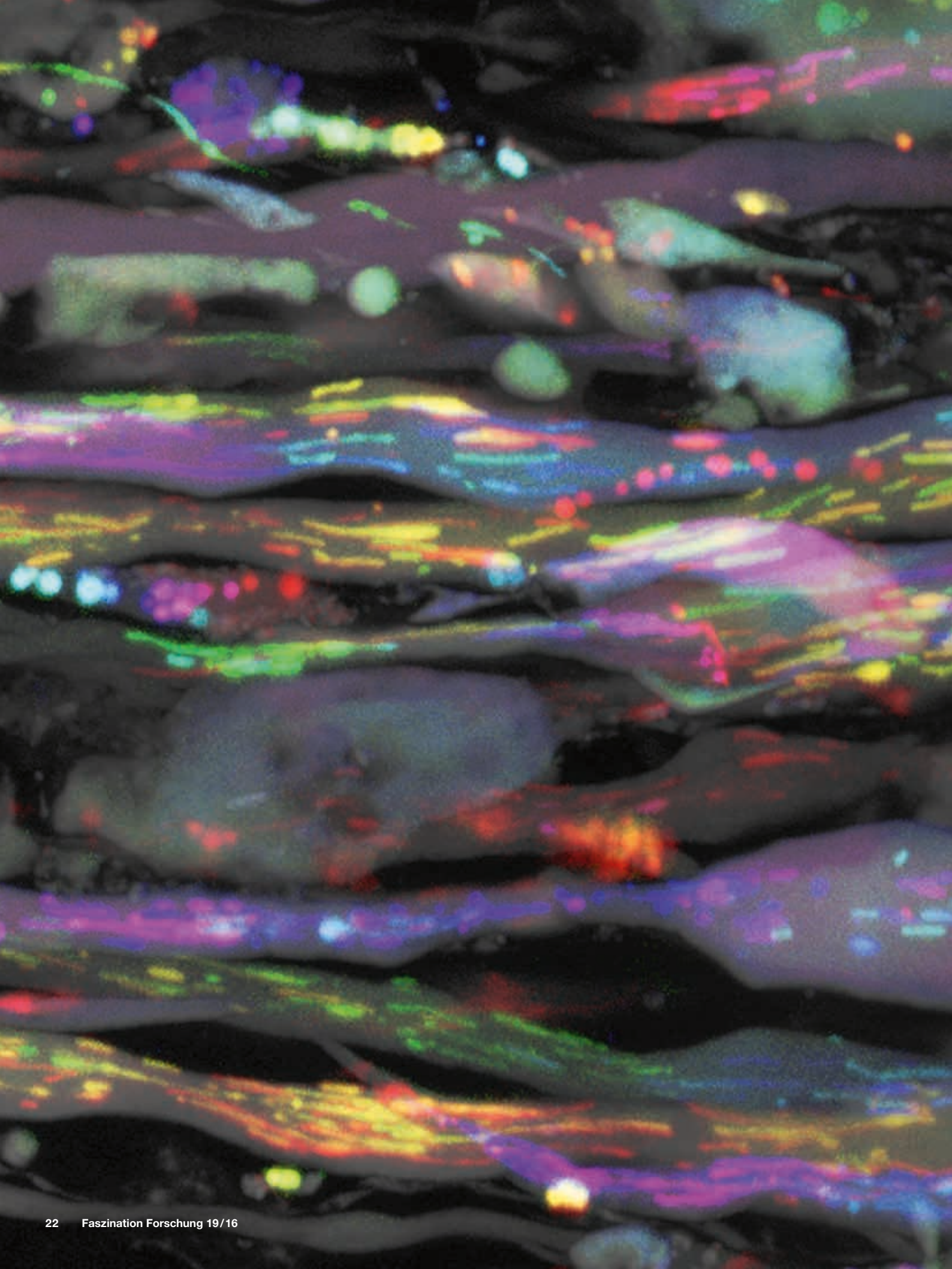
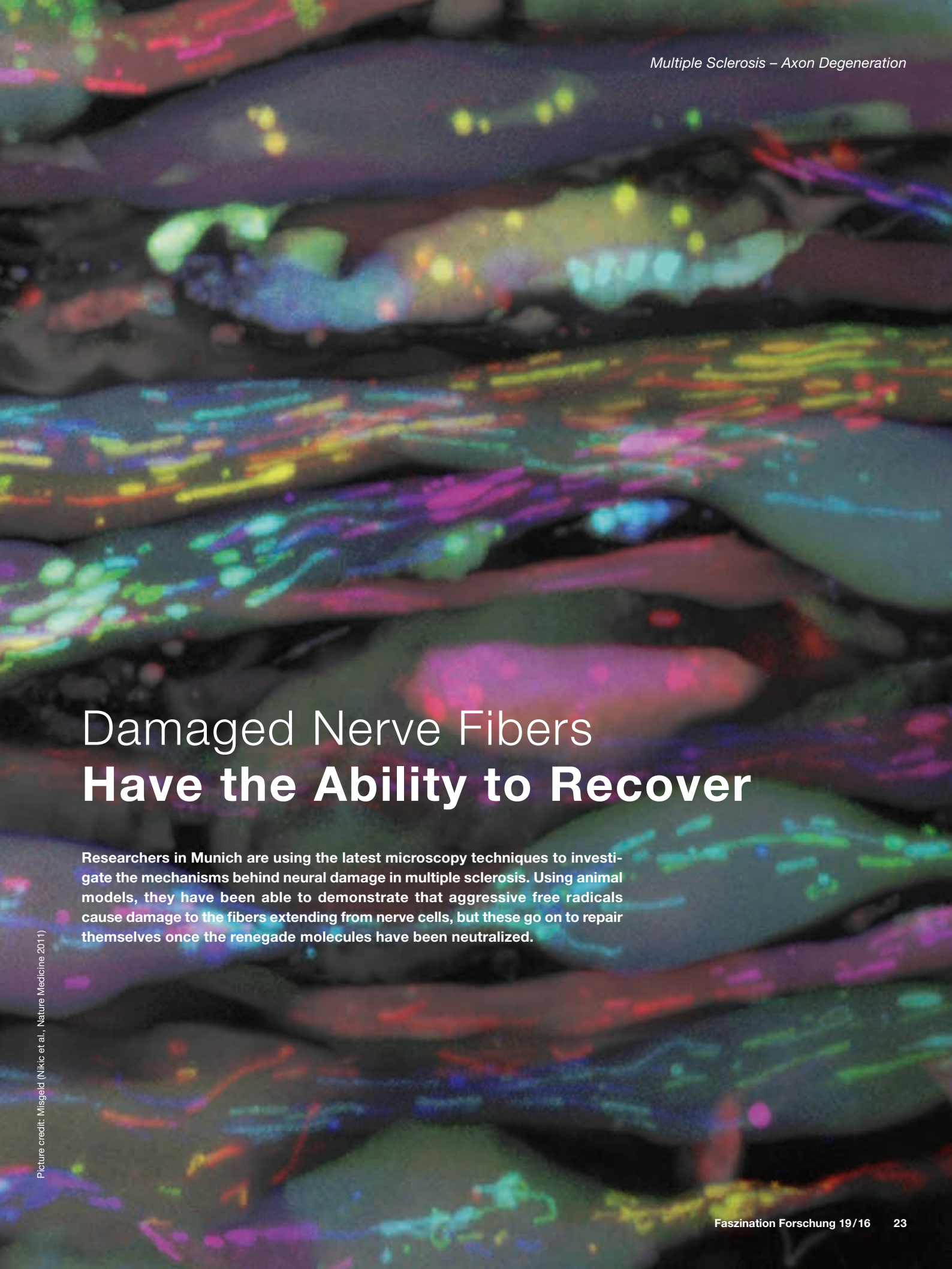
