FUTURA

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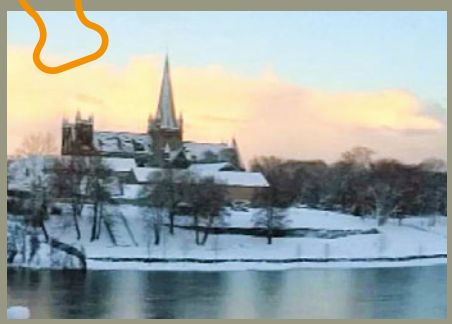
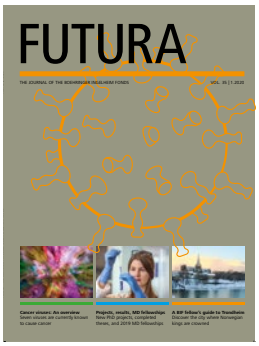
Cancer viruses: An overview
Seven viruses are currently known to cause cancer

Projects, results, MD fellowships
New PhD projects, completed theses, and 2019 MD fellowships



A BIF fellow's guide to Trondheim
Discover the city where Norwegian kings are crowned



The cover illustration shows a simplified model of Kaposi's sarcoma-associated herpesvirus (KSHV), which is one of seven viruses currently known to cause cancer. It was discovered by Patrick Moore of Columbia University, NY, USA, who managed to separate human and tumour DNA sequences in a sample of Kaposi's sarcoma tissue taken from an AIDS patient.

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CANCER VIRUSES: AN OVERVIEW

FELLOWS

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PHD RESULTS

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FOUNDATION

HALFWAY AROUND THE WORLD

PUBLISHING INFORMATION

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RESEARCH IN THE SPOTLIGHT

»The current pandemic provides a lesson in rigorous scientific reasoning.«

Since the start of the current pandemic, biomedical science has been at the center of public attention – not only its results, but its research processes, methods, and reasoning.

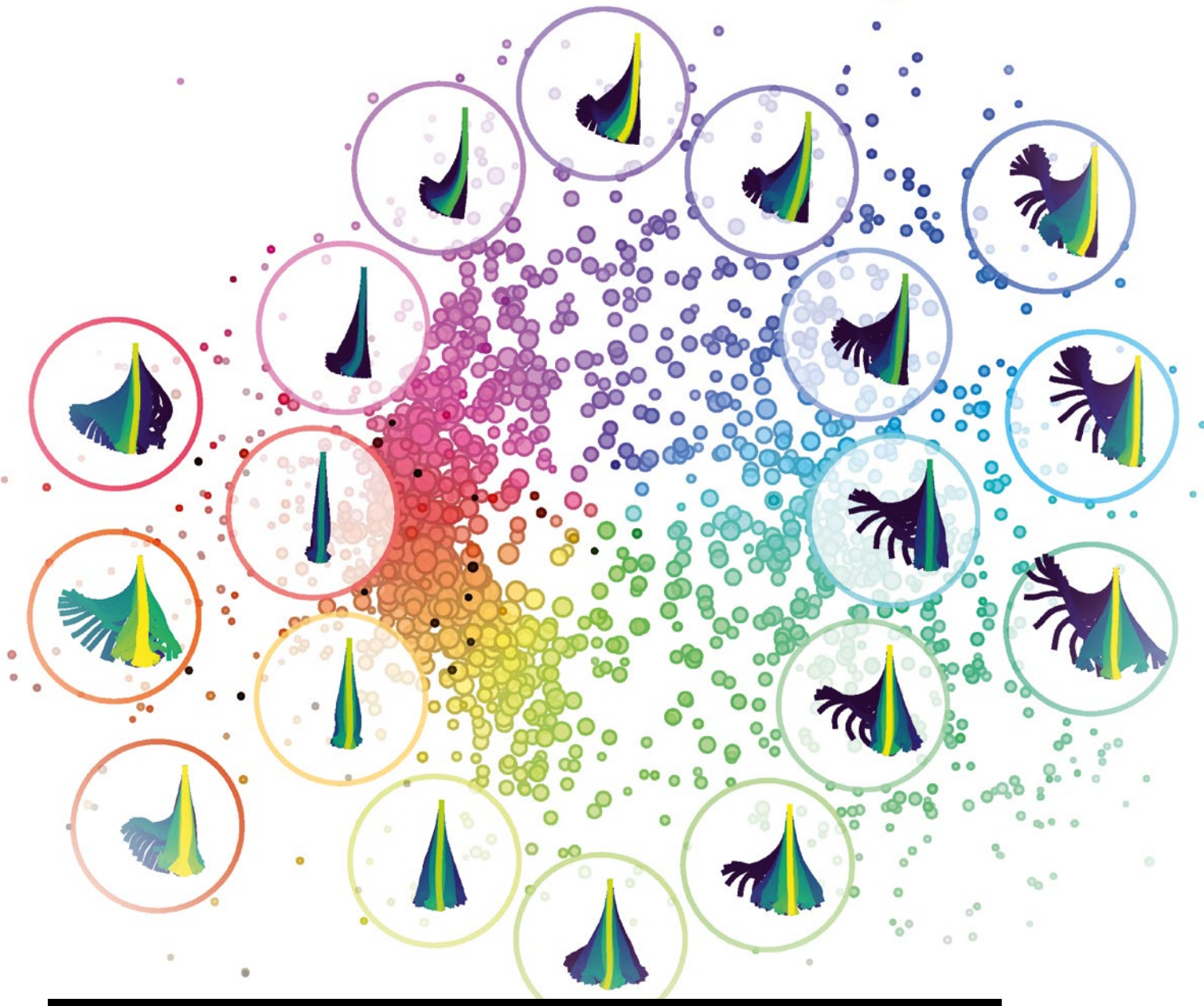
Usually, research results reach the public after they have withstood the scrutiny of the scientific community, after the usual exchange of arguments that gradually leads to better designed experiments and studies, more data, and a better interpretation of data. Now, this normally invisible process about what conclusions can be drawn depending on data and study design is extensively covered in the media. And even studies at the pre-print stage are hotly debated.

This is a unique chance. An unprecedented number of people around the world are watching almost in real time as scientists investigate this new culprit, generate data, and base their conclusions on these data. The current pandemic showcases the lengthy and thorny path from observation, idea, and hypothesis to robust data and facts. Ideally, it will provide a lesson in rigorous scientific reasoning.

Probably more than ever before, it also exposes the public to the uncertainties, limitations, and dynamics inherent in the research process. What seems "right" today may need to be adjusted or may even be deemed "wrong" tomorrow in light of new and better data. This may be overwhelming to many who long for reassurance or firm answers. And many may be left with the impression that there is not much of a difference between the reliability of a scientist talking about his or her field's new data and some random post in a social media blog by a self-proclaimed expert.

But the good news is, the trust in scientists seems to be increasing as a result of their current visibility: a new survey in Germany shows that compared to some years ago, the public's overall trust in scientists, professors, and physicians has significantly risen. And many more agree in 2020 that researchers changing their conclusions in light of new data is not a sign of a lack of knowledge, but part of the scientific process of generating knowledge.

For this, we need to be grateful to the scientists around the world who have had the courage to face the often harsh public spotlight, who have tirelessly explained their positions, put data in context, given advice to politicians, and talked endlessly to journalists almost around the clock. They are not only excellent scientists, but also excellent and authentic communicators in the best sense of Aristoteles' three means of persuasion: logos, ethos, and pathos. They are scientists who are realistic and modest enough to know that they can only offer their discipline's share of arguments, and that political decisions have to take into account more. A big thank you for helping us navigate this overwhelming tide of data and noise!



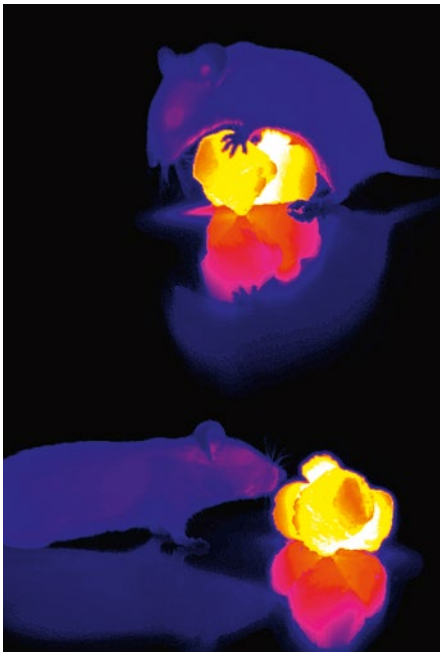
EYES ON THE PREY

By Duncan Mearns and Herwig Baier, Max Planck Institute of Neurobiology, Martinsried, Germany

The graph shows an analysis of thousands of tail movements of zebrafish while hunting. These fine movements, faster than the eye can follow, could only be distinguished by using high-speed cameras and an especially designed machine-learning algorithm developed in Herwig Baier's lab. The algorithm revealed that the half-millimetre long larvae always use a stereotyped sequence of behavioural components – orientation, approach, and capture – even though the exact moves vary with their position to and distance from the prey. The results show that the larvae strike when the prey is right in front of and slightly above their head. Such knowledge is important to find the neural circuits controlling a behaviour, which in this case should be in both brain hemispheres in order to judge location and distance.

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.

Photo: MPI of Neurobiology/Mearns



Mice investigating warm food.

HEAT PERCEPTION COMES IN FROM THE COLD

A team of researchers at the Max Delbruck Center for Molecular Medicine in Berlin, Germany, have upended the long-standing assumption that the signals for cold or warm are transmitted to the brain by different types of nerve cells, a theory known as "labelled lines". They have found that mice also need nerve cells with cold receptors to perceive warmth.

When analysing the signals from nerve cells in mouse paws, the researchers found no nerve cells that react only to warming. Most cells responded to both heat and pressure. So how does the mouse know whether a signal means warmth, cold, or pressure? The researchers found a group of cold-sensing nerve cells that is constantly active at an average mouse paw temperature of 27° C. Their activity increases if it gets colder, but decreases if it gets warmer.

They found that this group of cold-sensing cells is important for sensing warmth. When they genetically engineered the nerve cells of mice so that they lacked key heat-sensing channels, the mice could still feel warmth, but not as well as before. However, when they deleted a key cold-sensing channel, the mice could not feel non-painful heat anymore at all.

The team thinks that to sense warmth, the mice need both signals – increased firing from heat-sensing cells and decreased firing from cold-sensing cells. Cooling, on the other hand, is encoded only by increased activity of the cold-sensing cells. As mice sense temperature similar to humans, this might be true for us as well.

REFERENCE

Paricio-Montesinos *et al* (2020) The sensory coding of warm perception. *Neuron* DOI: https://doi.org/10.1016/j.neuron.2020.02.035

YOUR STOMACH HAS A SWEET TOOTH

Artificial sweeteners have never really been satisfying as a sugar substitute, despite activating the same taste receptors on the tongue. A team of researchers at Columbia University's Zuckerman Institute in New York, USA, may now have found an explanation. In experiments in mice, they showed that the brain responds to sugar not only on the tongue, but also in the gut.

It was known that blocking the sugar sensors on the tongue in mice did not keep them from the sweet stuff. When their tongue's sugar receptors are blocked, mice still look for it. To find out why, the researchers delivered sugar or sweetener directly to the gut of mice, while imaging neural activity. They found that sugar, but not sweetener, activated neurons in a region of the brain-stem called the caudal nucleus of the solitary tract.

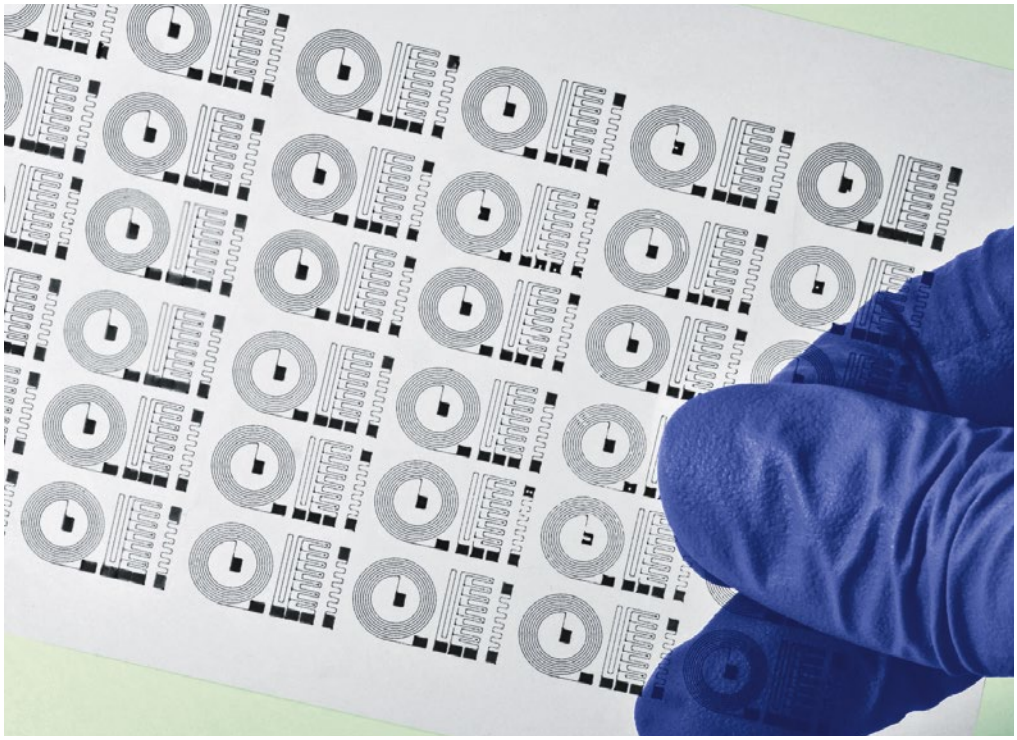
To dig deeper, the team monitored activity along the vagus nerve, which connects the gut to the brain. They found a cluster of neurons whose activity changes in response to glucose delivered to the gut. They then blocked the protein SGLT-1, which transports sugar in the gut, and found that this abolished the neural responses, suggesting it is a key sugar sensor in the gut. The team next genetically engineered mice to delete their gut-brain circuit connections, and showed that these mice did not seek out sugar. Finally, they used a technique called chemogenetics to change sugar-responsive neurons in mice brains so they could be activated by drugs, effectively hijacking the mice's neural sugar responses. Activating the neurons whenever the mice drank a sugar-free drink caused the animals to develop cravings for it, as they would for sugar.

