RNA part showed that the ROQ domain binds with nanomolar affinity to RNA stem-loop structures, recognizing mainly their shape rather than a sequence motif. My findings increase the understanding of Roquin-mediated post-transcriptional gene regulation, which underlies the prevention of excessive inflammation and autoimmunity.

PUBLICATIONS

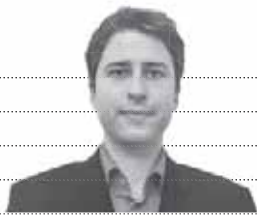
Jeltsch KM, Hu D, Brenner S, Zöller J, Heinz GA, Nagel D *et al* (2014) Cleavage of roquin and regnase-1 by the paracaspase MALT1 releases their cooperatively repressed targets to promote TH17 differentiation. *Nat Immunol* **15:** 1079–1089

Schlundt A, Heinz GA, Janowski R, Geerlof A, Stehle R, Heissmeyer V *et al* (2014) Structural basis for RNA recognition in roquin-mediated post-transcriptional gene regulation. *Nat Struct Mol Biol* **21:** 671–678

Vogel KU, Edelmann SL, Jeltsch KM, Bertossi A, Heger K, Heinz GA *et al* (2013) Roquin paralogs 1 and 2 redundantly repress the Icos and Ox40 costimulator mRNAs and control follicular helper T cell differentiation. *Immunity* **38:** 655–668

pVHL-DEPENDENT microRNA SIGNALLING IN RENAL CELL CARCINOMA

cf. BIF FUTURA, VOL. 25 | 3.2010



MICHAEL HELL
Discipline: Molecular Biologist, Diploma
Institute: Institute of Molecular Health Sciences, ETH Zurich, Zurich, Switzerland
Supervisor: Prof. Wilhelm Krek

Clear cell renal cell carcinoma (ccRCC) is the main form of kidney cancer in humans and is genetically tightly linked to mutations in the von Hippel-Lindau (*VHL*) tumour suppressor gene. The Krek group has previously shown that the loss of the VHL protein (pVHL) in various cell models leads to aneuploidy, which is a deviation in the normal chromosome number as a result of chromosome missegregation. The aim of my PhD was to study how pVHL loss results in aneuploidy and to determine the significance of this pathway in the development of ccRCC. I showed via quantitative microRNA detection and microarray technology that the absence of pVHL induces the microRNA miR-28-5p in cultured cells, kidney-specific *Vhl*-knockout mice, as well as in ccRCC patients. Using polysome and luciferase assays, I found that miR-28-5p in turn downregulates the spindle checkpoint protein Mad2 (Mad2L1) through interaction with the 3' untranslated region (3'UTR) of Mad2 mRNA. Importantly, the addition of oligonucleotides known as antagomirs to silence microRNA restored the checkpoint *in vitro* and *in vivo,* indicating that miR-28-5p inhibition could constitute a therapeutic approach in ccRCC. To investigate the role of pVHL *in vivo,* we established a surgically induced mouse model in which the study of chromosome segregation in otherwise senescent kidney cells is possible. In this model, we could show that kidney-specific *Vhl*-knockout mice – unlike control littermates – had aberrant aneuploidy levels and cystic and dysplastic pre-cancer lesions reminiscent of human ccRCC histopathology. My results therefore suggest a novel mechanism of aneuploidy suppression via a pVHL-regulated microRNA pathway. Taken together, these data will contribute to our understanding of kidney cancer progression and to the development of future therapeutic options.

PUBLICATIONS

Hell MP, Duda M, Weber TC, Moch H, Krek W (2014) Tumor suppressor VHL functions in the control of mitotic fidelity. *Cancer Res* **74:** 2422–2441

Hell MP, Thoma CR, Fankhauser N, Christinat Y, Weber TC, Krek W (2014) miR-28-5p promotes chromosomal instability in VHL-associated cancers by inhibiting Mad2 translation. *Cancer Res* **74:** 2432–2443

DLG1 IS REQUIRED FOR MYOFIBRILLAR ARRANGEMENT IN THE *DROSOPHILA* HEART

cf. BIF FUTURA, VOL. 26 | 1.2011

ANNETTE HELLBACH
Discipline: Molecular Medic, Diploma
Institute: Max Planck Institute of Biochemistry, Martinsried, Germany
Supervisor: Dr Frank Schnorrer

The heart is the central organ of the circulatory system. Cardiac muscle cells, known as cardiomyocytes, comprise myofibrils, which mediate regular heartbeat – without which no higher life would be possible. The tubular heart of *Drosophila* is built by two rows of cardiomyocytes, which house regularly arranged myofibrils that originate at the cell–cell junction between the two rows (called the longitudinal junction) and run in parallel circularly around the heart tube. In the mammalian heart, myofibrils are anchored at the intercalated disc, which connects cardiomyocytes. In both *Drosophila* and mammalian hearts, it is not known how this anchoring site for myofibrils is established and how it distinguishes itself from other spaces within the cell. Understanding these processes could help to address hereditary and sporadic human cardiomyopathies, which represent an important public health problem. In my PhD studies, I hypothesized that the location of the longitudinal junction within the cardiomyocyte might be further characterized by discovering proteins with essential functions in epithelial polarity. Therefore, I used RNA interference (RNAi) to knock down all epithelial polarity proteins for which either antibodies or RNAi lines were available in the *Drosophila* heart. Then, by imaging the hearts using a fluorescence microscope, I tested how myofibrillar arrangement changed as a result. Of those proteins, Discs large 1 (Dlg1) not only localized around the longitudinal junction, but also caused a severe misarrangement of myofibrils: myofibrils either showed a chaotic arrangement or, in the most severe cases, changed their running direction by 90° such that they ran longitudinally along the heart tube instead of around it. In addition, I was able to identify expression of the beta PS integrin as a marker for the presence and location of the longitudinal junction. My results provide the first insight into the mechanisms governing myofibrillar arrangement in the *Drosophila* heart. Because the longitudinal junction in *Drosophila* heart is similar in morphology and function to the intercalated disc in vertebrates, my findings are important to the study of human cardiac pathologies.

PUBLICATIONS

The results of this PhD project have not yet been published.

INTEGRIN SUBTYPE-SPECIFIC EFFECT ON CELL CONTRACTILITY AND ACTIN-BASED SIGNALLING

cf. BIF FUTURA, VOL. 26 | 2.2011

MICHAELA-ROSEMARIE HERMANN
Discipline: Molecular Biomedic, Diploma
Institute: Max Planck Institute of Biochemistry, Martinsried, Germany
Supervisor: Prof. Reinhard Fässler

Integrins are α/β heterodimeric cell-adhesion molecules that connect the extracellular matrix with the actin cytoskeleton. By assembling into focal adhesions (FAs), integrins establish a molecular platform where mechanical forces, cytoskeletal organization and biochemical signals intersect and modulate cellular functions such as cell migration, proliferation, differentiation, and survival. Mammalian cells usually co-express several integrins that recognize different extracellular matrix proteins, become activated and trigger their own intracellular signalling events, but also cross-talk with other signalling cascades. *In-vivo* and *in-vitro* studies have shown that fibronectin (FN)-binding integrins (e.g. β1- and all αV-class) have both specific and redundant roles, but how these distinct classes accomplish their individual functions and whether they co-operate was unclear. The goal of my PhD studies was to dissect FN-binding integrin-specific signalling and to investigate their individual and co-operative action. Because the major FN-binding integrins, α5β1 and αV-class, are co-expressed on most cells, I used pan-integrin-null kidney fibroblasts in which αV- or β1-class heterodimer subsets were re-expressed separately or together. By combining biochemical isolation protocols with quantitative mass spectrometry and standard biochemical assays, I analysed the molecular composition of FAs assembled by a single integrin class. I found that α5β1 integrins are responsible for force generation, whereas αV-class integrins induce the generation of actin filaments. I also investigated whether and how αV- and/or β1-class integrins mediate changes in gene expression. I showed that these integrins regulate cytoskeletal dynamics and break the interaction between actin and the transcriptional co-activator megakaryocyte acute leukemia (MAL) to drive MAL/serum-response-factor (SRF)-mediated gene expression. My results show that the signalling cascade comprising αV-/β1-class integrins and MAL/SRF precisely adjusts the adhesion and actin remodelling required for cell spreading, migration, and invasion.