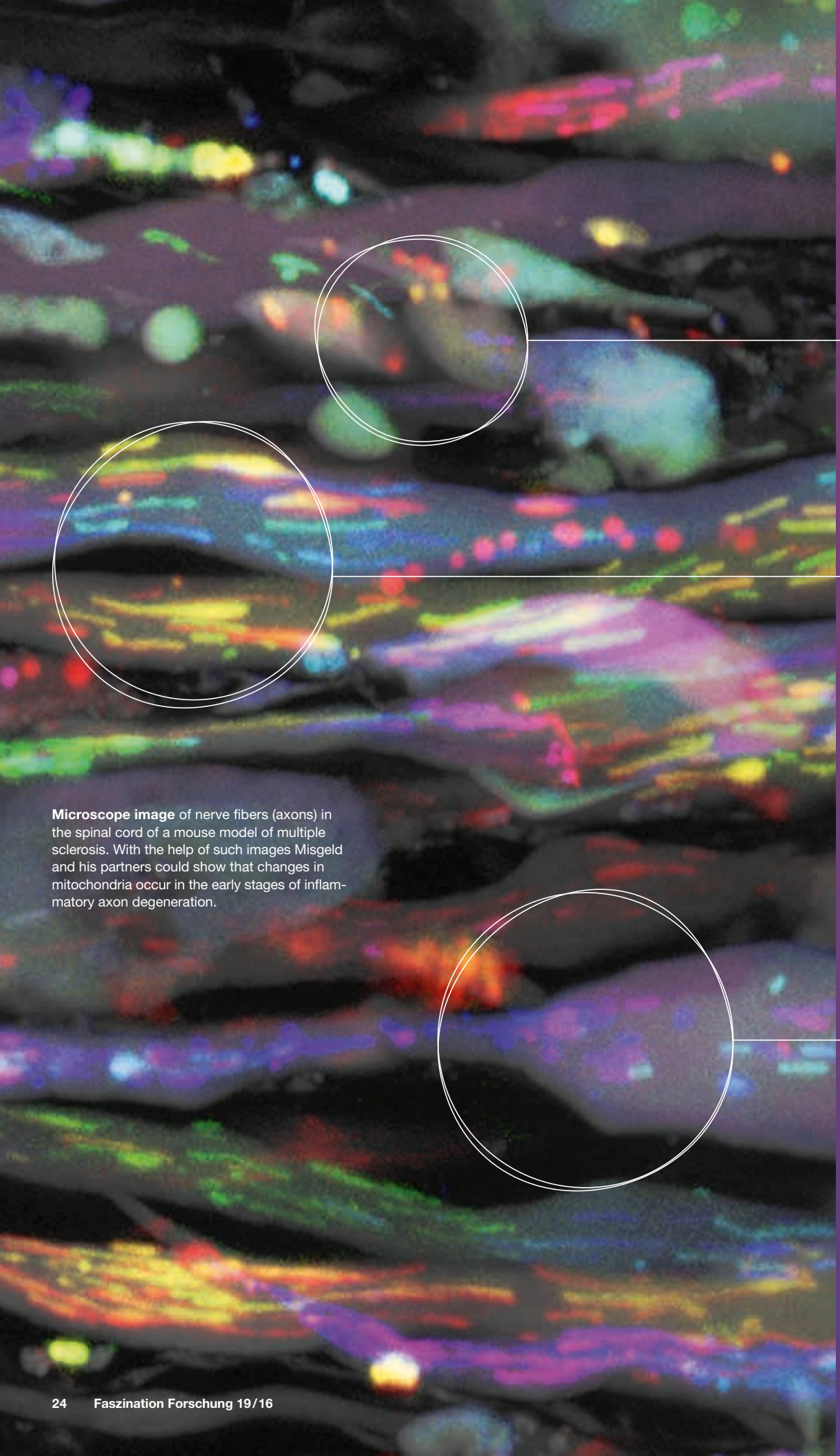