It seems there are two sugar-sensing mechanisms, one receiving information from the tongue, the other monitoring the gut. This has implications for developing strategies for curbing sugar over-consumption, which is a significant global health problem. Researchers could target various parts of the circuit, or develop sugar substitutes that activate both mechanisms.

REFERENCE

Tan *et al* (2020) The gut–brain axis mediates sugar preference. *Nature* DOI: 10.1038/s41586-020-2199-7

Photos: Lewin/Poulet Lab, MDC (top); iStock/Tammi Mild (bottom)



A new technique may make possible the speedy, on-demand design of softer, safer neural devices.

PRINTABLE SOFT BRAIN IMPLANTS

Brain implants are used to treat conditions including depression, Parkinson's disease, and epilepsy, and typically consist of electrode arrays made from rigid materials, usually metal. This can be a problem when such devices are required long term, because, over time, rigid implants can cause inflammation, tissue damage, and ultimately scar tissue, as well as degrade in performance. But researchers at the Massachusetts Institute of Technology, USA, have developed a technology that enables the 3D printing of soft, flexible brain implants that could be used both for medical devices or electrodes for research.

The researchers first transformed a liquid conductive polymer into a viscous substance they could feed into a 3D printer, and then used this as the raw material to print several soft electronic devices. One was an electrode they tested in a live mouse; they found it was able to monitor the activity of a single neuron in a freely moving animal. They also showed that their method could produce complex multi-electrode arrays far more rapidly than the conventional method, known as lithography.

The team hope that the technology will enable other researchers to make numerous different soft electronic devices quickly, streamlining the development of soft neural interfaces. One potential application could be improving neural control systems for amputees with prosthetic limbs.

REFERENCE

Yuk *et al* (2020) 3D printing of conducting polymers. *Nature Communications* DOI: https://doi.org/10.1038/s41467- 020-15316-7

2,900

That's the number of substances suspected of acting as anti-infection agents, taken from 1,600 plant species on the Indonesian island of Java. This was one finding from an analysis of genetic relatedness, distribution, and metabolites present in 7,500 seed plant species on the island. Methods used in the study could be applied to speed up searches for other groups of bioactive compounds in other parts of the world.

Source: Holzmeyer *et al* (2020) Evaluation of plant sources for anti-infective lead compound discovery by correlating phylogenetic, spatial, and bioactivity data. *Proceedings of the National Academy of Sciences* DOI:10.1073/pnas.1915277117

THE CELL'S THERMOMETER

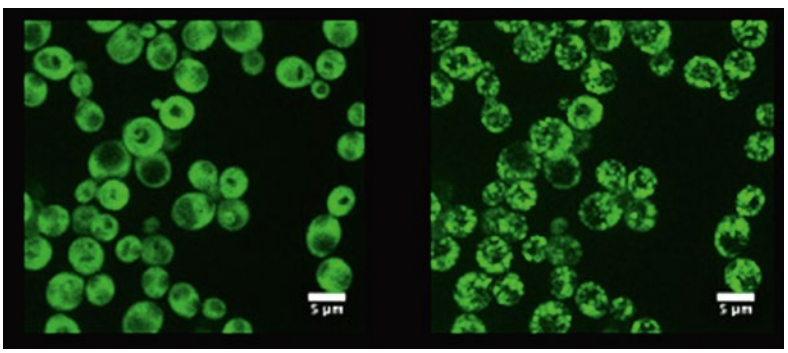
Any organism exposed to potentially harmful levels of heat stops growing and shifts into a protective mode known as the "heat shock response". This involves a drastic change in which genes are active, resulting in increased production of a family of protective proteins known as heat shock proteins, together with suppression of the usual set of "housekeeping" proteins. But scientists do not completely understand how this transformation in cellular output is achieved, or how harmful temperatures are detected.

Researchers at the Max Planck Institute of Molecular Cell Biology and Genetics in Dresden, Germany – among them BIF fellow Cecil Jegers – have now found that the protein Ded1, an essential translation factor involved in producing the proteins cells normally need, appears to be crucial for triggering the heat shock response. Under normal circumstances, Ded1 is evenly distributed, dissolved in the plasma inside cells. However, it becomes insoluble at higher temperatures, causing it to undergo a process called phase separation in which it precipitates out to form solid clumps. The team conducted experiments in yeast cells, showing that this process acts as a temperature sensor in cells. The protein clumps interact with RNA molecules to form soft gel-like condensates, which revert once temperatures fall back to safe levels. As Ded1 is needed for translating RNA molecules into house-keeping proteins, this transformation switches the cellular machinery from a housekeeping state to a heat shock state.

The team also showed that Ded1 proteins from different organisms do this at temperatures corresponding to the maximum safe growth temperature for each species, suggesting this is a general mechanism common across many forms of life. Left unchecked, stressors like heat cause proteins to take deformed shapes that can be toxic (the heat shock proteins' job is to prevent this). Such misfolded proteins are centrally involved in neurodegeneration, so the work may have implications for understanding neurodegenerative conditions like Alzheimer's or Parkinson's disease.

REFERENCE

Iserman *et al* (2020) Condensation of Ded1p promotes a translational switch from housekeeping to stress protein production. *Cell* DOI: 10.1016/j.cell.2020.04.009



Ded1p protein of baker's yeast.

Photos: Massachusetts Institute of Technology (top); Technische Universität Dresden (bottom)

GLUCOSE LINKED TO SEVERE FLU SYMPTOMS

A low percentage of people with flu suffer from an immune system over-response known as a cytokine storm, which can result in hospitalization, or even death. The reasons for this remains largely unknown. Researchers in China have now linked this phenomenon to glucose metabolism, suggesting that one of the three main ways in which glucose is metabolized may interact with the immune system to promote excessive responses. The work has implications not only for influenza, but probably also COVID-19.

The researchers examined the production of immune system molecules in mice infected with influenza. They found that mice that were given glucosamine to raise their blood sugar level produced significantly more inflammatory molecules known as cytokines and chemokines. They also analysed blood samples from flu patients collected at two Wuhan University hospitals between 2017 and 2019 and found that higher levels of blood glucose were associated with higher levels of cytokines.

The team then conducted studies in human cells that showed that influenza A virus induces one of the major glucose metabolizing pathways, the hexosamine biosynthesis pathway. During an infection, high blood sugar causes an enzyme within this pathway to interact with molecules involved in the immune system, the interferones, resulting in increased production of cytokines. These findings suggest that glucose metabolism plays a crucial role in inducing cytokine storms during flu infections, and high blood glucose levels may explain severe symptoms in some patients.

Diabetes, a disease of impaired glucose metabolism, is known to significantly increase the risk of death in patients with COVID-19, which can also involve cytokine storms. The researchers are therefore now studying how glucose metabolism may affect COVID-19 patients.

REFERENCE

Wang *et al* (2020) O-GlcNAc transferase promotes influenza A virus–induced cytokine storm by targeting interferon regulatory factor–5. *Science Advances* DOI: 10.1126/sciadv.aaz7086

PROFILE OF **THE ZEBRA FINCH**

By Mitch Leslie

Male zebra finches (*Taeniopygia guttata*) display a striped throat like their namesake, the African zebra, and learn their song – a "beep, meep, oi!" or "a-ha!" – from their fathers in a particular developmental phase. Because of this, the native bird of Australia and Indonesia has become the star of neurological research into the development of language.

A baby has a lot in common with a young male zebra finch. Both start their lives babbling and producing strings of disjointed syllables, but then hone their vocal output until they are communicating like adults. The similarities between how humans learn to speak and how zebra finches learn to sing have made this small bird one of the most important model organisms. Scientists have published more papers on zebra finches than on almost any other bird species.

Like several other model organisms, zebra finches were popular in the pet trade long before scientists recognized their value for research. In the 1950s, biologists began using the birds to study behaviour because they were "ideally suitable for laboratory observations", as one researcher put it. The finches thrive in captivity, do

not require a specialized diet, breed readily without regard to season, and mature in just three months, a very short time for birds. They also perform many of the same behaviours in a cage that they do in the wild – and they are not shy when scientists are watching.

One of their drawbacks, on the other hand, is that they require larger cages than mice because they need room to fly. Also, the shell of their eggs prevents easy access to the zygote for genetic manipulation through methods such as CRISPR. A further complication is the zygote's complex yolky structure.

Nonetheless, the birds took off as model organisms when neuroscientists began to probe vocal learning, in which animals acquire the ability to communicate through sound by imitating other animals around them. Humans show this type of learning, but other traditional animal models, such as mice, do not. Researchers have discovered that young male zebra finches learn to sing by memorizing and then rehearsing the songs of their fathers or other males. Scientists have mapped the brain circuits that permit the birds to learn songs. They have traced the complex effects of hormones on these circuits and pinpointed many of the necessary genes, many of which are also important to human speech learning.

Scientists in other fields have also capitalized on the qualities of zebra finches, investigating subjects as diverse as how animals choose mates, how they optimize their use of time as they forage for food, and how heat waves affect development.

CV OF ZEBRA FINCH (*TAENIOPYGIA GUTTATA*)

- I weigh 10 to 16 g.
- I live for two to five years in the wild.
- I eat grass seeds.
- I work mainly in neurobiology, behavioural studies, and endocrinology.

Photos: iStock/Capstoc (top); AdobeStock/EdeStudio (right)

Only a small number of people infected by cancer-causing viruses go on to develop cancer.

CANCER VIRUSES: AN OVERVIEW

By Mitch Lesley

Twenty years after the discovery of viruses, it became clear that something smaller than cells can cause cancer. Here, we describe the search for and discovery of the seven viruses currently known to cause cancer in humans.

Patrick Moore spent his career investigating infectious diseases in far-flung locations like Nigeria and Nepal, not studying cancer. In the early 1990s, however, Moore and Yuan Chang, his wife, both at Columbia University in New York, USA, decided to focus on a type of tumour that had vexed scientists for more than a decade – Kaposi's sarcoma. Patients with AIDS often developed these purple or brown tumours on the skin or other parts of the body, and their prevalence in people with weakened immune systems suggested they resulted from an infection. But researchers had tested more than 20 potential culprits without success.

A new technique that allowed researchers to separate normal and tumour DNA sequences could finally expose the guilty party, Moore suspected, so the scientists obtained a tumour from a patient with AIDS. "We subtracted all the human DNA, and what was left was the DNA that belonged to a virus," he recalls. That virus was the Kaposi's sarcoma-associated herpesvirus (KSHV). Together with colleagues, Moore and Chang, who are now at the University of Pittsburgh, repeated the feat in 2008 using a different technique and discovered a second cancer-causing virus, the →

Photo: gettyimages/Artur Debat

Merkel cell polyomavirus (MCV), which sparks a rare and aggressive type of skin tumour called Merkel cell carcinoma.

Researchers uncovered the first evidence that viruses can trigger cancer more than a century ago. In 1911, Peyton Rous of the Rockefeller Institute for Medical Research in New York revealed that he could produce tumours in chickens by injecting them with material from the tumours of other chickens. Even when Rous filtered the material to screen out cells, it still instigated tumours when he injected it into the birds, suggesting that something smaller than a cell, a virus, was responsible. Other scientists from Denmark had performed similar experiments three years earlier. But in the early 1900s, researchers knew little about viruses, which had been discovered only 20 years before. Many scientists remained skeptical that these mysterious infectious particles had any role in cancer. In 1966, Rous, who went on to demonstrate that a different virus promoted tumours in rabbits, received the Nobel Prize for his findings, which is the longest gap between discovery and award in Nobel history.