PUBLICATIONS

Schiller HB, Hermann MR, Polleux J, Vignaud T, Zanivan S, Friedel CC *et al* (2013) β1- and αv-class integrins cooperate to regulate myosin II during rigidity sensing of fibronectin-based microenvironments. *Nat Cell Biol* **15:** 625–636

NOVEL STRATEGIES TO ASSESS RECOGNITION OF TUMOUR CELLS BY T CELLS

cf. BIF FUTURA, VOL. 24 | 2.2009

CARSTEN LINNEMANN
Discipline: Immunologist, Diploma
Institute: Netherlands Cancer Institute, Amsterdam, Netherlands
Supervisor: Prof. Ton N. M. Schumacher

T cells protect the human body against pathogens. T cell receptors (TCRs) identify infected cells by mediating the recognition of pathogen-derived peptides, which are presented at the cell surface as peptide–MHC (pMHC) complexes. T cells use the same MHC-mediated recognition mechanisms to eradicate cancer cells, but the identities of the pMHC complexes involved in tumour eradication are not known. Tumours carry mutations, but whether these mutations make tumours visible to the immune system has been difficult to determine due to their high frequency and diversity. In my PhD, I studied the mechanisms of anti-tumour T cell immunity. As part of this process, I developed two novel tools to study the interaction between the tumour and T cells by deciphering its core components: tumour-associated pMHC complexes and tumour-specific TCRs. First, I created a platform based on immortalized, autologous B cells to load these antigen-presenting cells with candidate antigens. Using my technique, I demonstrated that 80% of pMHC complexes with mutated peptides were recognized by intratumoural $CD4^+$ T cells in melanoma. Second, I developed a TCR gene capture approach in human cells to rapidly identify TCR genes by targeted sequencing of their genomic loci. This method facilitates the rapid assembly of libraries of TCR genes, e.g. from tumour antigen-specific T cells. Such TCR libraries are valuable for studying the frequency with which specific pMHC complexes on tumour cells are recognized by T cells. They can also be used to find the genes of TCRs known to recognize a pMHC complex on a tumour cell, which could be exploited for therapeutic application. The tools developed during my PhD enable the systematic analysis of the relevance of different tumour antigens to the treatment of human cancer.

PUBLICATIONS

Linnemann C, van Buuren M, Bies L, Verdegaal EM, Schotte R, Calis JJA *et al* (2015) High throughput epitope discovery reveals frequent recognition of neo-antigens by CD4+ T-cells in human melanoma. *Nat Med* **21:** 81–85

Linnemann C, Heemskerk B, Kvistborg P, Kluin RJC, Bolotin DA, Chen X *et al* (2013) High-throughput identification of antigen-specific TCRs by TCR gene capture. *Nat Med* **11:** 1534–1541

Bendle GM, Linnemann C, Hooijkaas AI, Bies L, de Witte MA, Jorritsma A *et al* (2010) Lethal graft-versus-host disease in mouse models of T cell receptor gene therapy. *Nat Med* **5:** 565–570

REGULATED DISULPHIDE BOND FORMATION IN THE ER PREVENTS OXIDATIVE DAMAGE OF THE CYTOSOL

cf. BIF FUTURA, VOL. 26 | 3.2011

THOMAS RAMMING
Discipline: Molecular Medic, Diploma
Institute: Department of Pharmaceutical Sciences, University of Basel, Basel, Switzerland
Supervisor: Dr Christian Appenzeller-Herzog

Many secretory or membrane proteins must form disulphide bonds to maintain their structure or function. These covalent linkages between two cysteine side chains often play a key role in folding native polypeptide chains in the endoplasmic reticulum (ER). The bonds are introduced by a disulphide relay system comprising the flavoenzyme endoplasmic oxidoreductin 1 (Ero1) and protein disulphide isomerase (PDI). Ero1 generates disulphides *de novo* via its bound flavin adenine dinucleotide (FAD) co-factor. Since FAD uses oxygen (O_2) as an electron acceptor, the reactive oxygen species (ROS) hydrogen peroxide (H_2O_2) is formed as a byproduct. Newly generated disulphides are transferred to PDI, which engages with various substrates to facilitate protein maturation. Although Ero1 consumes O_2 and releases H_2O_2, none of the Ero1 crystal structures reveal a potential path for the entry or exit of these molecules. Before my PhD work, regulation of Ero1 activity by PDI was not fully understood and the fate of the reactive byproduct H_2O_2 was completely obscure. By genetically encoding two sensors specific to H_2O_2 and glutathione and depleting glutathione peroxidase 8 (GPx8) in human cells using small interfering RNA, I examined phenotypic changes using fluorescence assays, quantitative polymerase chain reaction, and cell viability readouts. I found that GPx8 detoxifies Ero1-derived H_2O_2 before it can diffuse from the ER lumen, thereby protecting the cytosol from oxidative damage. Furthermore, I mapped the GPx8-binding site of Ero1 to a protein loop between two Cys residues that were formerly thought to form a structural disulphide. I showed that this loop serves instead as a PDI-regulated switch, allowing the O_2 molecule to access FAD. My results describe the first disulphide-regulated O_2 access into a flavoprotein and unravel the degradation pathway for Ero1-derived H_2O_2 in mammalian cells.

PUBLICATIONS

Ramming T, Hansen HG, Nagata K, Ellgaard L, Appenzeller-Herzog C (2014) GPx8 peroxidase prevents leakage of H2O2 from the endoplasmic reticulum. *Free Radic Biol Med* **70:** 106–116

Ramming T, Appenzeller-Herzog C (2013) Destroy and exploit: Catalyzed removal of hydroperoxides from the endoplasmic reticulum. *Int J Cell Biol* **2013:** 180906

Ramming T, Appenzeller-Herzog C (2012) The physiological functions of mammalian endoplasmic oxidoreductin 1: On disulfides and more. *Antioxid Redox Signal* **16:** 1109–1118

SILENCING GENOMIC INTRUDERS BY SMALL RNAs

cf. BIF FUTURA, VOL. 26 | 3.2011

GRZEGORZ SIENSKI
Discipline: Molecular Biologist, MSc
Institute: Institute of Molecular Biotechnology of the Austrian Academy of Sciences (IMBA), Vienna, Austria
Supervisor: Dr Julius Brennecke

Argonaute proteins are essential components of the RNA-silencing pathways that control gene expression in eukaryotes. A subset of these proteins belonging to the germline-restricted PIWI clade acts predominantly in the nucleus. Piwi and its bound Piwi-interacting RNAs (piRNAs) are central players in a gonad-specific genome surveillance system that keeps transposable elements under control. In the absence of the piRNA pathway, RNA levels of several transposons increase several hundred fold, ultimately leading to widespread transposition events, genomic damage, and sterility. The major aim of my PhD was to systematically dissect the mechanism of Piwi silencing in *Drosophila melanogaster.* I used genome-wide sequencing approaches to measure steady-state and nascent RNAs and to analyse distinct chromatin modifications. I found that Piwi silences its targets primarily at the transcriptional level and that this is accompanied by extensive methylation of histone H3 lysine 9, a hallmark of transcriptionally repressed chromatin. Piwi-mediated transposon silencing requires its binding partner DmGtsf1 and the Maelstrom protein and severely impacts on the expression of genes in the vicinity of transposon insertions. In addition to its role in silencing, the Piwi–piRNA complex instructs several distinct loci to produce piRNAs. These findings provide the first genome-wide demonstration of a transcriptional silencing process directed by small RNAs in *Drosophila*. Given the large similarities between fly and mammalian piRNA systems, my studies have direct implications for small RNA-mediated genome defence in animals. They also provide insights into how transposons and the host pathways devoted to their silencing influence gene expression, and thus support a central role for transposons in genome evolution.