In humans, the first virus that causes cancer came into view in the early 1960s, when British scientists spotted viral particles in cells from a patient with Burkitt's lymphoma, a cancer of the immune system. Dubbed the Epstein-Barr virus (EBV), the pathogen turned out to be the culprit in several other types of cancer. In the 1970s, Harald zur Hausen, then at the University of Erlangen-Nuremberg in Germany, and colleagues began research that linked human papillomavirus (HPV) to cervical cancer, a finding that earned him a share of the Nobel Prize in 2008. The rogue's gallery of confirmed human cancer viruses also includes the hepatitis B and C viruses, which produce liver tumours, and HTLV-1, which spurs a type of leukemia.

These seven viruses turn out to be some of the most important causes of human cancers. According to the World Health Organization (WHO), 25% of cancers in the developing countries stem from viral infections. Several of these cancers result in large numbers of deaths. HPV, for example, is responsible for most cases of cervical cancer, which the WHO estimates kills more than 300,000 women every year.

The viruses causing this misery are diverse. Five use DNA as their genetic material, while the other two use RNA. MCV only needs six proteins to turn one of our cells cancerous, whereas KSHV relies on almost 100. However, the viruses have one thing in common – they are adept at manipulating our cells to generate more viruses. Human cancer viruses are challenging to study because some do not infect animal models, notes Henri-Jacques Delecluse of the German Cancer Research Center in Heidelberg. Nonetheless, scientists have uncovered many of the tricks that enable these viruses to replicate in our cells and persist in our bodies. KSHV, for example, has stolen several human genes involved in signalling pathways that help control cell growth and survival or that regulate the immune system. "These signalling pathways are really important for the virus," says Moore. One gene the virus picked up encodes a type of cyclin, a protein that prompts cells to advance through the cell cycle – and thus produce more viruses. Another stolen gene prevents apoptosis, which is one of the body's important weapons against virally infected cells.

EBV exploits the immune system's B cells to ensure its replication. Antigens from pathogens normally trigger these cells to reproduce and produce specific antibodies against the invader. But EBV invades the B cells and spurs them only to divide. "The virus fools the B cell into thinking there is an antigen and it must proliferate," says Delecluse. And each time a B cell divides, it also replicates the virus.

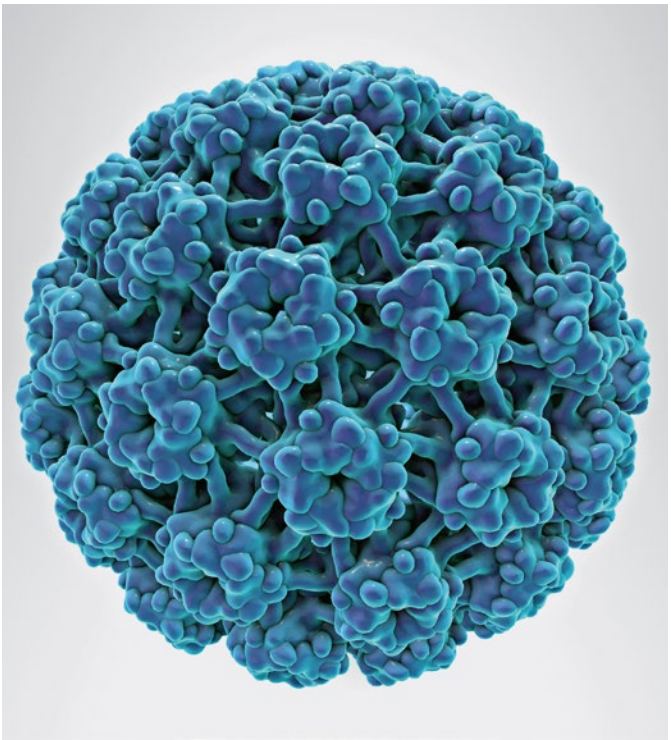
Causing cancer is not the viruses' goal, Moore notes, but the changes they engineer for their own benefit can lead to abnormal cell division. Viruses induce cancer in at least three ways, says Karl Munger of Tufts University School of Medicine in Boston. The first way is through oncogenes that unleash abnormal cell growth. The HPV genome encodes the proteins E6 and E7 that block two key tumour suppressors, p53 and the retinoblastoma protein, which help guard us against cancer. The second mechanism involves inciting

Causing cancer is not the viruses' goal, but the changes they engineer for their own benefit can lead to abnormal cell division.

long-term inflammation that favours cancer development. The hepatitis B and C viruses spur liver tumours in this way. The third mechanism is disrupting the genome. Munger and his colleagues have, for example, shown that HPV stimulates genome instability by causing cells to make extra centrosomes, structures that help orchestrate the separation of chromosomes during mitosis.

The cancer-causing viruses pose some puzzles. For one thing, they infect far more people than ever develop cancer. Researchers estimate that 90% to 95% of the world's population has contracted EBV, for instance. Teens and young adults may get the flu-like disease infectious mononucleosis, but most people suffer few or no symptoms and never have EBV-related cancers. "Although we are all infected, why do only a few people go on to develop a tumour?" is the key question, Delecluse says.

Scientists are beginning to identify the underlying mechanisms. MCV is almost as common as EBV, with up to 80% of the world's population infected. Yet, there are fewer than 2,000 cases of Merkel cell carcinoma in the United States each year, a recent analysis of national statistics found. "Most of us harbour it in our skin, and it's no problem," says Moore. Researchers now know that the virus does not trigger cancer unless one of its genes, which encodes a protein known as the large T antigen, undergoes a mutation. The mutation prevents the virus from replicating inside the skin cell where it resides. However, the virus's DNA remains in the genome of its host cell, which continues to manufacture cancer-promoting viral proteins, eventually leading to a tumour. Because the mutation is rare, the cancer is rare as well.



Some human papilloma viruses like HPV-16 can cause cancer, but a vaccine for HPV-16 already exists.

EBV presents a further mystery because it results in different cancers in different parts of the world. In Sub-Saharan Africa, EBV-caused Burkitt's lymphoma is the most common tumour in children. In southern China and other parts of Asia, nasopharyngeal carcinoma, a tumour that grows at the back of the nasal cavity, is 50 times more prevalent than in the United States or Europe. Paul Farrell of Imperial College in London notes that EBV may also be responsible for 8% to 10% of stomach cancers, and seems to be particularly important in some parts of Eastern Europe, Asia, and South America.

Researchers are still trying to pin down the reasons for this geographic variation. In hopes of identifying mutations that are involved, Farrell and his team have analyzed the genomes of more than 200 strains of the virus from around the world. Determining which alterations are significant is difficult, says Farrell, because the virus has so many genes that contribute to the oncogenic process. Still, he and his team have found that sequence differences that result in the virus replicating more efficiently are associated with African Burkitt's lymphoma and Asian nasopharyngeal carcinoma. And other differences have emerged. In December 2019, Delecluse and his colleagues revealed that one strain that causes nasopharyngeal carcinoma is particularly good at inducing inflammation, which promotes this type of cancer. He adds that factors such as diet and other infections may also have a role.

Most scientists agree that the described seven types of viruses cause cancer. But thousands of varieties of viruses enter our bodies. Whether any of them is also oncogenic remains controversial. A virus that has drawn suspicion is cytomegalovirus, which infects much of the population and occasionally prompts a cold-like illness. Some researchers argue that it can also trigger breast cancer and glioblastoma, a type of brain tumour. Other proposed cancer culprits include the monkey virus SV40 and HSV-1, which causes cold sores. But Moore is skeptical. "We have to be very, very cautious in saying a virus causes cancer. The evidence has to be overwhelming." He says that none of the proposed viral candidates meets the criteria.

A study published in 2020 bolsters this view. Peter Lichter of the German Cancer Research Center and colleagues analyzed complete genome and RNA sequences for more than 2,600 cancer samples. The researchers found evidence of the accepted →

Photo: iStock/Dr_Microbe

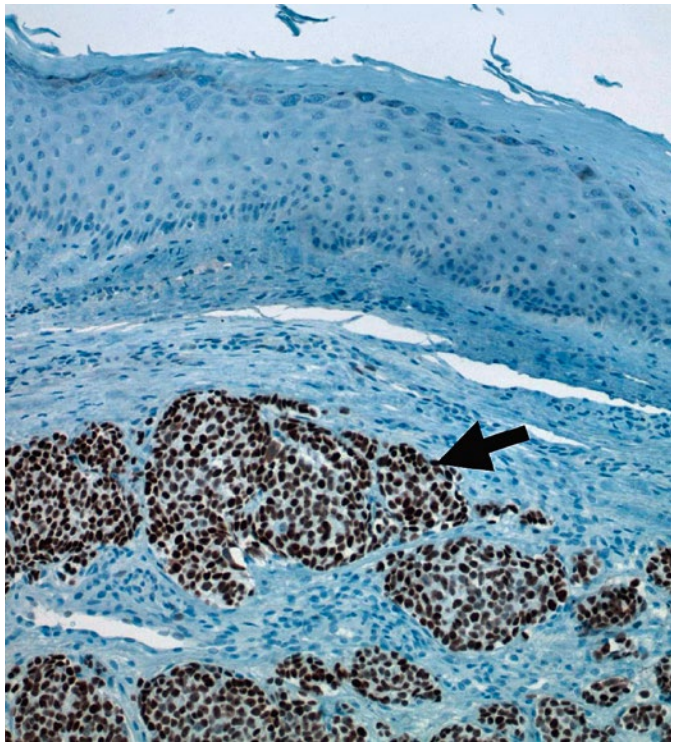
cancer-causing viruses in the tumours. However, they did not find signs of other viruses. For instance, they did not detect cytomegalovirus in brain tumours.

A different issue is whether unknown viruses lead to cancer. Munger says it is a possibility. "I think that there are other viruses that cause cancer that we haven't discovered yet." But if these viruses exist, they may be elusive. "We may be at the stage where we have found most, if not all, of the easily identified viruses that cause cancer," Moore says.

Even if there are only seven culprits, the discovery of viruses' role in cancer has already had a profound impact on patients. Vaccines against the hepatitis B virus and HPV have prevented millions of cases of cancer, and some scientists predict that widespread vaccination could even eradicate cervical cancer. There is no vaccine against KSHV, but Moore says that developing one could also lead to the elimination of Kaposi's sarcoma, which is the most common cancer in Sub-Saharan Africa. The evidence indicates that the virus is vulnerable, he says. "It's an immunologic weakling. If we give it a gentle push, it will go over the cliff."

Preventive vaccines may not be the solution for other cancer-causing viruses, however. Researchers have not been able to design an effective vaccine against EBV. One candidate produced by a pharmaceutical company and tested in 2007 showed some benefits – it protected college students against mononucleosis. But it did not stop them from being infected by EBV, and the company shelved it. Several groups are now working on other potential EBV vaccines. Delecluse's team is one of them, but whether any of the approaches can prevent infection – which is necessary to stop the virus from inciting cancer – remains unclear, he cautions.

Scientists are investigating a variety of approaches besides preventive vaccines to stimulate the immune system to attack virally induced cancers. Later this year, researchers in the United Kingdom plan to launch a clinical trial of a therapeutic vaccine that may halt the development of cervical cancer in patients who are already infected with HPV. And for more than 20 years, Cliona Rooney, now at Baylor College of Medicine in Houston, and colleagues have been coaxing T cells to take on tumours. Several types of cancers are vulnerable to this approach, she says. "If the virus is in the tumour and expresses proteins, you can target those proteins." She and her colleagues expose T cells to antigens from tumours. Cells that recognize the antigens multiply, and then the researchers transfer those cells into patients. They have shown that the strategy works in patients who have undergone haematopoietic stem cell transplants to treat leukemia or other conditions and are susceptible to B cell tumours triggered by EBV. The cells "are very safe and potent, but we need to make them more effective", says Rooney.



Photomicrograph of Merkel cell carcinoma infiltrating the skin (arrow). Tumour cell nuclei are stained brown by an antibody to the Merkel cell polyomavirus large T antigen.

The scientists are now genetically engineering the T cells to improve their performance. In 2018, Rooney and her coworkers demonstrated that T cells that had been modified to prevent tumours from shutting them down were beneficial in patients with Hodgkin's lymphoma, a cancer triggered by EBV. Along with Carlos Ramos of Baylor College of Medicine and colleagues, they are also running a clinical trial of the upgraded T cells in patients with tumours caused by HPV. Further study of viruses like EBV and HPV may open up additional opportunities for treatment – and might enable us to prevent even larger numbers of cancers and reduce suffering worldwide.

Photo: Original creation/Patrick S. Moore

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.

Shared first authorship is indicated with an *.

ESTABLISHMENT AND REMODELLING OF THE DENDRITIC CELL NETWORK IN TISSUES

cf. BIF FUTURA, VOL. 31 | 2.2016

MAR CABEZA-CABRERIZO
Discipline: Immunologist, MSc
Institute: The Francis Crick Institute, London, UK
Supervisor: Prof. Caetano Reis e Sousa

Dendritic cells (DCs) are leucocytes that act as sentinel cells, sensing the extracellular environment and initiating immune responses against infection and cancer. By being strategically positioned in all tissues, DCs can encounter pathogens and subsequently instruct other immune cells to target and kill them. DCs develop from a haematopoietic progenitor in the bone marrow (BM) that travels via the blood in the form of a pre-DC to seed tissues. What happens when pre-DCs colonize different organs, whether this is affected by infection, and how BM production is matched to DC demand in the periphery remain poorly understood. During my PhD, I developed new microscopy techniques to examine how pre-DCs from the BM arrive and proliferate in the tissue to establish a network of DCs. I set up a microscopy method to visualize DCs in large areas of the lung and small intestine. I also collaborated with mathematicians and data scientists to design software to quantify cell proliferation in tissues. Using these techniques, I showed that during influenza virus infection, the network of DCs in the lung is remodelled by increasing the number of lung DCs without increasing local proliferation. I found that the BM detects changes in the periphery and sends more pre-DCs specifically to the infected lung. During this process, pre-DCs move closer to the sinusoids, capillary-like structures that they use to exit the BM. My results revealed that once in circulation, pre-DCs use the C-C chemokine receptor type 2 (CCR2) to follow a gradient of ligands secreted by the infected tissue. Finally, I showed that in the infected lung, pre-DCs differentiate and generate new waves of DCs that are needed to instruct more influenza-specific immune cells. My work demonstrates that pre-DCs from the BM are an essential component of immunity against infections. This could have implications for scenarios like cancer, allergies, or vaccination, and could be used in the design of new immunological therapies.

PUBLICATIONS

Cabeza-Cabrerizo M*, van Blijswijk J*, Wienert S, Heim D, Jenkins RP, Chakravarty P *et al* (2019) Tissue clonality of dendritic cell subsets and emergency DCpoiesis revealed by multicolor fate mapping of DC progenitors. *Sci Immunol* **4**: eaaw1941

CHRONIC STRESS INDUCES NEURAL CIRCUIT REMODELLING, LEADING TO IMPAIRED MOTIVATION

cf. BIF FUTURA, VOL. 31 | 1.2016

IGNAS CERNIAUSKAS
Discipline: Neuroscientist, MSc
Institute: Helen Wills Neuroscience Institute, University of California, Berkeley, CA, USA
Supervisor: Prof. Stephan Lammel

Despite the high prevalence of major depression disorder, currently available antidepressants, which target several neurotransmitter systems in the brain, are still lacking. Less than half of patients respond to treatment, and antidepressants have many side effects. A different approach to treatment conceptualizes depression as a neural circuit instead of a neurotransmitter dysfunction. In recent years, the lateral habenula (LHb) – a small brain nucleus important in rewarding information processing – and its associated circuits have have been linked to depression. LHb is connected with dopaminergic neurons in the ventral tegmental area (VTA), a midbrain structure that has a central role in goal-oriented behaviours and learning. Changes in dopamine release have been associated with depression-related behaviours. However, the neurobiological mechanisms that contribute to the hyperactivity of LHb neurons in depression, and how this hyperactivity translates to depression behaviours, are still unknown. In my PhD project, I exposed mice to chronic mild stress and used different behavioural paradigms to assess what depression-like phenotypes they developed. Using brain slice electrophysiology, I identified a distinct neuronal population in the LHb that becomes hyperactive in a depression state. This LHb hyperactivity most likely inhibits dopamine release in the VTA and is exclusively associated with reduced motivation, a symptom of depression that is common to mice and humans. Using chemogenetics, I was able to decrease the activity of the LHb in response to stress, which abolished the reduced motivation. Lastly, using single-cell RNA sequencing, I identified genes that were overexpressed in the LHb of mice that developed a depressive state after exposure to chronic stress. My findings link a distinct behavioural phenotype observed in depressed patients to specific molecular, cellular, and circuit changes in the LHb. Identifying and targeting this and similar circuits could lead to breakthrough discoveries in depression research and to the development of novel patient-tailored antidepressants.

PUBLICATIONS

Cerniauskas I, Winterer J, de Jong JW, Lukacsovich D, Yang H, Khan F *et al* (2019) Chronic stress induces activity, synaptic and transcriptional remodeling of the lateral habenula associated with deficits in motivated behaviors. *Neuron* **104**: 899–915

SOLVING THE STRUCTURE OF A LABILE MEMBRANE PROTEIN COMPLEX

cf. BIF FUTURA, VOL. 31 | 1.2016

VADIM KOTOV
Discipline: Structural Biologist, MSc (equivalent)
Institute: University Medical Center Hamburg-Eppendorf (UKE), Hamburg, Germany
Supervisor: Prof. Thomas C. Marlovits

Membrane protein complexes have essential biochemical and physiological functions. Understanding these processes requires knowledge of the atomic organization of the proteins, but in the past, solving a membrane protein structure was challenging. Recent breakthroughs in lipid and detergent technologies, crystallization techniques, and cryo-electron microscopy (cryo-EM) have opened new avenues in this area. In my PhD project, I addressed two major challenges: finding the optimal sample buffer, and solving the structures of asymmetric components within a larger symmetric assembly. A stable protein sample is needed for any biochemical and structural analysis. The most widely used proxy for protein stability is thermostability. By combining well-established thermodynamic models of protein unfolding with modern high-throughput equipment, I estimated Gibbs free energy of unfolding on a large scale and used it as a quantitative measure of protein stability. I demonstrated that this approach decreases the false-positive rate in the nano differential scanning fluorimetry (nanoDSF) assay, which is compatible with most protein samples, including membrane protein complexes. Thus, I established a pipeline for efficient screening of buffer compositions for structural and biochemical research. In the second part of my project, I worked with other Marlovits lab members to solve the structure of the export apparatus (EA) inside an assembled pathogenic type III secretion system. The EA is a labile asymmetric membrane protein assembly that is masked by an outer protein ring with 24-fold rotational symmetry. Our structure revealed how the molecular features of the EA control the proper assembly of the entire 3.5 MDa secretion system. The results of my work broaden the range of membrane protein samples that can be characterized by cryo-EM and represent an important technological and methodological development in structural biology.

PUBLICATIONS

Kotov V, Bartels K, Veith K, Josts I, Subhramanyam UKT, Günther C *et al* (2019) High-throughput stability screening for detergent-solubilized membrane proteins. *Sci Rep* **9**: 10379

Further results of this project can be discussed on BioRxiv: 714097.

MECHANISTIC INSIGHT INTO THE ROLE OF polyP IN NEURODEGENERATION

cf. BIF FUTURA, VOL. 32 | 1.2017

JUSTINE LEMPART
Discipline: Biochemist, MSc
Institute: Department of Molecular, Cellular and Developmental Biology, University of Michigan, Ann Arbor, MI, USA
Supervisor: Prof. Ursula Jakob

Built as a linear chain of inorganic phosphate units, polyP is one of nature's simplest molecules. The Jakob lab identified polyP as a physiological modifier of neurodegenerative diseases that accelerates amyloid fibre formation *in vitro* and protects neuronal cells against amyloid toxicity. The goal of my PhD project was to characterize the interaction between polyP and two proteins involved in neurodegenerative diseases: α-syn, in Parkinson's disease; and tau, in Alzheimer's disease. Using biophysical assays, I found that the first step of α-syn fibre formation is polyP-independent. Subsequent oligomerization of α-syn is then rapidly accelerated by the presence of polyP. I showed that reversible binding of polyP to mature α-syn fibrils altered the morphology of the fibrils, making them less prone to shedding toxic intermediates. The formation of a polyP-α-syn fibre complex prevented fibrils associating with the cell membrane and significantly reduced the internalization of amyloids into differentiated neuronal cells. In collaboration with a group at the University of Pennsylvania, I showed that polyP interacted directly with monomeric tau. This interaction led to the formation of a distinct aggregation-prone conformation of tau, which we characterized using intramolecular fluorescence resonance energy transfer. Using aggregation assays, I showed that each tau isoform had multiple polyP binding sites. This suggests that polyP can act as a scaffold for one tau molecule, changing its structure, or several tau molecules, bringing them closer together and possibly accelerating amyloid formation. My results form the foundation of future studies to establish polyP as a therapeutic strategy for preventing the cellular spread of these two diseases.

PUBLICATIONS

Wickramasinghe SP, Lempart J, Merens HE, Murphy J, Huettemann P, Jakob U, Rhoades E (2019) Polyphosphate initiates tau aggregation through intra- and intermolecular scaffolding. *Biophys J* **117**: 717–728

Lempart J, Tse E, Lauer JA, Ivanova MI, Sutter A, Yoo N *et al* (2019) Mechanistic insights into the protective roles of polyphosphate against amyloid cytotoxicity. *Life Sci Alliance* **2**(5): e201900486

Cremers CM, Knoefler D, Gates S, Martin N, Dahl JU, Lempart J *et al* (2016) Polyphosphate: a conserved modifier of amyloidogenic processes. *Mol Cell* **63**: 768–780

STRUCTURAL AND MECHANISTIC ANALYSIS OF THE SMG1 KINASE AND ITS COMPLEXES

cf. BIF FUTURA, VOL. 30 | 2.2015

MAHESH LINGARAJU
Discipline: Structural Biologist, MSc
Institute:
Max Planck Institute of Biochemistry,
Martinsried, Germany
Supervisor: Prof. Elena Conti

Messenger RNA (mRNA) surveillance and quality control are crucial for cellular function. Nonsense mediated decay (NMD) recognizes and degrades premature stop codons containing aberrant mRNA. NMD also has a role in regulating physiological gene expression. UPF1 (regulator of nonsense transcripts 1) is essential for NMD. Its ATPase activity is thought to be required to remodel the messenger ribonucleoprotein particles. In metazoans, N- and C-terminal unstructured regions of UPF1 are phosphorylated by the kinase SMG1, which is considered to be the committed step in the NMD pathway. Phosphorylation of UPF1 is an important signal to recruit the endonuclease SMG6, which performs the first cleavage of the aberrant mRNA. Most of the proteins involved in NMD have been structurally and biochemically characterized. However, atomic information on SMG1 was lacking. The goal of my PhD project was to obtain structural information for SMG1 and its associated complex. I established mammalian cell lines stably expressing SMG1, alone or in combination with its interaction partners SMG8 and SMG9. Then I established a robust purification protocol for SMG1. This enabled me and my lab colleagues to reconstruct the SMG1-SMG8-SMG9 complex at 3.45 Å resolution using cryo-electron microscopy. The structure revealed how SMG1 interacts with SMG8 and SMG9. Comparing the structure to that of the *Caenorhabditis elegans* SMG8-SMG9 complex showed that the SMG8-SMG9 dimer interface is conserved. Structural and biochemical analyses also revealed unexpected ligands and properties. Specifically, we found that inositol hexaphosphate is critical for the kinase activity of SMG1. Analysis of the structures of SMG1-related kinases revealed that SMG1, mTOR (mammalian target of rapamycin), and possibly other kinases bind phosphoinositol, potentially regulating their activity. The work paves the way for understanding how SMG1 might phosphorylate UPF1 and enables further research into the initial steps of NMD.

PUBLICATIONS

Gat Y, Schuller JM, Lingaraju M, Weyher E, Bonneau F, Strauss M *et al* (2019) InsP6 binding to PIKK kinases revealed by the cryo-EM structure of an SMG1-SMG8-SMG9 complex. *Nat Struct Mol Biol* **26**(12): 1089–1093

Li L, Lingaraju M, Basquin C, Basquin J, Conti E (2017) Structure of a SMG8-SMG9 complex identifies a G-domain heterodimer in the NMD effector proteins. *RNA* **23**: 1028–1034

FUNCTIONAL DISSECTION OF A GENE EXPRESSION OSCILLATOR IN *C. ELEGANS*

cf. BIF FUTURA, VOL. 31 | 2.2016

MILOU MEEUSE
Discipline: Molecular Biologist, MSc
Institute: Friedrich Miescher Institute for Biomedical
Research (FMI), Basel, Switzerland
Supervisor: Prof. Helge Grosshans

Temporal control of biological events is crucial for organism development and survival, as exemplified by somitogenesis and circadian rhythms. Oscillations in gene expression can act as timekeeping mechanisms that regulate such repetitive processes. The Grosshans lab has identified thousands of genes whose expression levels oscillate during *Caenorhabditis elegans* larval development. However, whether and how these oscillations control developmental programmes were unclear. In my PhD project, I aimed to characterize the properties of the ensemble of oscillating genes – the oscillator – at the global level and to study the mechanisms underpinning the oscillations and their effects. Using RNA-sequencing data covering the *C. elegans* life cycle from embryo to adult, I found that the expression of ~3,700 genes peaked once per larval stage, from L1 to L4. Although the oscillation periods and larval stage durations varied, the oscillations were coupled to the molts – when the old cuticle is replaced by a new one at the end of each stage. Moreover, I discovered that the oscillations initiated in embryos, arrested in early L1 larvae and starvation-induced arrested larvae, and ceased in adults. The oscillator always arrested in a particular phase that coincided with molt exit, when development can be stalled, suggesting a developmental checkpoint function. To provide insight into the molecular mechanism, I investigated whether and how oscillations arise from rhythmic transcription. RNA polymerase II (RNAPII) chromatin immunoprecipitation (ChIP) sequencing revealed rhythmic occupancy of RNAPII at the promoters of oscillating genes, suggesting transcriptional regulation at the level of RNAPII recruitment. I screened 92 oscillating transcription factors and identified six with molting phenotypes, among them the GRainyHead homologue (GRH-1). My genetic studies showed that GRH-1 is required for timely completion of the molt, for the prevention of cuticle rupturing during the molt, and for oscillatory expression of molting-related genes. Thus, I propose GRH-1 as a putative component of the *C. elegans* oscillator. My work not only provides insight into the oscillator, but also facilitates its further dissection.