PUBLICATIONS

Mohn F, Sienski G, Handler D, Brennecke J (2014) The rhino-deadlock-cutoff complex licenses noncanonical transcription of dual-strand piRNA clusters in *Drosophila*. *Cell* **157:** 1364–1379

Donertas D, Sienski G, Brennecke J (2013) *Drosophila* Gtsf1 is an essential component of the Piwi-mediated transcriptional silencing complex. *Genes Dev* **27:** 1693–1705

Sienski G, Dönertas D, Brennecke J (2012) Transcriptional silencing of transposons by Piwi and maelstrom and its impact on chromatin state and gene expression. *Cell* **151:** 964–980

FLIPPING THE REDOX SWITCH: SPECIFIC AND EFFICIENT THIOL OXIDATION IN MAMMALS

cf. BIF FUTURA, VOL. 25 | 3.2010

MIRKO C. SOBOTTA
Discipline: Biochemist, Diploma
Institute: German Cancer Research Center (DKFZ), Heidelberg, Germany
Supervisor: Dr Tobias P. Dick

Hydrogen peroxide (H_2O_2) belongs to the group of reactive oxygen species (ROS) and is a strong oxidant that is commonly used as a disinfectant and bleaching agent. For a long time, H_2O_2 was thought to be an unwanted by-product of cellular respiration that needed to be removed by scavenging enzymes such as the abundant peroxiredoxins (Prx). However, during the past 10 years it has become clear that H_2O_2 is actively produced by the cell and is involved in signal transduction pathways such as growth factor signalling or the response to antigens. It regulates the function of redox-regulated proteins by oxidizing distinct cysteinyl thiols to disulphide bonds in a reversible, non-toxic process. One of the unsolved questions in redox biology is the specificity of H_2O_2 oxidation: how does the small H_2O_2 molecule act on certain proteins and their thiols but leaves other proteins untouched? In addition, it is not clear why the small amounts of H_2O_2 that are produced during signalling are not immediately scavenged by Prx enzymes before they can react with their target proteins. In my PhD project, I showed that in mammalian cells, proteins are oxidized by H_2O_2 through a redox relay involving peroxiredoxins. These enzymes use oxidative equivalents from H_2O_2 to specifically oxidize other proteins. Using redox proteomics, I found that Prx2 specifically and efficiently oxidizes the transcription factor STAT3 under pro-oxidative conditions as well as during cytokine signalling. Specific thiols of STAT3 are targeted by Prx2, resulting in disulphide-linked STAT3 oligomers with attenuated transcriptional activity. Thus, the activity of STAT3 is modulated by Prx2 expression levels. My study suggests that peroxidase-based redox relays are common in mammals and may represent a general principle explaining the specificity and efficiency of intracellular peroxide signals.

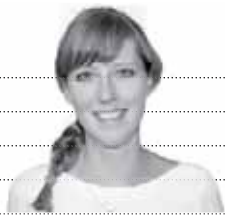
PUBLICATIONS

Sobotta MC, Liou W, Stöcker S, Talwar D, Oehler M, Ruppert T *et al* (2015) Peroxiredoxin-2 and STAT3 form a redox relay for H_2O_2 signaling. *Nat Chem Biol* **11:** 64–70

Sobotta MC, Barata AG, Schmidt U, Mueller S, Millonig G, Dick TP (2013) Exposing cells to H_2O_2: A quantitative comparison between continuous low-dose and one-time high-dose treatments. *Free Radic Biol Med* **60:** 325–335

INSIGHTS INTO NOVEL CAVEOLAR PROTEINS: EHD2 CONFINES CAVEOLAE TO THE PLASMA MEMBRANE

cf. BIF FUTURA, VOL. 25 | 3.2010



MIRIAM STÖBER
Discipline: Biochemist, MSc
Institute: Institute of Biochemistry, ETH Zurich, Zurich, Switzerland
Supervisor: Prof. Ari Helenius

Caveolae are 50–80 nm flask-shaped plasma membrane domains that cover up to one-third of the cell surface in endothelial and smooth muscle cells, fibroblasts, and adipocytes. They function in mechanosensing, membrane organization, signalling, and endocytosis. Most caveolae are immobile but they can be activated to undergo internalization; however, the cellular factors that regulate the balance between static caveolae and motile caveolar carriers are not known. The Eps-15 homology domain-containing protein 2 (EHD2) was identified as one of several proteins at caveolae isolated from human adipocytes. In my PhD project, I analysed the specific subcellular localization and the cellular function of EHD2. I performed a multidisciplinary study combining biochemistry with various fluorescence-based live cell microscopy assays to show that EHD2 constitutes a major, novel component of invaginated caveolae. By creating several EHD2 mutants, I uncovered that EHD2 is recruited from the cytosol to the plasma membrane, where it is present as large 60–75S homo-oligomers. The formation of the EHD2 complexes requires ATP binding and membrane association. Both the depletion of EHD2 by RNAi and the expression of a dominant-negative EHD2 mutant significantly elevate the fraction of mobile caveolae, indicating that the function of EHD2 is to confine caveolae to the plasma membrane. Consistent with a constraining role, I found that EHD2 negatively regulates the endocytosis of two caveolar cargos, cholera toxin B and simian virus 40. Further experiments showed that EHD2 associates with actin filaments and thereby mediates an intimate connection between stationary caveolae and the actin cytoskeleton. Thus, my results have revealed that EHD2 is a crucial regulator of the dynamic behaviour of caveolae and, consequently, their diverse roles in physiological processes.

PUBLICATIONS

Stoeber M, Stoeck IK, Hänni C, Bleck CKE, Balistreri G, Helenius A (2012) Oligomers of the ATPase EHD2 confine caveolae to the plasma membrane through association with actin. *EMBO J* **31:** 2350–2364

Hayer A, Stoeber M, Bissig C, Helenius A (2010) Biogenesis of caveolae: Stepwise assembly of large caveolin and cavin complexes. *Traffic* **11:** 361–382

Hayer A, Stoeber M, Ritz D, Engel S, Meyer HH, Helenius A (2010) Caveolin-1 is ubiquitinated and targeted to intralumenal vesicles in endolysosomes for degradation. *J Cell Biol* **191:** 615–629

MOLECULAR MECHANISM OF LIGHT ACTIVATION IN THE BLUE LIGHT PHOTORECEPTOR BLUF

cf. BIF FUTURA, VOL. 25 | 3.2010

ANIKÓ UDVARHELYI
Discipline: Physicist, Diploma
Institute: Max Planck Institute for Medical Research, Heidelberg, Germany
Supervisor: Dr Tatiana Domratcheva

Photoreceptor proteins translate photon energy into biological information. Light absorption induces a photochemical reaction in the photoreceptor's chromophore that switches the protein conformation from the dark state to the light state. The BLUF (blue light using flavin adenine dinucleotide) photoreceptor exploits photo-induced proton-coupled electron transfer (PCET) for this reaction. BLUF dark and light states bind the oxidized flavin chromophore in hydrogen-bonding networks that differ in the conformation of a conserved glutamine residue. To resolve the considerable controversy over how Gln takes part in these networks, computational studies are indispensable for adequately interpreting the spectroscopy data and fully understanding the reaction mechanism. In my PhD studies, I presented the first systematic computational study of photo-induced PCET in BLUF. I created BLUF models containing different Gln rotamers and tautomers and compared their energies to determine the lowest and most likely structures of the BLUF dark and light states. To characterize the BLUF photoreaction, I mapped the pathway connecting the dark and light states on the excited-state potential-energy surface. As a prerequisite, I established a computational procedure using state-of-the-art electronic-structure methods, and identified quantum-mechanical cluster and hybrid quantum-mechanical/molecular-mechanical models that satisfied my requirements. After benchmarking the protocol, I computed the most complete BLUF photoreaction pathways to date. My results explain for the first time how PCET activates the dark state but at the same time conveys photostability to the light state in BLUF. My calculations also provide the framework for understanding available experimental data on redox-tuning effects in BLUF proteins.