PUBLICATIONS

The results of this project can be discussed on BioRxiv 755421.

CATHEPSIN L ENSURES THE SURVIVAL OF THE FITTEST CD4 T-CELL REPERTOIRE

cf. BIF FUTURA, VOL. 31 | 1.2016

ELISABETTA PETROZZIELLO
Discipline: Immunologist, MSc
Institute: Institute for Immunology,
Ludwig Maximilian University,
Martinsried, Germany
Supervisor: Prof. Ludger Klein

The survival of the fittest T-cell repertoire is accomplished through thymic positive selection. During this process, cortical thymic epithelial cells (cTECs) present self-peptides on major histocompatibility complexes (self-pMHC) to developing T cells, in order to test their T-cell receptor (TCR) reactivity. Only the T cells expressing a TCR with low affinity to the self-pMHC survive, so the mature repertoire comprises T cells bearing the proper MHC restriction and a functional TCR. Since cTECs use a unique antigen-processing machinery to generate self-pMHC, it is believed that these self-peptides are optimized to mediate positive selection and are not found elsewhere in the body. However, it is unclear why mildly self-reactive T cells are selected to protect us against pathogens. The aim of my PhD project was to investigate the impact of the cTEC-specific protease cathepsin L (Ctsl) on shaping a functional T-cell repertoire. Since Ctsl generates MHCII-bound peptides, I focused on the selection of CD4 T cells. I began by coupling a Ctsl conditional knockout mouse model, which lacks Ctsl exclusively in cTECs, to a transgenic mouse model bearing a TCR-oligoclonal T-cell repertoire. I then performed flow cytometric characterization and TCR sequencing analyses of the CD4 T-cell compartment in Ctsl-deficient and Ctsl-sufficient animals in which positive selection was carried out on an altered and physiological array of peptides, respectively. Ctsl deficiency during positive selection severely impaired the CD4 T-cell compartment, resulting in fewer T cells and less TCR diversity than in Ctsl-sufficient mice. Furthermore, T cells developing in Ctsl-deficient thymi showed a defective response to polyclonal TCR stimulation, which could be due to decreased functionality of TCRs or to the lack of some specific TCR clones. I also identified some TCRs that are selected regardless of Ctsl expression, but it will be crucial to test if they are still functional. My results bring us a step closer to determining if the peptides produced by Ctsl have a direct role in the generation of a functional T-cell compartment by regulating not only cell numbers and TCR diversity, but also the proper tuning of the TCR signalling cascade.

PUBLICATIONS

The results of this project have not yet been published.

iRHOMS DRIVE EGF CLEARANCE THROUGH THE LYSOSOME AND THE PROTEASOME

cf. BIF FUTURA, VOL. 31 | 2.2016

BORIS SIEBER
Discipline: Biologist, MSc
Institute: Sir William Dunn School of Pathology,
University of Oxford, UK
Supervisor: Prof. Matthew Freeman

Cells communicate with each other by producing signals that trigger responses in receiving cells, such as cell proliferation, differentiation, or migration. Some of the most important signals for proliferation are growth factors, whose dysregulation is a central cause of human cancers. Most growth factors, such as the epidermal growth factor (EGF), are produced as transmembrane proteins by signal-sending cells. In these cells, the first regulatory events occur when pools of signals undergo degradation instead of secretion. Two main degradation machineries are present in the cell: the proteasome and the lysosome. Although inactive homologues of rhomboid proteases (iRhoms) were known to drive EGF degradation, the underlying molecular mechanism remained unclear. In my PhD project, I used a combination of biochemical and microscopy approaches to identify the components required for iRhom-driven degradation in human cells. I first demonstrated that the proteasome and the lysosome cooperate in the iRhom-driven degradation of EGF, thereby establishing this mechanism as a dual degradation route. By focusing on iRhom-driven lysosomal degradation, I investigated the importance of EGF trafficking through the secretory pathway for its clearance. I used two complementary approaches. First, I showed that a variant of EGF that is retained in the endoplasmic reticulum (ER) is still degraded upon iRhom expression. Second, I established that the pool of EGF sent for lysosomal degradation by iRhom shows the characteristic glycosylation signature of an ER-resident protein. These results demonstrate that EGF trafficking is not required for iRhom-driven degradation, thereby suggesting that this process is akin to ER-phagy (autophagy of ER content). This conclusion is further supported by my work showing the involvement of Beclin-1, a core component of autophagy, in iRhom-driven degradation. Although further work is required to establish how EGF is specifically targeted to an ER-phagy pathway, my PhD work highlights the importance of autophagy proteins in the degradation of EGF. This discovery opens up a potential new path towards the development of therapies for cancer treatment.

PUBLICATIONS

The results of this project have not yet been published.

RETROTRANSLOCATION OF MISFOLDED PROTEINS FROM THE ENDOPLASMIC RETICULUM

cf. BIF FUTURA, VOL. 31 | 1.2016

VEDRAN VASIC
Discipline: Molecular Biologist, MSc
Institute: Max Planck Institute for Biophysical Chemistry, Göttingen, Germany
Supervisor: Dr Alexander Stein

Swift disposal of misfolded proteins is essential for cellular viability. Misfolded proteins in the endoplasmic reticulum (ER) are transported across the ER membrane into the cytosol, where they are ubiquitinated, marking them for proteasomal degradation. This process is known as ER-associated protein degradation (ERAD). A major question in ERAD concerns the mechanism of retrotranslocation – that is, how misfolded proteins exit the ER. Evidence implicates the conserved eukaryotic ubiquitin ligase Hrd1 in forming the retrotranslocation channel, yet its mechanism of action was unknown. In my PhD project, I reconstituted purified Hrd1 into model membranes to directly study channel formation. Using single-channel electrophysiology and quantitative biochemistry, I found that Hrd1 forms a pore upon auto-ubiquitination. I showed that Hrd1 interaction with a misfolded protein causes the pore to expand to diameters observed in other protein translocases. When I removed polyubiquitin chains on Hrd1 using a deubiquitinating enzyme, the pore closed and became unresponsive to substrates, demonstrating that pore formation is reversible. Furthermore, I found two binding sites in Hrd1 for misfolded proteins. I showed that when Hrd1 is reconstituted into artificial vesicles, it binds misfolded proteins on its cytosolic side with high affinity upon auto-ubiquitination, which is required for efficient ubiquitination of the substrate. By contrast, when Hrd1 is reconstituted into nanodiscs – synthetic lipid bilayers in which both the luminal and cytosolic sides are accessible – it binds misfolded proteins on its luminal side with a lower affinity. Finally, I used lysine mutants of Hrd1 and identified ubiquitination sites that are crucial for channel activity and substrate ubiquitination. I propose a model in which the affinity gradient from luminal to cytosolic binding sites drives retrotranslocation by trapping the substrate at the cytosolic side and preventing it from sliding back to the luminal side. My results demonstrate that Hrd1 functions as a ubiquitin-gated channel for misfolded proteins and provide important insight into how misfolded proteins exit the ER.

PUBLICATIONS

Vasic V*, Denkert N*, Schmidt CC, Riedel D, Stein A, Meinecke M (2020) Hrd1 forms the retrotranslocation pore regulated by auto-ubiquitination and binding of misfolded proteins. *Nat Cell Biol* **22**: 274–281

DECODING GluN2C/2D-SPECIFIC NMDAR ANTAGONISM USING SMALL-MOLECULE COMPOUNDS

cf. BIF FUTURA, VOL. 31 | 2.2016

JUE XIANG WANG
Discipline: Biochemist, MSci
Institute: Cold Spring Harbor Laboratory, Cold Spring Harbor, NY, USA
Supervisor: Prof. Hiro Furukawa

N-Methyl-D-aspartate receptors (NMDARs) are ion channels that are critical for maintaining proper function of the central nervous system. Their heterotetrameric composition results in subtypes with distinct functional properties and spatio-temporal distribution in the brain, which raises the possibility of treating neurological diseases by pharmacologically targeting specific subtypes. Highly specific and potent compounds that target canonical NMDARs containing GluN2A or GluN2B subunits are well established. By contrast, compounds that target NMDARs containing GluN2C or GluN2D subunits are underdeveloped, with low potency and uncharacterized binding modes. The goal of my PhD project was to understand this substrate specificity in more detail. I showed that the novel small-molecule compound UBP791 is a competitive antagonist that inhibits GluN2C- and GluN2D-containing NMDARs with higher specificity than it does GluN2B- and GluN2A-containing NMDARs, as shown by 16–17-fold and 47–50-fold l ower inhibition potencies, respectively. The inhibition potencies were measured using two-electrode voltage clamp electrophysiology on *Xenopus laevis* oocytes injected with complementary RNA encoding various GluN2 subunits in combination with GluN1. I showed that the critical element for GluN2C- and GluN2D-specific binding was a combination of two residues, a Met and a Lys, at the antagonist binding site. This was achieved through structure determination of the ligand-binding domains of GluN2A and a GluN2D-like subunit in complex with UBP791 using X-ray crystallography and electrophysiology mutagenesis studies. Based on these insights, my collaborators developed the compound UBP1700, which I showed to be the most potent competitive GluN2 antagonist to date, with inhibition potencies for GluN2C- and GluN2D-containing NMDARs in the low nanomolar range. UBP1700 also has higher specificity for GluN2C- and GluN2D-containing NMDARs than UBP791. These novel UBP compounds can be used to elucidate the roles of GluN2C- and GluN2D-containing NMDARs in the brain. My work also proves that the ligand-binding domain can be effectively targeted for subtype-specific control of NMDARs.

PUBLICATIONS

Wang JX, Irvine MW, Burnell ES, Sapkota K, Thatcher RJ, Li M *et al* (2020) Structural basis of subtype-selective competitive antagonism for GluN2C/2D-containing NMDA receptors. *Nat Commun* **11**(1): 423

MD FELLOWS 2019

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 thirteen fellows who were granted an MD fellowship in 2019.

STEPHAN BLÜTHGEN

The role of m5C in coxsackievirus B3 biology

JEFFREY CONDE

Generation of hypoimmunogenic and functional human pluripotent stem cell-derived cardiomyocytes

LINA DÜRRWALD

Deciphering the role of glutamate in cortical interneuron maturation

JONATHAN GOLDSTEIN

Mechanisms of resistance to tumour immunology

FRANK HUANG

Emperipolesis as a new form of cell-in-cell interaction

ANDREAS HUTH

Characterizing corticothalamic ensembles in the posterior medial nucleus (POm) of the thalamus

MARIA KESSLER

Identifying the mechanisms of senescence escape in SETD1A knockdown cells

EMRE KOCAKAVUK

Epigenetic targeting of therapy-resistant subclones in recurrent glioblastoma

LUKAS LANGNER

Profiling on- and off-target activities of CRISPR-guided adenine and cytosine base editors in primary human T cells

CARINA LORENZ

Comprehensive analysis of clonal dynamics in response to neoadjuvant targeted therapy in $HER2^{+}$ breast cancer

LAURITZ MIARKA

Role of S100A9 in radiation resistance of brain metastases

SARAH STEGMANN

Influence of neuronal birth date on cell identity and function in the lateral septum

NINA TESKE

Potential role of brain pericytes in the innate immune response to pneumococcal central nervous system infection

THE ROLE OF M5C IN COXSACKIEVIRUS B3 BIOLOGY

STEPHAN BLÜTHGEN

Duration: 03/19–03/20

Project at: The Rockefeller University, Laboratory of Virology and Infectious Disease, New York, USA

Supervisor: Professor Dr Charles M. Rice

Home University: Heidelberg University Hospital

GENERATION OF HYPOIMMUNOGENIC AND FUNCTIONAL HUMAN PLURIPOTENT STEM CELL-DERIVED CARDIOMYOCYTES

JEFFREY CONDE

Duration: 09/19–08/20

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

Supervisor: Professor Richard T. Lee, MD

Home University: Ulm University Medical Center

DECIPHERING THE ROLE OF GLUTAMATE IN CORTICAL INTERNEURON MATURATION

LINDA DÜRRWALD

Duration: 03/19–03/20

Project at: Tufts University, Department of Neuroscience, Boston, MA, USA

Supervisor: Professor Chris G. Dulla, PhD

Home University: Charité – Universitätsmedizin Berlin

MECHANISMS OF RESISTANCE TO TUMOUR IMMUNOLOGY

JONATHAN GOLDSTEIN

Duration: 09/19–08/20

Project at: The Rockefeller University, Laboratory of Systems Cancer Biology, New York, NY, USA

Supervisor: Professor Sohail Tavazoie, MD, PhD

Home University: Medical Center – University of Freiburg

EMPERIPOLESIS AS A NEW FORM OF CELL-IN-CELL INTERACTION

FRANK HUANG

Duration: 09/19–08/20

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

Supervisor: Professor Peter A. Nigrovic, MD

Home University: Heidelberg University Hospital

CHARACTERIZING CORTICOTHALAMIC ENSEMBLES IN THE POSTERIOR MEDIAL NUCLEUS (POM) OF THE THALAMUS

ANDREAS HUTH

Duration: 01/20–10/20

Project at: Universität Heidelberg, Institut für Anatomie und Zellbiologie, Heidelberg, Germany

Supervisor: Professor Dr Thomas Kuner

Home University: University of Würzburg

IDENTIFYING THE MECHANISMS OF SENESCENCE ESCAPE IN SETD1A KNOCKDOWN CELLS

MARIA KESSLER

Duration: 09/19–07/20

Project at: Harvard Medical School, Massachusetts General Hospital, Cancer Center, Charlestown, MA, USA

Supervisor: Professor Shyamala Maheswaran, PhD

Home University: Heidelberg University Hospital

EPIGENETIC TARGETING OF THERAPY-RESISTANT SUBCLONES IN RECURRENT GLIOBLASTOMA

EMRE KOCAKAVUK

Duration: 01/19–11/19

Project at: The Jackson Laboratory for Genomic Medicine, Computational Biology, Farmington, CT, USA

Supervisor: Professor Roel Verhaak, PhD

Home University: University Hospital Essen

PROFILING ON- AND OFF-TARGET ACTIVITIES OF CRISPR-GUIDED ADENINE AND CYTOSINE BASE EDITORS IN PRIMARY HUMAN T CELLS

LUKAS LANGNER

Duration: 10/19–08/20

Project at: Harvard Medical School, Massachusetts General Hospital, Charlestown, MA; USA

Supervisor: Professor J. Keith Joung, MD, PhD

Home University: Medical Center – University of Freiburg

COMPREHENSIVE ANALYSIS OF CLONAL DYNAMICS IN RESPONSE TO NEOADJUVANT TARGETED THERAPY IN HER2^{+} BREAST CANCER

CARINA LORENZ

Duration: 09/19–08/20

Project at: Stanford University, School of Medicine Stanford, CA, USA

Supervisor: Professor Christina Curtis, PhD

Home University: University of Cologne

ROLE OF S100A9 IN RADIATION RESISTANCE OF BRAIN METASTASES

LAURITZ MIARKA

Duration: 01/20–10/2

Project at: Centro Nacional de Investigaciones Oncologicas (CNIO), Madrid, Spain

Supervisor: Manuel Valiente, PhD

Home University: Kiel University

INFLUENCE OF NEURONAL BIRTH DATE ON CELL IDENTITY AND FUNCTION IN THE LATERAL SEPTUM

SARAH STEGMANN

Duration: 02/19–02/20

Project at: Harvard Medical School, Department of Neurobiology, Boston, MA, USA

Supervisor: Professor Corey Harwell, PhD

Home University: University Hospital Cologne

POTENTIAL ROLE OF BRAIN PERICYTES IN THE INNATE IMMUNE RESPONSE TO PNEUMOCOCCAL CENTRAL NERVOUS SYSTEM INFECTION

NINA TESKE

Duration: 10/19–08/20

Project at: Amsterdam UMC Universitair Medische Centra, Department of Neurology, Amsterdam, The Netherlands

Supervisor: Professor Dr Diederik van de Beek

Home University: Klinikum der Universität München (LMU)

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.

HALFWAY AROUND THE WORLD

The two alumni Clemens Franz and Johannes le Coutre are currently "on the other side of the world" – at least as seen from the BIF office. They talk about work and life in Japan and Australia in this double interview.

By Kirsten Achenbach

AUSTRALIA

Johannes le Coutre studied biology in Regensburg, Germany, and completed his PhD in 1995 at the Max Planck Institute for Nutritional Physiology in Dortmund, Germany. For his postdoctoral training, he moved to the Howard Hughes Medical Institute at UCLA, Los Angeles, USA. In 2000, he returned to Europe to work at the Nestlé Research Centre in Lausanne, Switzerland, where in 2004 he became head of Perception Physiology. In addition, from 2009 to 2017, he held a visiting professorship at the University of Tokyo in Japan. In 2019, he decided to return to academia and accepted a full professorship for food and health at the University of New South Wales (UNSW) School of Chemical Engineering in Sydney, Australia. He still teaches at the EPFL in Lausanne and is a visiting professor at Imperial College London, UK. Finally, he is founding chief editor of *Frontiers in Nutrition*. In Sydney, Johannes is developing a broad research agenda in the emerging field of cellular agriculture.

Johannes, what exactly is cellular agriculture?

Cellular agriculture is the name for a growing field of activities that are deeply rooted in existing technology to produce food. It combines elements of agriculture, life science, medical research, and engineering with the goal of growing edible tissues from vegetables or meat at the cellular level. The aim is to greatly modernize and invigorate our traditional agricultural methods, which often exhaust the land and use enormous amounts of water. Usually, food, nutrition, and health programmes are run in life science environments, so I think our materials science approach can make a valuable contribution. It's cellular agriculture that will enable us one day to eat meat without having to kill an animal. By the way, I'm actively looking for people at all levels who are interested in exploring this uncharted territory to join me in the lab.

After living in several different cultures, do you feel less culture shock?

As you know, in Germany we say "andere Länder, andere Sitten" – "different countries, different customs". Moving to a new country definitely takes you out of your comfort zone. There is not really a culture shock, but you pick up many more things as compared to just going for a visit or a vacation.

Interestingly, you could even make the case that scientific work is the most constant parameter and that the way it is conducted is a unifying activity, simply because it is so similar across the world.

In Australia, we're geographically distant to just about every other country in the world. This is reflected in shipping

Yellow-crested cockatoo

times and costs for laboratory material and equipment. Also, seminars featuring international speakers take place less frequently than in Europe or the United States.

Australia has the image of being very laid back. If that's true, does it apply to the scientific world as well?

This is really just an image and a bad one at that – maybe related to the amount of sunshine and the large number of beautiful beaches here. It clearly isn't true. The fact is, people are hardworking, and normally there is always someone working in the lab, seven days a week.

Compared to both Europe and the USA, I've noticed that Australian universities are very purpose-driven and that the ties between academic work and major governmental initiatives or industry are very visible.

What is quintessentially Australian for you?

The flora and the fauna. Nature is spectacular here and different from anything I've ever seen. Because I'm a biologist at heart, this clearly has been one of the reasons to go to Australia – not only for myself but for the entire family. In addition, I'm a passionate sailor, and for harbour and offshore sailing, you can't beat the conditions here, even though the lakes in Switzerland certainly are beautiful as well. During our first week, we had a visitor in our home, a hand-sized spider, which got us all a bit nervous because we had read about poisonous spiders in the Sydney area. Fortunately, it turned out to be a huntsman spider, which is not really dangerous. We often see cockatoos and ibises. What's truly amazing is that we see hundreds of thousands of flying foxes – bats – every day. They grow to have a wingspan of about one metre. Finally, the beauty of the landscapes and the abundance of exotic plants are simply breathtaking.

You left academia to "add translatability to your scientific endeavors" – why return to academia now?

Sounds counterintuitive, doesn't it? Yet, my decision to work in industry (2000) and my return to academia (2019) are about 20 years apart. A lot has changed, and academic life is becoming way more output-oriented. Every mid-size university these days has a technology transfer office and the goal is to publish patents and expand entrepreneurial activities. Personal publication lists as the single success metric are not enough. While in industry, I was always connected with academic life – and I do enjoy teaching.

Interestingly, the mindset and motivation behind my scientific activities have actually remained quite constant. At UNSW we are now aiming to build world class research capacity in the emerging field of cellular agriculture. While beer brewing and cheese making serve as proofs of principle for the fermentative aspects of this field, it is cultivated meat or vegan cheese that are today's targets. To succeed, we have to conduct and translate excellent fundamental science into visionary engineering technology.

Also, after all these years in the food industry, I'm interested in drawing on the findings and seeing if we can develop an entrepreneurial thrust in this field in Australia. There are multiple start-ups in this field on almost every continent, so maybe one day we'll commercialize our discoveries as well.

You moved to Australia at the start of the worst wildfire season the country has ever seen – how did it impact you personally or scientifically?

Bushfires are normal every year in Australia. However, the 2019/2020 season has been extreme. And yes, both the bushfires and Covid-19 have slowed down my transition to the new environment. As a family we're all in lockdown for the moment on two different continents. New recruitments for the laboratory are possible only within state borders and some budgets are being deferred into next year.

In your career, Australia is continent no. 4 for you. Any plans to get the full set? And if you ever went to Antarctica, what would your dream project be?

Oh, you can be sure I would try to go sailing in Antarctica. But apart from that, I somehow think it best if there are places on earth untouched by humans. So let's cross our fingers that Antarctica will remain the way it is, free of unnecessary interventions. ⟵

Photos: private (left); iStock/phototrip (right)

CLEMENS FRANZ

JAPAN

Clemens Franz is an associate professor of biophysics at the Nano Life Science Institute (NanoLSI) at Kanazawa University, Japan. The facility is a World Premier Institute (WPI) and thus part of a group of elite institutes funded by the Japanese government to perform cutting-edge science, much like Germany's Clusters of Excellence. He focuses on cancer research and uses a novel high-speed microscopy technology to film individual molecules at work in cells. Clemens was born in Berlin, Germany, in 1973. He studied biochemistry in Berlin and London and received his PhD from University College London in 2003. He worked as a postdoctoral fellow at Harvard Medical School in Boston, MA, USA, as well as at the MPI for Cell Biology and Genetics, and the TU Dresden in Dresden, Germany. During this time, he investigated tumour cell adhesion and migration by atomic force microscopy. In 2007, he accepted a group leader position at the DFG Center for Functional Nanostructures at the Karlsruhe Institute for Technology. In 2018, he went to Japan.

You told me that you have to write three to four sentences for kindergarten every morning about how your son, who was born in Japan last year, slept the night before and what he ate. How's your Japanese?

I've been taking regular lessons, but I have to admit I'm still very much at a beginner's level. Simple conversational Japanese is relatively easy, as are grammar and pronunciation. At any rate, people usually understand me. But there are some peculiarities, such as the different counting systems depending on the nature or shape of objects. I quickly learned two of the three Japanese alphabets, hiragana and katakana. Unfortunately, to read proper Japanese texts, you also have to know some 2,000 or more "kanjis" (Chinese characters). It usually takes Japanese children most of their school life to master these and I am afraid it is beyond my abilities. Luckily, app-based translation tools have made it much easier to quickly understand documents or product labels. However, it's fairly easy to get along with English at work when communicating with other researchers and the administrative staff.

What were your reasons for going to Japan?

We have family in Japan and have been there several times, but we felt we had barely scratched the surface. Also, we missed the Japanese friendliness. However, the immediate decision to move was entirely work-related. In my research, I use scanning probe microscopy, or SPM, in which a nanometre-sized tip moves over a sample surface. With it, we can image and even touch and manipulate individual protein molecules, but it takes hours to create a single image. In 2018, the Nano Life Science Institute, called NanoLSI, was founded at Kanazawa University to explore normal and cancer cells using a high-speed version of this technology, which was developed there 15 years ago. It generates many images a second, allowing us to film proteins "at work". Currently, we are constructing a new research building that will bring all NanoLSI researchers together, and we will assemble more than 50 high-speed SPM platforms. This is an unparalleled pooling of resources and a great opportunity to continue my research. So it was an easy decision.

Do you see a difference in the way science is done and perceived?

There are many differences – for instance, the role of students in research. Most undergraduates seem to want to quickly enter a large company, while few go on to a mas-

Photos: private

ter's or a PhD programme. This might be related to the fact that PhD students in Japan normally have to pay their way, through *arubaito,* part-time jobs that take their name from the German word for work. However, our new NanoLSI graduate school now offers decent scholarships for both master's and PhD students. Traditionally, teachers, or *sensei,* are treated with great respect by students, which leads to a less casual style than you have at American or British universities, where everyone's on a first-name basis. Also, group consensus is generally more important than individual interests, and critique is usually offered in a reserved and constructive fashion, rather than in a blunt and direct way. Understanding such subtle nuances sometimes requires reading between the lines.

What are/were the biggest differences in culture for you, especially in the lab?