PUBLICATIONS

Domratcheva T, Udvarhelyi A, Shahi ARM (2014) Computational spectroscopy, dynamics, and photochemistry of photosensory flavoproteins. In Flavins and Flavoproteins: Methods and Protocols, Weber S, Schleicher E (eds) pp 191–228. Springer, New York, NY, USA

Udvarhelyi A, Domratcheva T (2013) Glutamine rotamers in BLUF photoreceptors: A mechanistic reappraisal. *J Phys Chem B* **117:** 2888–2897

Udvarhelyi A, Domratcheva T (2011) Photoreaction in BLUF receptors: Proton-coupled electron transfer in the flavin-Gln-Tyr system. *Photochem Photobiol* **87:** 554–563

MECHANISMS OF DIFFERENTIAL CENTRIOLE INHERITANCE IN *CAENORHABDITIS ELEGANS*

cf. BIF FUTURA, VOL. 26 | 1.2011

LUKAS VON TOBEL
Discipline: Biochemist, MSc
Institute: Swiss Federal Institute of Technology (EPFL), Lausanne, Switzerland
Supervisor: Prof. Pierre Gönczy

Centrosomes are the main microtubule-organizing centres in most animal cells and they consist of centrioles and the surrounding pericentriolar material. In many metazoan species, centrioles need to be eliminated during oogenesis and maintained during spermatogenesis so that the zygote has the correct number of centrioles after fertilization. Centrioles are exceptionally stable structures but very little is known about how their stability is guaranteed. The first aim of my PhD thesis was therefore to analyse how and when centrioles are eliminated during oogenesis. Using immunofluorescence and electron microscopy, we showed that centriole elimination during *Caenorhabditis elegans* oogenesis occurs within approximately 30 minutes during the diplotene stage of meiotic prophase I. Furthermore, through the analysis of various deletion mutants as well as animals depleted in specific genes by RNA interference, we uncovered that the RNA helicase CGH-1 is important for this process as is the germ-cell karyotype, but that the surrounding somatic tissue does not contribute significantly to elimination. In the second part of my project, I characterized the role of a gene called *sas*-1 in centriole integrity. Wild-type oocytes fertilized by *sas*-1 mutant sperm assemble a monopolar spindle in the first cell cycle. We were able to show that this defect arises because *sas*-1 mutant centrioles lose their stability shortly after fertilization, resulting in the absence of normal centrioles at the first mitosis. SAS-1 is a centriolar C2 domain-containing protein and we found that it is related to human C2CD3, which is also required for centriole formation in human cells. Intriguingly, we uncovered that SAS-1 can bind and stabilize microtubules in human cells. Thus, we propose that SAS-1 maintains *C. elegans* centrioles by binding and stabilizing centriolar microtubules. Taken together, my results open the door for a mechanistic dissection of centriole elimination and identify an initial player required for the maintenance of centriole integrity in *C. elegans*.

PUBLICATIONS

von Tobel L, Mikeladze-Dvali T, Delattre M, Balestra FR, Blanchoud S, Finger S *et al* (2014) SAS-1 is a C2 domain protein critical for centriole integrity in *C. elegans*. *PLoS Genet* **10:** e1004777

Mikeladze-Dvali T, von Tobel L, Strnad P, Knott G, Leonhardt H, Schermelleh L *et al* (2012) Analysis of centriole elimination during *C. elegans* oogenesis. *Development* **139:** 1670–1679

A MOLECULAR CHARACTERIZATION OF BREAST CANCER PROGRESSION AND METASTASIS

cf. BIF FUTURA, VOL. 25 | 3.2010

ELVIN WAGENBLAST
Discipline: Molecular Biotechnologist, BSc
Institute: Cold Spring Harbor Laboratory, Cold Spring Harbor, NY, USA
Supervisor: Prof. Gregory Hannon

Despite recent advances in the treatment of advanced breast cancer, most patients with metastatic disease succumb to their disease. During metastasis, primary tumour cells evolve the capacity to intravasate into the lymphatic or vasculature systems, extravasate into a target organ such as the lung, and eventually colonize secondary sites. Previous studies have demonstrated that individual cells within complex tumourigenic populations show heterogeneity in their capacity to form metastatic lesions. However, no model system has yet emerged that allows genetically modified cells to be followed in a polyclonal context throughout each stage of metastatic disease progression. During my PhD studies, I developed a mouse model to probe the role of breast tumour heterogeneity in multiple stages of disease using molecular barcodes and next-generation sequencing technologies. I found that distinct clones within a mixed population display specialization, for example, dominating the primary tumour, contributing to metastatic populations, or showing tropism for entering the lymphatic or vasculature systems. I correlated these stable properties with distinct gene-expression profiles. Those tumourigenic clones that efficiently entered the vasculature expressed two secreted proteins, Serpine2 and Slpi, which are necessary and sufficient to programme these cells for vascular mimicry. Through this process, highly aggressive tumour cells form channels to distribute blood to hypoxic regions (that is, lacking oxygen) of the tumour. My data indicate that Serpine2 and Slpi drive the formation of these extravascular networks, ultimately leading to the formation of distant metastases. Thus, these two proteins – and the phenotype they promote – may be broadly relevant as drivers of metastatic progression in human cancer.

PUBLICATIONS

Wagenblast E, Soto M, Gutiérrez-Ángel S, Hartl AH, Gable AL, Maceli AR *et al* (2015) A model of breast cancer heterogeneity reveals vascular mimicry as a driver of metastasis. *Nature* **520:** 358–362

Weissmueller S, Manchado E, Saborowski M, Morris JP IV, Wagenblast E, Davis CA *et al* (2014) Mutant p53 drives pancreatic cancer metastasis through cell-autonomous PDGF receptor β signaling. *Cell* **157:** 382–394

Knott SRV, Maceli AR, Erard N, Chang K, Marran K, Zhou X, Gordon A, Demerdash El O, Wagenblast E *et al* (2014) A computational algorithm to predict shRNA potency. *Mol Cell* **56:** 796–807

MD FELLOWS 2014

With its MD fellowships, the Boehringer Ingelheim Fonds helps outstanding medical students to pursue an ambitious experimental project in basic biomedical research. Candidates study in Germany and change their workplace (institution and city) for at least ten months to join an internationally renowned laboratory. Here we present the nine fellows who were granted an MD fellowship.