In my experience, Japanese scientists may sometimes appear more reserved and are, at first, less likely to initiate contact. They are highly focused on their individual projects, which they pursue with exemplary dedication and effort. In general, working days are long and Saturdays are also busy lab days, which puts a strain on the work–life balance and on family life. Some of my colleagues leave for dinner around 8 pm. but then return to finish up their lab day. Compared to the other places I've worked, there are probably fewer regular meetings between individual researchers and groups. However, the WPI programme, which our institute is part of, is strongly encouraging collaborative research projects, so we now have more lunchtime seminars, strategic meetings, and funding for explorative collaborations. Also, university cleaning personnel are not allowed to enter lab spaces, so we clean the labs ourselves. However, I did politely protest when the annual end-of-year cleaning was scheduled for 8.30 am on 25 December.

Have you encountered anything that you would call uniquely Japanese?

One thing that is probably unique to Japan is the personal stamp called a *hanko* that serves as your signature. It's the first thing you need to get upon arrival. You can get one with your name in Japanese within minutes from an ATM-like box in one of the shopping centres. The *hanko* is then used to sign contracts, leases, and any other document in red ink. I need it, for example, to stamp the extensive timesheets I use to keep a record of my work days.

On another note, every morning I pass through a tranquil garden that is part of the museum dedicated to the renowned Zen philosopher Suzuki. It's a welcome quiet moment before entering the busy university campus. The integration of modern architecture and beautiful landscaping continues to amaze me.

How easy was it for you to settle into everyday Japanese life?

The biggest help at the beginning was the fantastic support from the administrative staff at the institute. WPI centres are encouraged to increase the share of foreign staff to about 30 to 40 percent. Several of our staff members are dedicated solely to their support, which far exceeds what one would experience in Europe. As far as our family life goes, we moved to a district in which we may be the only non-Japanese. Despite a few language barriers, our neighbours received us in a very friendly fashion and we are constantly exchanging recipes and gardening herbs. In winter, we all shovel the snow on our street – a nice bonding experience. Our neighbours also helped us to understand the legendary intricacies of Japanese waste separation and recycling procedures. That said, privately we still interact more with other foreigners, sharing new discoveries in Kanazawa and discussing strategies to handle the inevitable red tape. I do try to observe common rules of Japanese curtesy, but naturally I sometimes fail to read situations properly. In my experience, Japanese overlook the occasional faux pas from foreigners.

What is your advice for other fellows wanting to go to Japan?

I can recommend Japan to anyone considering moving abroad. While the overall number of foreigners in Japan is still low (less than 4 percent), Japan universities are becoming increasingly international, making it much easier to adapt than in the past. Needless to say, Japanese cuisine itself is a good reason to move. For scientists, the ability to attract additional funding is paramount. I initially struggled a bit, but recently managed to obtain some welcome external funding. Interestingly enough, even large grants require comparatively short proposals in terms of the number of pages, but this is partly due to the extraordinary information density of written Japanese. When applying in English – which is possible for almost all grants now – we have been given the same amount of space. The forced brevity is hard, but good.

By the way, at NanoLSI, we have a number of programmes for short, medium, and long-term stays at our institute for anyone interested in learning more about high-speed SPM and the Japanese way of life. We also welcome PhD students interested in a transdisciplinary project combining life science and nanotechnology.

PAPERS IN THE SPOTLIGHT

In "Papers in the spotlight", we present papers from current fellows and recent BIF 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, send an email to kirsten.achenbach@bifonds.de.

EVOLVED TO LOVE A PUNGENT SMELL

Drosophila sechellia, endemic to the Seychelles archipelago, feeds exclusively on the toxic noni fruit, also called "vomit fruit" for its tell-tale smell. However, almost all other *Drosophila* species, including the cosmopolitan generalists *D. melanogaster* and *D. simulans*, ignore or even avoid it. BIF alumnus Thomas Auer from Richard Benton's group at the University of Lausanne, Switzerland, developed new tools through CRISPR-Cas9 genome editing to study the genetic and neuronal differences that lead *D. sechellia* to seek out noni fruit.

It turns out that *D. sechellia* does not have a specific olfactory receptor (OR) that detects the odours of the vomit fruit, but mainly relies on a slightly altered version of the receptor OR22a. Without it, the fly does not react to the noni smell. When the authors exchanged OR22a between *D. melanogaster* and *D. sechellia*, the former became more attracted to noni fruit while the latter's interest waned greatly. At the molecular level, the authors found that the OR22a proteins of the two fly species differ in just a handful of amino acids, which tune the *D. sechellia* receptor to the smell of noni fruit, especially its methyl ester components. The authors also found that *D. sechellia* has up to three times more olfactory sensory neurons that express OR22a. In addition, the interneurons connected to these sensory neurons branch differently in the brains of *D. sechellia* than in *D. melanogaster*. Both of these changes might alter how the fly processes smells. With their new methods, Auer and colleagues have thus identified key changes at the molecular, physiological, and anatomical level that offer an initial explanation of *D. sechellia*'s love of this pungent fruit. Moreover, this work lays an important foundation to use the highly specialized fly as a model to study how genetic and neuronal changes allow other types of behaviours to evolve.

D. sechellia on its exclusive food staple, the noni fruit.

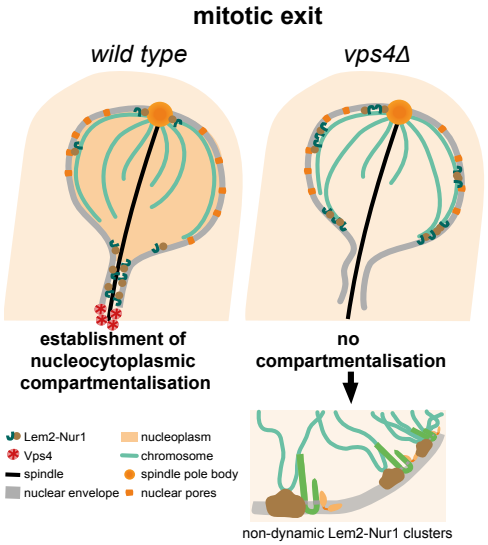
REFERENCE

Auer TO, Khallaf MA, Silbering AF, Zappia G, Ellis K, Álvarez-Ocaña R *et al* (2020) Olfactory receptor and circuit evolution promote host specialization. *Nature* **579**: 402–408.

Thomas Auer, fellow: 2011–2013

LETTING GO FOR CLOSURE DURING MITOSIS

When cells divide, they need to deal with the membrane around their nucleus, which keeps nucleoplasm and cytoplasm apart. While animals and plants have an open mitosis, in which they completely dismantle their nuclear envelope (NE), many lower organisms keep it intact (closed mitosis) or only open it in distinct places (semi-open mitosis). However they do it, they all use the same highly conserved molecular players. Gerard Pieper from Snezhana Oliferenko's group at the Francis Crick Institute in London, UK, studied the semi-open mitosis of the fission yeast *S. japonicus* and the role of the ESCRT-III complex in it. During interphase, Lem2-Nur1 anchors heterochromatin (HC) to the inner nuclear membrane, a process important for genome organization. In this phase, the authors found, Lem2-Nur1 plays a continu-



Photos: T. Auer (private), Benjamin Fabian (bottom left), Gerard Pieper (bottom right)

ous game of catch and release with HC: Lem2-Nur1 binds to HC and thus attracts the different parts of the ESCRT-III complex, starting with Cmp7, which in turn recruits particularly Vps32 and Vps4. Vps4 promptly catalyses the release of all of the involved players, including HC. Loose again, Lem2-Nur1 moves along the NE to start the game anew. Towards mitotic exit, however, Lem2-Nur1 moves along the NE to where the spindle sticks through the ruptured NE (see figure). The authors were able to show the molecular events required for Lem2-Nur1 to closely wrap the loose ends of the NE, its tails, to the spindle to effectively close the nuclear membrane. However, Lem2-Nur1 can only do this when it is released from HC by Vsp4. If this does not happen, the nuclei fail to compartmentalize, Lem2-Nur1 and HC form immobile clusters during interphase, and the chromosomes stay bound to the NE throughout mitosis. The latter never occurs in healthy cells, no matter what form of mitosis they employ. As a consequence, the authors saw severe growth defects in their vps4-deficient mutants of *S. japonicus*. The results should help us to understand the fundamental process of mitosis better and hint at further functions of the ESCRT complex.

REFERENCE

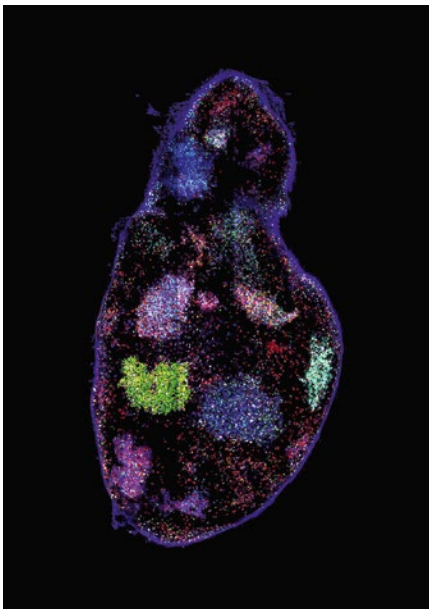
Pieper GH, Sprenger S, Teis D, Oliferenko S (2020) ESCRT-III/Vps4 controls heterochromatin-nuclear envelope attachments. *Developmental Cell* **53**: 1–15

Gerard Pieper, fellow: 2016–2018

Photos: G. Pieper (private); Ariën Schiepers (private); the Rockefeller University (top right)

FURTHER EDUCATION FOR B CELLS FOR BETTER VACCINES

To develop a universal vaccine against pathogens such as the flu, which can infect people multiple times, we must better understand how our immune system reacts to repeated encounters with different strains of the same pathogen. When encountering a pathogen, some B cells – the cells that produce antibodies and can remember which ones helped to defeat past infections – enter germinal centres in the lymph nodes. In these "boot camps", they undergo rapid mutations to find the best-fitting antibodies, which they then release to fight the invader. However, these antibodies might no longer fit if they later encounter a different strain or evolved version of the same pathogen.



Germinal centres with clusters of activated B cells (fluorescently marked) in a mouse lymph node.

Ariën Schiepers and his supervisor, Gabriel Victoria, of the Rockefeller University in New York, USA, have now discovered that B cells only rarely return to a germinal centre during a second encounter with the same pathogen. When the two researchers compared both encounters, it turned out that, in both cases, the germinal centres contained hundreds of different B cell clones. However, of the B cells in the secondary germinal centres, not even 10 percent had been in a germinal centre before. Thus, the immune system does not efficiently improve on the experience it had when first encountering a pathogen. If it had this ability, the B cells would be able to hone their antibodies with every encounter and finally may even develop so-called broadly neutralizing antibodies. These can recognize a certain pathogen in all of its forms, leading to a broad immunity against most, if not all, forms of this pathogen, even future ones. Therefore, in order to develop a universal vaccine against the flu or HIV, it might be a good strategy to find ways to recall more experienced B cells to the germinal centres for further education.

REFERENCE

Mesin L*, Schiepers A*, Ersching J, Barbulescu A, Cavazzoni CB, Angelini A *et al* (2020) Restricted clonality and limited germinal center reentry characterize memory B cell reactivation by boosting. *Cell* **180**: 92–106

Ariën Schiepers, fellow: 2018–2020

PERSPECTIVES

FROM PHD TO CONSULTING TO VENTURE CAPITAL

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

INTERVIEW WITH YANXIANG ZHOU

Yanxiang Zhou was born in Beijing, China, in 1987, but grew up in Germany. At the age of 17 he won the prize for the best interdisciplinary project in "Jugend forscht", a nationwide science, engineering, technology, and math competition for researchers younger than 21. For his project, he developed an algorithm for sequence alignments in homology modelling. He went on to earn a BSc in bioinformatics at TU Munich, a BSc in chemistry and biochemistry at LMU Munich, an MRes in cancer biology at Imperial College London, and a PhD in molecular biology at the London Research Institute (now part of the Francis Crick Institute). In 2015, after completing his PhD, he joined the Boston Consulting Group (BCG), where he worked with different healthcare companies on projects ranging from technology assessment and sales to marketing strategy. In 2018, he became an associate at Illumina Ventures, a venture capital (VC) firm with offices in Dublin, Ireland, and the San Francisco Bay Area. It invests in early-stage companies that are pioneering new genomics applications and enabling precision medicine.

What does your job at Illumina Ventures entail?

When you first join a VC firm, you typically "source" companies and technologies that could be used in new start-ups. They should offer solutions that are at least 10 times better than what is currently on the market and have the potential for the kind of growth that looks like a hockey stick on a chart. VC is not about incremental changes. You search for companies through university tech transfer offices, at networking events, at scientific conferences, and sometimes simply by using Google. The next important step is "due diligence". This involves analysing the company in greater detail, their idea and the data supporting it, their market, intellectual property, team, etc.

You also help your portfolio companies – those you've already invested in – to become more successful. What they need can be very different and also depends on what stage they're in. Currently, for example, we're helping one company with sourcing nose swabs for SARS-CoV-2 tests through our network.