NINA AUERBACH

The physical role of the adhesion G protein-coupled receptor GPR133 in zebrafish

MORINA BRINGEZU

Perceptual learning of temporal sequences: Probing the emergence of novel sensory memories for audition and vision

TERESA GERHARDT

Peptide-MHC-II tetramers as biomarkers for successful vaccination strategies in atherosclerosis

KONRAD HOEFT

Effect of hepcidin in mouse models of acute lung injury

LUKAS JOHN

The role of oxalate-mediated inflammation on the progression of chronic kidney disease

RAJIV KHAJURIA

Deciphering the molecular basis of the hypoplastic anaemia in cartilage-hair hypoplasia

LEONARD KIRN

Tumor–stroma crosstalk in colorectal cancer: The role of paracrine hedgehog signalling

JONA KROHN

The role of VCAM-1 and related factors on endothelial-to-mesenchymal transdifferentiation in the tethered mitral valve

STEFAN KUHLMANN

Defining the molecular cause of altered response to antiarrhythmic drugs in Pitx2-dependent atrial fibrillation

THE PHYSICAL ROLE OF THE ADHESION G PROTEIN-COUPLED RECEPTOR GPR133 IN ZEBRAFISH

NINA AUERBACH

Project at: Department of Developmental Biology, Washington University School of Medicine, St. Louis, MO, USA

Supervisor: Prof. Kelly Monk, PhD

Home university: University of Leipzig

PERCEPTUAL LEARNING OF TEMPORAL SEQUENCES: PROBING THE EMERGENCE OF NOVEL SENSORY MEMORIES FOR AUDITION AND VISION

MORINA BRINGEZU

Project at: Département d'Études Cognitives, École Normale Supérieure, Paris, France

Supervisor: Dr Daniel Pressnitzer

Home university: University of Heidelberg

PEPTIDE-MHC-II TETRAMERS AS BIOMARKERS FOR SUCCESSFUL VACCINATION STRATEGIES IN ATHEROSCLEROSIS

TERESA GERHARDT

Project at: Division of Inflammation Biology, La Jolla Institute for Allergy and Immunology, La Jolla, CA, USA

Supervisor: Prof. Klaus Ley, MD

Home university: Medical Center – University of Freiburg

EFFECT OF HEPCIDIN IN MOUSE MODELS OF ACUTE LUNG INJURY

KONRAD HOEFT

Project at: Department of Anesthesia & Critical Care, Massachusetts General Hospital, Boston, MA, USA

Supervisor: Warren M. Zapol, MD

Home university: RWTH Aachen University Clinic

THE ROLE OF OXALATE-MEDIATED INFLAMMATION ON THE PROGRESSION OF CHRONIC KIDNEY DISEASE

LUKAS JOHN

Project at: The Anlyan Center, Yale University, New Haven, CT, USA

Supervisor: Prof. Peter S. Aronson, MD

Home university: University Erlangen-Nuremberg

DECIPHERING THE MOLECULAR BASIS OF THE HYPOPLASTIC ANAEMIA IN CARTILAGE-HAIR HYPOPLASIA

RAJIV KHAJURIA

Project at: Boston Children's Hospital, Harvard Medical School, Boston, MA, USA

Supervisor: Dr Vijay Sankaran, MD

Home university: Charité-Universitätsmedizin Berlin

TUMOR–STROMA CROSSTALK IN COLORECTAL CANCER: THE ROLE OF PARACRINE HEDGEHOG SIGNALLING

LEONARD KIRN

Project at: Department of Bioscience and Nutrition, Karolinska Institutet, Huddinge, Sweden

Supervisor: Prof. Rune Toftgard

Home university: Charité-Universitätsmedizin Berlin

THE ROLE OF VCAM-1 AND RELATED FACTORS ON ENDOTHELIAL-TO-MESENCHYMAL TRANSDIFFERENTIATION IN THE TETHERED MITRAL VALVE

JONA KROHN

Project at: Brigham and Women's Hospital, Harvard Medical School, Boston, MA, USA

Supervisor: Prof. Elena Aikawa, MD

Home university: Heidelberg University Hospital

DEFINING THE MOLECULAR CAUSE OF ALTERED RESPONSE TO ANTI-ARRHYTHMIC DRUGS IN PITX2-DEPENDENT ATRIAL FIBRILLATION

STEFAN KUHLMANN

Project at: Center for Cardiovascular Sciences, University of Birmingham, Birmingham, UK

Supervisor: Prof. Paulus Kirchhof

Home university: Max Delbrück Center for Molecular Medicine

THE FOUNDATION The Boehringer Ingelheim Fonds (BIF) is a public foundation – an independent, non-profit organization for the exclusive and direct promotion of basic research in biomedicine. The foundation pays particular attention to fostering junior scientists. From the start it has provided its fellowship holders with more than just monthly bank transfers: seminars, events, and personal support have nurtured the development of a worldwide network of current and former fellows.

PERSPECTIVES

HOW TO POSITION YOURSELF

In this section, we introduce BIF alumni from various scientific backgrounds and professional contexts. They describe their career paths and highlight important steps and decisions that helped them to reach their current position.

INTERVIEW WITH DR SYLVIA ERHARDT, CENTER FOR MOLECULAR BIOLOGY IN HEIDELBERG, GERMANY

Photo: Sylvia Erhardt: Universität Heidelberg

Sylvia Erhardt was born in Karlsruhe, Germany, in 1971 and studied biology in nearby Heidelberg. Her diploma was in the then young field of epigenetics at the Center for Molecular Biology (ZMBH). In 1999, her PhD in the field of epigenetic regulation of primordial germ cells took her to Cambridge, UK, for four years. For her postdoc at the University of Berkeley, USA, she switched to studying epigenetic regulation of centromeres in fruit flies. In 2007, Sylvia's career came full circle when she returned to the ZMBH as an independent group leader of the CellNetworks Excellence Cluster. She turned down a grant from the NIH to do so. Her husband, now a chemist at the German Cancer Research Center (DKFZ), and her three-year-old daughter accompanied her on this move. Her work at the ZMBH focuses on how chromosomes segregate during cell division in normal and tumour cells, with a special emphasis on epigenetic changes in and around the centromere. She now has two daughters aged ten and five.

Between your PhD and your postdoc you repositioned yourself. Why did you do that and would you do it again?

During my PhD, I worked on epigenetic mechanisms in early mouse development and the embryonic germ line – a topic that still fascinates me – and my current research may in fact lead back to this field. However, during my PhD I became intrigued by chromosome segregation. As epigenetically controlled centromeres are at the base of this process, the next career step was quickly decided. I personally think that changing fields from PhD to postdoc can be very beneficial. You can build up a unique combination of ideas and know-how. If, at some point, you are able to combine all your experience, you may have found a niche for your independent research.

How did third-party funding influence your career choices?

I made my decisions where to go for my PhD and postdoc before applying for independent funding. I realized early on that getting your own research money is an important lesson to learn and makes you more independent. It also always gave me a big boost and it may have made me work even harder and focus more on my goals. Also, when you write a grant, you have to outline your project and think about pitfalls, alternatives, and all the potential outcomes of your experiment – these are all good things to know before starting a project.

Did you get support in your move back to Germany as a dual-career couple?

Many people helped. The CellNetworks Excellence Cluster, the ZMBH, and the DKFZ all worked together to find a suitable position for my husband as a chemist at the DKFZ. The only advice I can give is that it is important to mention early on that there is a "dual-career issue". I think most universities/employers are willing to help, but it is a very individual process. As a woman, my experience is that you will be asked about dual-career needs. Men may still have to initiate the topic themselves if it is important to them. A back-up plan will make negotiations easier, but is often a luxury that you may not have.

NEW TEAM MEMBER: SIMONE FREIMUND

Simone Freimund is a new assistant at the Boehringer Ingelheim Foundation – not the Fonds. In October, she took over from Genia Rosellen-Meckel. She is the first person applicants contact for the PLUS 3 and Exploration Grant programmes. Simone organizes the scientific advisory board meetings and supports both the Heinrich Wieland Prize and the Boehringer Ingelheim Prize. After completing her diploma as an international management assistant in 1999, she organized executive board-level workshops for the clients of an international consulting firm. From 2002 until she joined the Boehringer Ingelheim Foundation, Simone was responsible for organizing events and trade fair stands within the communication department of an international real estate company. During that time, she was able to add diplomas as a public relations consultant and social media manager to her resumée. Simone's strong background in organization and budgeting together with her international experience proved its worth when she joined us in the middle of the preparations for the 50th anniversary of the Heinrich Wieland Prize in October. In her spare time, Simone loves to be with her family, cook, and listen to music.

BIF'S SISTER FOUNDATIONS

ROCK PAINTINGS, ILLEGAL IMMIGRANTS, AND PHILOSOPHERS

These are just a few of the book topics that can be found in the conference room at BIF's office in Mainz. The printing of nearly 2,600 books covering the walls has been supported by the Siblings Boehringer Ingelheim Foundation for the Humanities, which shares the premises with BIF. The smallest of the three Boehringer Ingelheim Foundations is also the oldest, predating BIF by 27 years. Its purpose is to promote the humanties, however, focusing on works related to the German culture because of their authors, topics, or publishers. Nowadays it awards print subsidiaries, mostly for PhD theses. After all, at many German universities, you are only allowed to use the title once your thesis is printed in book form. This is something that can set back art history students tens of thousands of euros, since their work traditionally contains many illustrations and photographs. Funding recommendations are made by an honorary board of scientific advisors. Its four members sift through around 300 applications a year, each averaging 70 to 80 pages. Around 40% will be funded with amounts of up to 10,000 euros out of the foundation's annual budget of 360,000 euros. However, the authors need to contribute to the printing costs as well. They also need to acknowledge the funding in the printed book – and send one copy to the foundation, thereby adding to its growing collection.

Check out the website at www.boehringer-geisteswissenschaften.de

PROFILES

Dr Henrik Bringmann,
Max Planck Institute for Biophysical Chemistry, Göttingen, Germany
Fellowship: 2004–2006

Dr Martin Denzel,
Max Planck Institute for Biology of Ageing, Cologne, Germany
Fellowship: 2005–2008

Dr Baris Tursun,
Max Delbrück Center, Berlin, Germany
Fellowship: 2002–2005

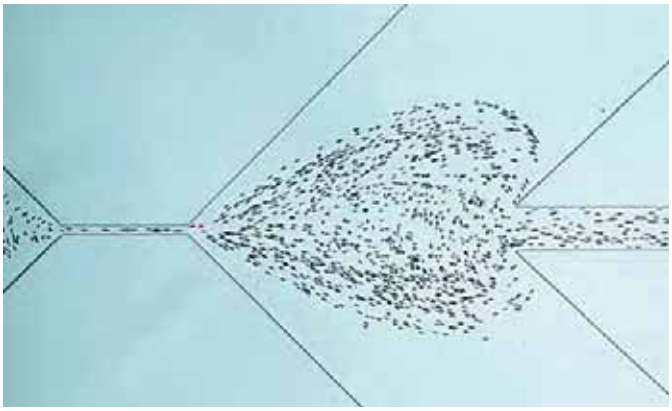
Three BIF fellows received ERC Starting Grants in December 2014. Dr Henrik Bringmann, leader of the "Sleep and Waking" group at the Max Planck Institute for Biophysical Chemistry, Göttingen, Germany, received a grant for his proposal "The Mechanism of Sleep Control through a Sleep-Active Sleep-Promoting Neuron". Dr Martin Denzel, shortly after being named leader of the group "Metabolic and Genetic Regulation of Ageing" at the Max Planck Institute for Biology of Ageing, Cologne, Germany, received a grant to research the role of protein quality control in ageing. Dr Baris Tursun, leader of the group "Gene Regulation and Cell Fate Decision in *C. elegans*" at the Max Delbrück Center, Berlin, Germany, will use his grant to study the molecular mechanism of direct reprogramming in cells. In 2014, for the first time, Germany topped the list of received grants, with 70 of 328 grants going to German-based institutions.

Photos: Carsten Costard (top left); Max Planck Institute for Biophysical Chemistry (top right), Göttingen; Max Planck Institute for Biology of Ageing, Cologne (right middle); Max Delbrück Center for Molecular Medicine, Berlin (right bottom)

PAPERS IN THE SPOTLIGHT

In "Papers in the Spotlight" we present papers from current BIF fellows and alumni. The selection criteria are based not only on scientific merit, but also on the general interest of the topic. If you would like to see your paper here, contact Kirsten at kirsten.achenbach@bifonds.de.

CELL FORM FOLLOWS FUNCTION



In fast-flowing liquids, cells are forced into bullet-like shapes, which in turn can be used to identify them.

Identifying different cell types with high-throughput methods without labelling or molecular markers? Thanks to BIF fellow Oliver Otto and his supervisor Professor Jochen Guck at the Biotechnology Center, TU Dresden, Germany, this is now possible. In fast-flowing liquids, shear stress deforms cells from more or less rounded to bullet-shaped. How far and how fast depends on the cells' stiffness, which in turn depends on its type and state – red blood cell or leukocyte, healthy or cancerous, dividing or not. So far, methods using such shape changes have been too slow for many applications. The new method – real-time deformability cytometry (RT-DC) – speeds things up by a factor of 10,000, analysing several hundred cells per second. The team created an image-analysing algorithm that measures cell shape change and size. They developed a suspension medium in which cells deform at speeds slow enough to keep them intact but fast enough for the throughput of a flow cytometer. But most importantly, they showed that at these speeds you can match cell shape change to functional type. "We can already track the differentiation of blood stem cells and identify the activation of very small leukocyte sub-populations in whole blood," explains Oliver. "On top of that, our method works with all animal cells." RT-DC promises to profoundly change the way cell samples are analysed, needing no intrusive and time-consuming labelling of cells. It cannot yet sort cells, but Oliver is confident: "That is only a question of engineering and time – at most a year."

REFERENCE

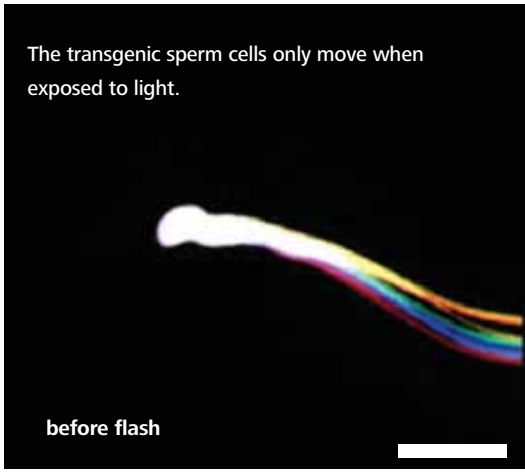
Otto O, Rosendahl P, Mietke A, Golfier S, Herold C, Klaue D *et al* (2015) Real-time deformability cytometry: On-the-fly cell mechanical phenotyping. *Nat Methods* **12:** 199–202

Oliver Otto, fellowship 2008–2011

ENLIGHTENED SPERM

Fertilization is usually not dependent on light. Thanks to optogenetics it now is – at least for a mouse strain in the labs of BIF alumna Dagmar Wachten and Benjamin Kaupp, member of BIF's Board of Trustees, at the Center of Advanced European Studies and Research (caesar) in Bonn, Germany. Sperm cells are propelled by the beat of their flagella, the frequency of which is controlled by the level of messenger cyclic adenosine monophosphate (cAMP) in the cell. In wild-type mice, cAMP is produced by the soluble adenylyl cyclase SACY, which in turn is controlled by Ca^{2+}-influx. In the soil bacterium *Beggiatoa*, cAMP is produced by the adenylyl cyclase bPAC, when activated by light. After the insertion of bPAC into mice sperm cells, it generated additional cAMP upon illumination. This made the transgenic sperm swim faster than normal sperm. In a second step, the researchers crossed the bPAC mice with SACY-deficient mice. In the dark, the sperm of these mice did not move. After light stimu-