How will your job change as you advance?

The higher up in the firm, the more involved you are in negotiating deals and working with portfolio companies directly. A big part of your life entails advising companies as a member of their board of directors.

What do you like best about your job?

Meeting entrepreneurs who are full of energy and optimism. Sometimes you also meet companies that develop solutions to problems close to your heart. I recently talked to a company that is developing more sensitive food allergy tests. For people with severe food allergies, like my six-year-old daughter, they could save weeks of very unpleasant conventional testing.

What would you recommend to other fellows wanting to take this path?

VC is not for everyone. Knowing yourself and what type of work you want to do is key. Internships are possible to get first-hand experience. In VC, you have to talk to a lot of people: entrepreneurs, scientists, medical doctors, lawyers, your investors, etc. For an introvert, this can be very draining. In addition, in stark contrast to many other jobs, there is a smaller team around you. You are much more on your own.

However, one aspect that is pleasant is that in VC you're there to provide funding to other people. It's not a service industry like consulting in which the client ultimately decides. On a practical level, this means fewer ad-hoc "emergencies" and better control of your schedule. However, when closing a deal – that is, just before making an investment – the hours can be longer, and there will never be "enough" time to figure out all details. The key is to focus on items that matter in the big picture.

All in all, you're involved with people finding solutions to current problems. That's exciting to see!

Photo: Illumina Ventures

PROFILES

PROFESSOR DIRK BAUMJOHANN
Institute: University Hospital Bonn (UKB), Germany
Fellowship: 2006–2008

In February, Dirk Baumjohann took up a professorship in autoimmunity at the Department of Internal Medicine III, Oncology, Hematology, and Rheumatology, at University Hospital Bonn, Germany. He will build upon his existing scientific knowledge of molecular and cellular aspects of T cell diversity and their role in autoimmune disease such as multiple sclerosis. With his work, he will support the immunoSensation2 Cluster of Excellence and use the close link between bench and bedside in Bonn for the benefit of patients.

PROFESSOR STEFANIE DIMMELER
Institute: University of Frankfurt, Germany
Fellowship: 1991–1992

Stefanie Dimmeler has been elected the new spokesperson of the Board of Directors of the German Center for Cardiovascular Research (DZHK) and will take up her position in December 2020. During her three-year term, she plans to focus on intensifying the cooperation between DZHK sites and between the DZHK and its national and international partners. At seven sites throughout Germany, DZHK researchers are developing new therapies and diagnostic procedures for cardiovascular diseases. The DZHK aims to rapidly and efficiently transfer the results from basic research into clinical practice.

PROFESSOR VOLKER HAUCKE
Institute: Director at the Leibniz Institute for Molecular Pharmacology (FMP), Berlin, Germany
Fellowship: 1994–1997

PROFESSOR RÜDIGER KLEIN
Institute: Director at the Max Planck Institute of Neurobiology, Martinsried, Germany
Fellowship: 1988–1990

PROFESSOR EDWARD LEMKE
Institute: Institute of Molecular Biology (IMB), Mainz, Germany
Fellowship: 2003–2005

Three BIF alumni are among the 185 researchers to receive an **ERC Advanced Grant,** worth up to 3.5 million euros, in the 12th round of the funding programme. As Mariya Gabriel, European Commissioner for Innovation, Research, Culture, Education, and Youth, explains, "By supporting frontier research, the EU enables our brightest scientists to push the frontiers of knowledge for the long-term benefit of all."

With his project "SynapsBuild: Mechanisms of Presynaptic Biogenesis and Dynamic Remodeling", **Volker Haucke** will analyse human nerve cells to understand how presynaptic vesicles and their precursors are formed and mature, how they are transported and assembled, and, finally, how these processes are coordinated and regulated. In addition to the ERC Advanced Grant, Volker has been awarded the 2020 Feldberg Prize by the Feldberg Foundation. This organization aims to promote Anglo-German scientific exchange in the sphere of experimental medical research and awards the prize on an annual basis to one outstanding researcher in Germany and one in the UK.

With his project "BrainRedesign: Redesigning Brain Circuits in Development", **Rüdiger Klein** will attempt to unravel how the amygdala helps us to learn by attaching positive or negative emotions to events and objects. He will use so-called guidance molecules to change how the neurons in the amygdala of mice connect during development. This leads to reorganized circuits, thereby transforming the innate and learned emotional behaviour of the mice.

In his project "MultiOrganelle Design: Multiple Designer Organelles for Expanded Eukaryotic life", **Edward Lemke** will use designer organelles in cells engineered to effectively have two genetic codes to study so-called intrinsically disordered proteins (IDPs). These abundant proteins are very difficult to study due to their flexible and dynamic structure. The two genetic codes will allow the researchers to incorporate fluorescent groups at specific locations in proteins. They will thus be able to label proteins at multiple specific sites to visualize and study conformational changes of IDPs at unprecedented resolution without altering the cell's host physiology.

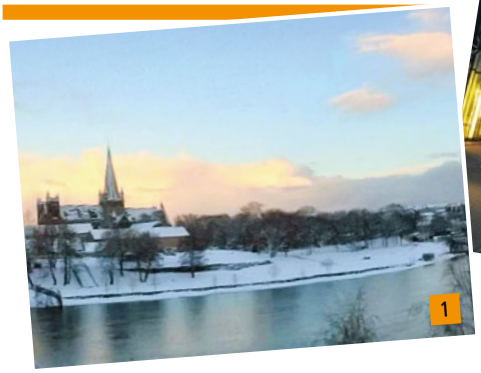
STEPHANIE PANIER
Institute: Max Planck Institute for Biology of Ageing, Cologne, Germany
Fellowship: 2008–2010

Stephanie Panier has started her own group, "Genome Instability and Ageing", at the Max Planck Institute for Biology of Ageing and the CECAD Cluster of Excellence in Cologne. She will study how cells detect and repair DNA damage to safeguard their genetic information. In particular, she will look at the regulatory mechanisms underlying DNA recombination and RNA metabolism at global sites of DNA damage, at telomeres, and at distressed replication forks.

Photos: Baumjohann: private; Dimmeler: University of Frankfurt; Haucke: Silke Oßwald, FMP; Klein: MPI of Neurobiology; Lemke: IMB, c_T. Hartmann; Panier: V. Borel

A BIF FELLOW'S GUIDE TO ...

TRONDHEIM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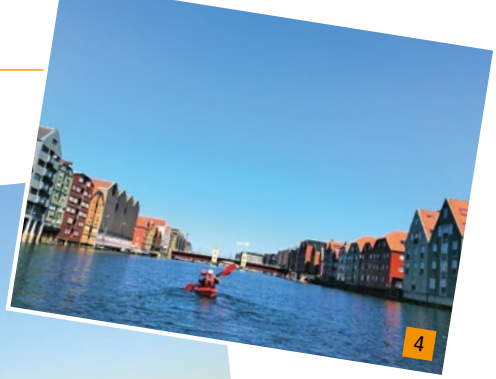
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Travelling is fun – especially if you get insider tips from locals! In each edition of FUTURA, one fellow shows you around his or her city. In this edition, your guide is Christa Ringers. She reports from Trondheim, Norway, the city best known as the place where Norwegian kings are crowned.

FACTS & FIGURES

Country: Norway
Population: 199,039
Area: 321.81 km²
Students: About 36,000
Known for the St. Olav's Way pilgrimage, student life, and the world's first and only bicycle lift.
Website: trondheim.com, visittrondheim.no

BEST SIGHTS

Nidaros Cathedral 1 3**:** the church built on St. Olav's grave is the largest medieval building in Scandinavia.
Gråkallbanen: ride the northernmost tramline straight to the city's Bymarka Forest.
Munkholmen: plan a daytrip to the island in the fjord, once home to a monastery, a prison, and a war bunker, and enjoy the beach.

NIGHTLIFE

Den Gode Nabo: an old wharf turned cosy bar with a large assortment of beer.
Work-Work: have a blast with beer and board games.

RESTAURANTS

Kommandanten 2 (next to the city fortress): order reindeer meat with hand-picked mushrooms from the forest (seasonal).
Taquiros and Tequila: one of the best Mexican restaurants in town with great vegetarian and vegan options.
Bror: affordable and tasty homemade burgers.

WHERE TO STAY

Singsaker Sommerhotell: cosy and affordable hotel run entirely by students.
Scandic Nidelven: worth visiting for their breakfast, which has been selected as Norway's best several years in a row.
Camping: take advantage of the freedom-to-roam law and go camping. Just clean up after yourself and keep a reasonable distance to people's houses.

ACTIVITIES

Winter: go skiing in Våsfjellet, or drink *gløgg* at the Christmas market.
Spring 4**:** celebrate 17 May (National Day) or screw up your courage and swim in the fjord near Korsvika.
Summer: hike all seven hills of Bymarka in one day (30 km) and get a TOPP7 T-shirt. *Skål* (toast) like a *trønder* during the Bryggerifestivalen (brewer's festival).
Fall: see the northern lights (norway-lights.com). Enjoy the cakes at Mormors Tue ("grandma's living room").

Contributors wanted! If you would like to introduce your city, send an email to kirsten.achenbach@bifonds.de

Christa Ringers is 26 years old. She is studying at the Kavli Institute for Systems Neuroscience, Trondheim, Norway. Her supervisors are Dr Nathalie Jurisch-Yaksi and Professor Dr Emre Yaksi.

Photos: Christa Ringers

PROFILES

PROFESSOR TOBIAS ROSE
Institute: University Bonn, Germany
Fellowship: 2003–2005

Tobias Rose has been appointed assistant professor at the University of Bonn. On 1 May, he launched the group "Circuit Mechanisms of Behaviour" to study how stable memories can arise despite the fact that neural circuits constantly rearrange themselves. He and his team will use naturalistic learning tasks and unrestrained behaviour in mice to investigate learning-related changes in the mouse visual system on multiple scales, ranging from the synaptic to the circuit level. To do so, they aim to establish novel, densely quantified behavioural paradigms and develop innovative miniaturized microscopy technology.

PROFESSOR MARIA HONDELE
Institute: ETH Zurich, Switzerland
Fellowship: 2008–2011

Maria Hondele will join the Biozentrum of the University of Basel as a new assistant professor on 1 September. Her research focuses on the fate of messenger RNAs (mRNAs), copies of the cell's genetic material that serve as matrices for protein production. These mRNA molecules are transported, altered, stored, repressed, or degraded by certain proteins. Maria's main topic of interest are membraneless organelles formed by liquid-liquid phase separation, for example, P-bodies or nuclear speckles, which are thought to control the fate of mRNA molecules.

PROFESSOR PETER KOHL
Institute: University Heart Centre Freiburg – Bad Krozingen, Freiburg, Germany
Fellowship: 1992–1995

Peter Kohl is the coordinator of Collaborative Research Centre 1425 "The Heterocellular Nature of Cardiac Lesions: Identities, Interactions, Implications", which started on 1 July and will be funded by the DFG with 11 million euros for four years. Only one third of the cells in the heart are muscle cells, but our knowledge of non-myocytes, their interactions, and their utility for steering tissue repair is still in its infancy. CRC 1425 aims to develop new methods for the diagnosis and therapy of heart disease. In doing so, researchers are not primarily targeting scar prevention or retransformation into functional muscle tissue; rather, they are pursuing a new and complementary approach: working with nature's own repair processes to allow scars to fulfil their important mechanical repair function with minimal side effects. CRC 1425 brings together 26 scientists from the University Heart Centre Freiburg – Bad Krozingen, the University Hospital Freiburg, the medical, biological, and technical departments of the University of Freiburg, the Max Planck Institute of Immunobiology and Epigenetics in Freiburg, as well as the universities of Heidelberg, Bonn, and Frankfurt.

TOBIAS RUFF
Institute: ETH Zurich, Switzerland
Fellowship: 2015–2017

Tobias Ruff has been awarded a Human Science Frontier Program (HSFP) Long-Term Fellowship for his project "Investigating Adult dLGN Reinnervation Using a Biohybrid Retinal Ganglion Cell Multielectrode Array", for which he moved to the ETH in Zurich, Switzerland. The Human Science Frontier Program awards these grants for innovative, ground-breaking projects. Applicants are expected to take a new research approach. For this, they receive funds to obtain training in a new area of research in an outstanding laboratory of their choice in another country.

UPCOMING EVENTS:

To protect our fellows and do our share in preventing the further spread of SARS-CoV-2, we have cancelled or postponed several of our meetings. As the situation is not yet resolved, further meetings might be affected. For this reason, we kindly ask you to check our website (bifonds.de/news-network/seminars-events) for current information on our seminars, communication training, and the ITCs.

13–14 NOVEMBER 2020
Meeting of BIF's Board of Trustees
The trustees decide on the allocation of fellowships, review the proposals for the International Titisee Conferences, and settle all of the foundation's matters of fundamental importance.

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

Photos: Rose: University Bonn; Hondele: private; Kohl: Universitätsklinikum Freiburg/B. Schilling; Ruff: ETH Zürich

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