Photos: ZellMechanik Dresden (left); Oliver Otto (bottom left)



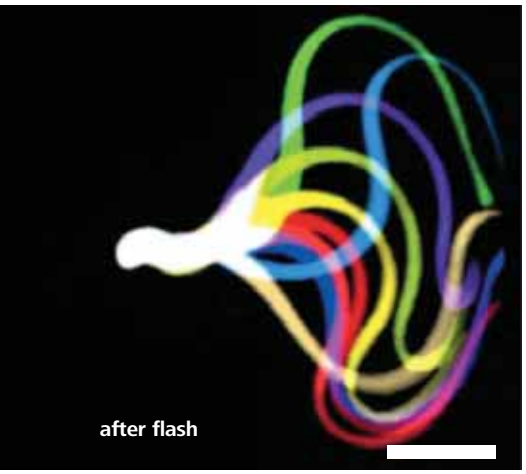
lation, however, bPAC produced cAMP and the sperm started to swim and even fertilize eggs. The light pulses used in this study are non-invasive, easily controlled and administered. Therefore, optogenetics allows for the disentanglement of cellular events that are independent of side effects caused, for example, by pharmacological tools. With these advantages, the authors were able to show that cAMP does not stimulate Ca^{2+}-influx, and thereby resolved a longstanding controversy. Thus, this work offers a tool to study complex signalling pathways with high spatial and temporal precision – not only in mammalian sperm, but also in other ciliated cell types.

REFERENCE

Jansen V, Alvarez L, Balbach M, Strünker T, Hegemann P, Kaupp UB, Wachten D (2015) Controlling fertilization and cAMP signaling in sperm by optogenetics. *Elife*, doi: 10.7554/eLife.05161

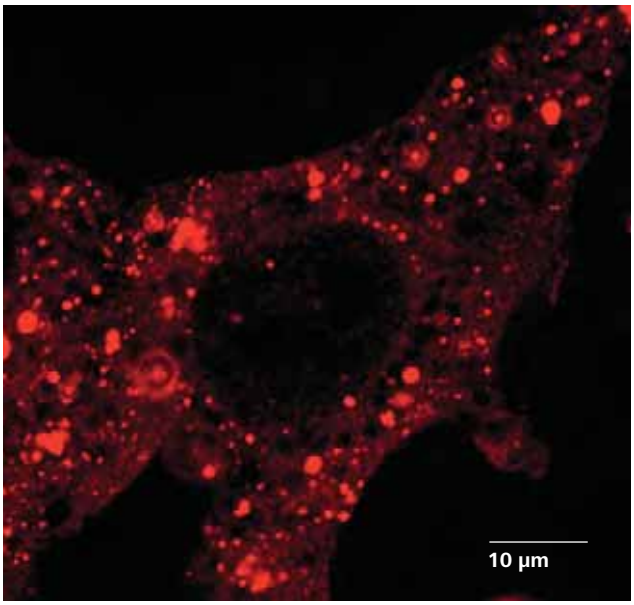
Dagmar Wachten, fellowship 2003–2006

Photos: Elife (bottom middle); Corinna Bernsdorff (caesar) (Portrait middle); dkfz.de (right middle)



NOT FOES, BUT PARTNERS IN SIGNALLING

In human cells, hydrogen peroxide (H_2O_2) works in tandem with peroxiredoxins to regulate target proteins despite its small size. This is the surprising result of a study by BIF alumnus Dr Mirko Sobotta from the German Cancer Research Center (DKFZ) and his supervisor Dr Tobias Dick. It was mainly seen as a dangerous by-product of cellular respiration that is neutralized by peroxiredoxins like other reactive oxygen species. But about ten years ago, it was found that most H_2O_2 in the cell is involved in cell signalling and regulation. It translates outside clues such as hormones or growth factors into cellular reactions. However, it was not known how the tiny H_2O_2 molecule manages to selectively regulate proteins or how it evades the highly efficient peroxiredoxins long enough to do its work. It turns out that H_2O_2 does get scooped up by peroxiredoxins. Contrary to belief, however, it is not neutralized but its oxidizing equivalent is transferred to the target protein. As an example, Sobotta and Dick showed that this redox relay dampens the transcription of STAT3, involved in inflammation and tumour development. Their results suggest that such redox relays are common in mammalian cells and that peroxidases may switch between scavenging excess oxidative equivalents and oxidative signalling.



Peroxiredoxin-2 and the transcription factor STAT3 meet in clearly defined areas within the cell (red flourescence).

REFERENCE

Sobotta MC, Liou W, Stöcker S, Talwar D, Oehler M, Ruppert T *et al* (2015) Peroxiredoxin-2 and STAT3 form a redox relay for H_2O_2 signaling. *Nat Chem Biol* **11:** 64–70

Mirko C. Sobotta, fellowship 2010–2012

BIF FELLOW'S GUIDE TO ...

AARHUS


1


2


3


4

Travelling is fun – especially if you get insider tips from locals! In each edition of FUTURA, one fellow shows you around his or her city. In this edition your guides are Sofiia Mortensen and Jan Herudek. They report from Aarhus, the second largest city in Denmark.

FACTS & FIGURES

Country: Denmark
Population: About 260,000
Area: 90 (urban) or 470 (municipal) km²
Students: About 44,500
Famous for: Vikings, beer, sailing, Danish design
Websites: www.visitaarhus.com

WHERE TO STAY

Cabinn Hotel: Modern and cosy hotel for a reasonable price.
Hotel Scandic Aarhus City: Central location and featuring modern Danish interior design.
House in Downtown Aarhus: Bed and Breakfast in a charming old house.

NIGHTLIFE

Train: Have fun in one of Denmark's leading and largest nightclubs.
Sherlock Holmes: This pub is loved for its live music five days a week and good drinks.
Tir Na Nóg: Visit an authentic Irish pub with Irish music and bartenders with a genuine Irish accent!
Herr Bartels: Offers the most fantastic choice of delicious cocktails.

RESTAURANTS

Römer Brunch & Bistro: In Aarhus, the words "cool" and "lounge" are synonymous with Römer.
Sharks: Excellent burgers and a great place for billiard fans.
Olive: Stylish French restaurant close to the harbour. You can bring your own wine!

ACTIVITIES

Den Gamle By (The Old Town): Must-see open-air museum of Danish urban history and culture in the heart of Aarhus. 1
Sailing: Because Aarhus is a very windy city, it is a paradise for sailors with plenty of marinas and competitions. 2
Deer park: Touch and feed wild deer surrounded by beautiful forests.
Aarhus by bike: Explore one of the best bike cities in the world on two wheels.

BEST SIGHTS

ARoS: In the art museum you can visit the "your rainbow panorama" in a circular, 150-metre panoramic path with a 360-degree view of the surrounding city. 3
Risskov: There is an excellent view over the Aarhus bay from the western side of the beautiful Riiskov forest.
Moesgaard Museum: A brand-new museum of anthropology.
University Park: A charming, relaxed park next to the university. 4

Contributors wanted! If you would like to introduce your city to the readers of FUTURA, contact Kirsten at kirsten.achenbach@bifonds.de.


Sofiia Mortensen and Jan Herudek

Names Sofiia Mortensen and Jan Herudek
Nationalities Russian and Czech
Ages 25 and 26
University Aarhus University, Department of Molecular Biology and Genetics

Photos: Jan Herudek und Sofia Sirotkina; schmidt hammer lassen architects (No 3)

PROFILES

Prof. Simon Elsässer,
Science for Life Laboratory at the Karolinska Institutet, Stockholm, Sweden
Fellowship: 2008–2010

In January 2015, Simon joined the ranks of BIF professors (now 180). He is assistant professor at the Science for Life Laboratory at the Karolinska Institutet in Stockholm, Sweden. His group in the Division of Translational Medicine and Chemical Biology will focus on applying new synthetic and chemical biology methods to understand chromatin structure and function.

Prof. Michael Hoch,
University of Bonn, Germany
Fellowship: 2005–2008

Michael will be heading the rectorate of the University of Bonn, Germany, as of 1 May 2015. He was selected for the six-year term unanimously. Hoch leads the department "Molecular Developmental Biology" and focuses on how metabolism influences growth and how energy homeostasis impacts on the activity of organ systems such as the immune system in health and disease.

Dr Carsten Linnemann,
The Netherlands Cancer Institute, Amsterdam, Netherlands
Fellowship: 2009–2010

Carsten received the institute's 2014 Antoni van Leeuwenhoek Prize on 12 January 2015 for his work on cancer immunotherapy (see Futura 2/2014). The award recognizes outstanding young researchers at the Netherlands Cancer Institute and comes with 6,000 euros in prize money.

Prof. Mary O'Connell,
Central European Institute of Technology (CEITEC), Brno, Czech Republic
Fellowship: 1990–1992

Mary has been appointed professor to an ERA chair in the Molecular Medicine Research Programme of the Central European Institute of Technology (CEITEC) in Brno, Czech Republic. ERA chairs are funded by the EU Horizon 2020 programme to attract top academics to less-developed regions. Mary will continue her work on inheritable information and its modification in connection with diseases of the immune system.

Prof. Jan Riemer,
University of Cologne, Germany
Fellowship: 2004–2006

Jan joined the Faculty of Mathematics and Natural Sciences of the University ofCologne on 1 February 2015. His research focuses on mitochondrial protein import, oxidative protein folding in mitochondria and redox signalling and its attendant physiological consequences.

Dr Matthias Rosenwald,
Medical Scientific Liaison for Diabetes at Janssen Cilag AG in Zug, Switzerland
Fellowship: 2009–2012

Matthias received the Pfizer Research Prize 2015 for turning white fat cells in mice into brown fat cells, which could help to devise therapies for obesity. The prize is one of the most prestigious Swiss science awards and comes with 15,000 Swiss francs. It recognizes the work of outstanding young scientists in Switzerland.

UPCOMING EVENTS

10–11 JULY 2015
Meeting of BIF's Board of Trustees in Boston, USA. The trustees decide on the allocation of fellowships, review the proposals for the International Titisee Conferences, and settle all the foundation's matters of fundamental importance.

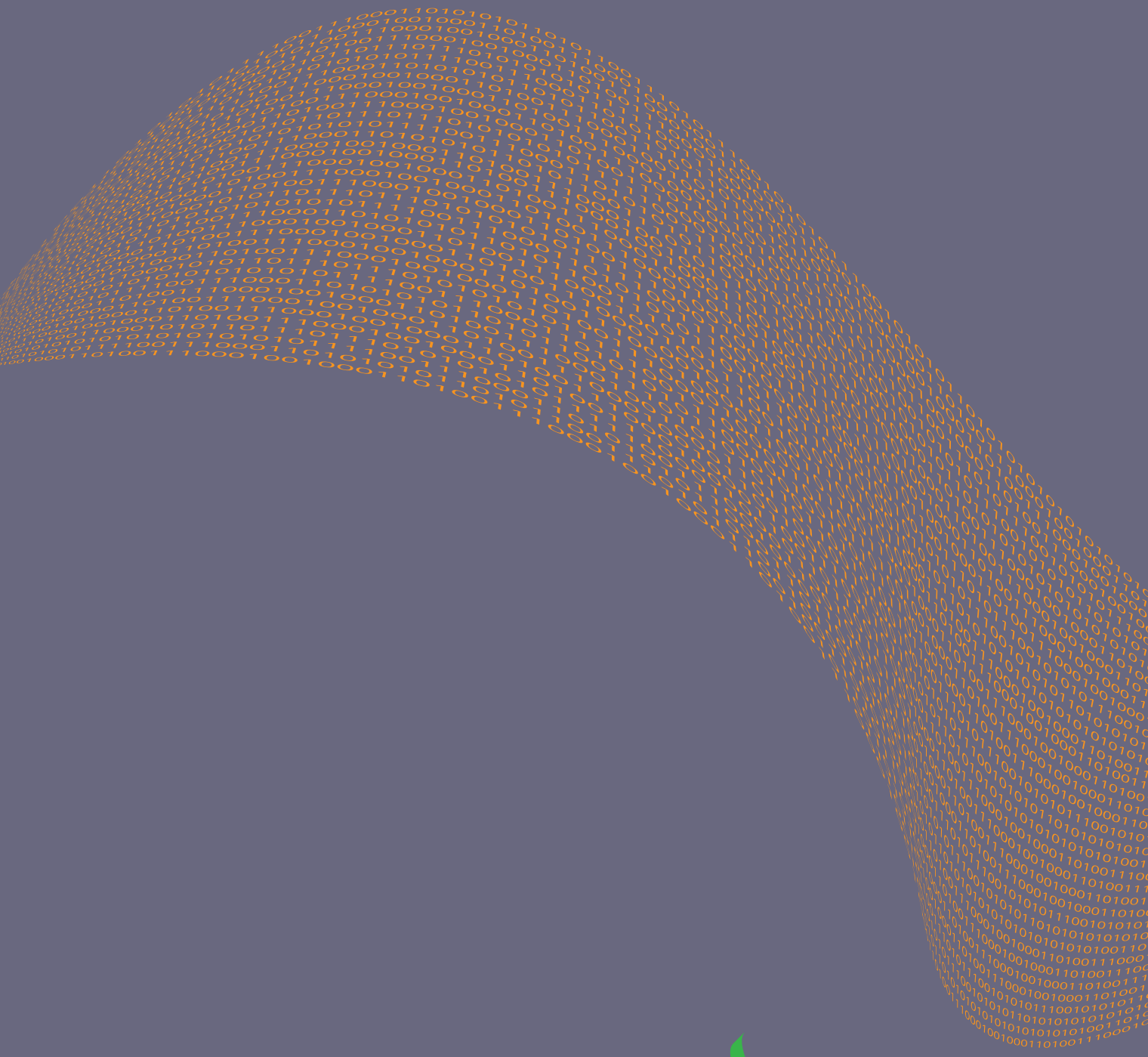
26–28 JULY 2015
Annual meeting of former alumni of all of BIF's programmes – based in Europe. The seminar takes place at Gracht castle in Erftstadt/Liblar near Cologne, Germany. This year's title is "Are We Alone? A Scientist's Guide to the Universe". Further details will be sent with the invitation.

8–14 AUGUST 2015
Seminar for current PhD fellows working in Europe in scenic Hirschegg (Kleinwalsertal), Austria. On the agenda: project presentations and discussion, career topics, and guided hiking tours in the surrounding Alps. Further details can be found in the invitation.

Need an update on upcoming events? Check our website at www.bifonds.de!

Have you received an award? Changed positions in academia or industry? Please let Anja (anja.hoffmann@bifonds.de) or Sandra (sandra.schedler@bifonds.de) know, so that we can share it with the BIF network.

Photos: Karolinska Institutet (top left); Jan Riemer; Carsten Linnemann; Volker Lannert (top right); CEITEC (middle right); www.pfizerforschungspreis.ch (bottom right)



Boehringer Ingelheim Fonds
Stiftung für medizinische
Grundlagenforschung

Schusterstr. 46-48
55116 Mainz
Germany
Tel. +49 6131 27508-0
Fax +49 6131 27508-11
E-mail: secretariat@bifonds.de
www.bifonds.de

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