Damaged Nerve Fibers
Have the Ability to Recover

Researchers in Munich are using the latest microscopy techniques to investigate the mechanisms behind neural damage in multiple sclerosis. Using animal models, they have been able to demonstrate that aggressive free radicals cause damage to the fibers extending from nerve cells, but these go on to repair themselves once the renegade molecules have been neutralized.

Picture credit: Misgeld (Nikic et al., Nature Medicine 2011)



Microscope image of nerve fibers (axons) in the spinal cord of a mouse model of multiple sclerosis. With the help of such images Misgeld and his partners could show that changes in mitochondria occur in the early stages of inflammatory axon degeneration.

Degenerated axon

Mitochondria (colored dots) are damaged and round shaped

Intact axon

Mitochondria (colored lines) are intact and elongated

Swollen axon

Mitochondria swell and begin deforming into round shapes

Picture credit: Misgeld (Nikic et al., Nature Medicine 2011); Graphics: edlundsepp

Link
www.neuroscience.med.tum.de

Evdoxia Tsakiridou

Geschädigte Nervenzellen erholen sich

Der Krankheitsmechanismus der Multiplen Sklerose (MS) ist komplex und immer noch nicht restlos aufgeklärt. Bekannt ist, dass die eigenen Immunzellen das zentrale Nervensystem angreifen. Es bilden sich lokale Entzündungsherde im Gehirn und Rückenmark und die Signalverarbeitung in den Nervenzellen (Neurone) ist gestört. Es folgen Schäden an den Schutzschichten (Myelinscheide) von Neuronen und ihren Fortsätzen (Axone). Im weiteren Krankheitsverlauf degenerieren die Axone mit der Folge, dass die Nervenzellen ihre Funktion einbüßen.

Bislang glaubten Forscher, dass der Abbau der Myelinschichten zum Absterben der Nervenzellen führt. Thomas Misgeld, Lehrstuhlleiter für Zellbiologie des Nervensystems an der TUM, und sein LMU-Kollege Martin Kerschensteiner fanden jedoch bei Studien am Maus-Modell heraus, dass auch Axone mit intakt erscheinender Myelinscheide untergehen. Diese sogenannte fokale axonale degeneration (FAD) läuft in mehreren Phasen ab: Zuerst schwellen an bestimmten Stellen die Nervenzellen an, später zerfallen sie in Einzelteile. Das geschieht nicht sofort. Zahlreiche Axone verharren für einige Zeit in geschwollenem Zustand, bevor der Zerfallsprozess einsetzt – und manche erholen sich spontan. Die beiden Forscher gehen deshalb davon aus, dass der Abbau der Myelinscheide nicht die einzige Ursache für die Axondegeneration sein kann. Zwischenstufen des FAD-Prozesses fanden die Wissenschaftler auch in Proben, die aus dem Gehirn von MS-Patienten stammen.

Charakteristisch für die FAD ist auch, dass die Mitochondrien, die Energielieferanten der Neuronen, deformiert sind. Diese Schäden werden vermutlich von Immunzellen verursacht, die Sauerstoff- und Stickstoffradikale produzieren. Mithilfe molekularer Bildgebung konnten Misgeld und Kerschensteiner in pharmakologischen Experimenten mit Mäusen zeigen, dass solche Molekülradikale die Mitochondrien schädigen und FAD auslösen können. Neutralisierten sie die aggressiven Moleküle mit entsprechenden Substanzen, konnten sich die betroffenen Axone wieder erholen. Die Forscher gehen deshalb davon aus, dass die FAD auch bei der MS umkehrbar sein könnte.

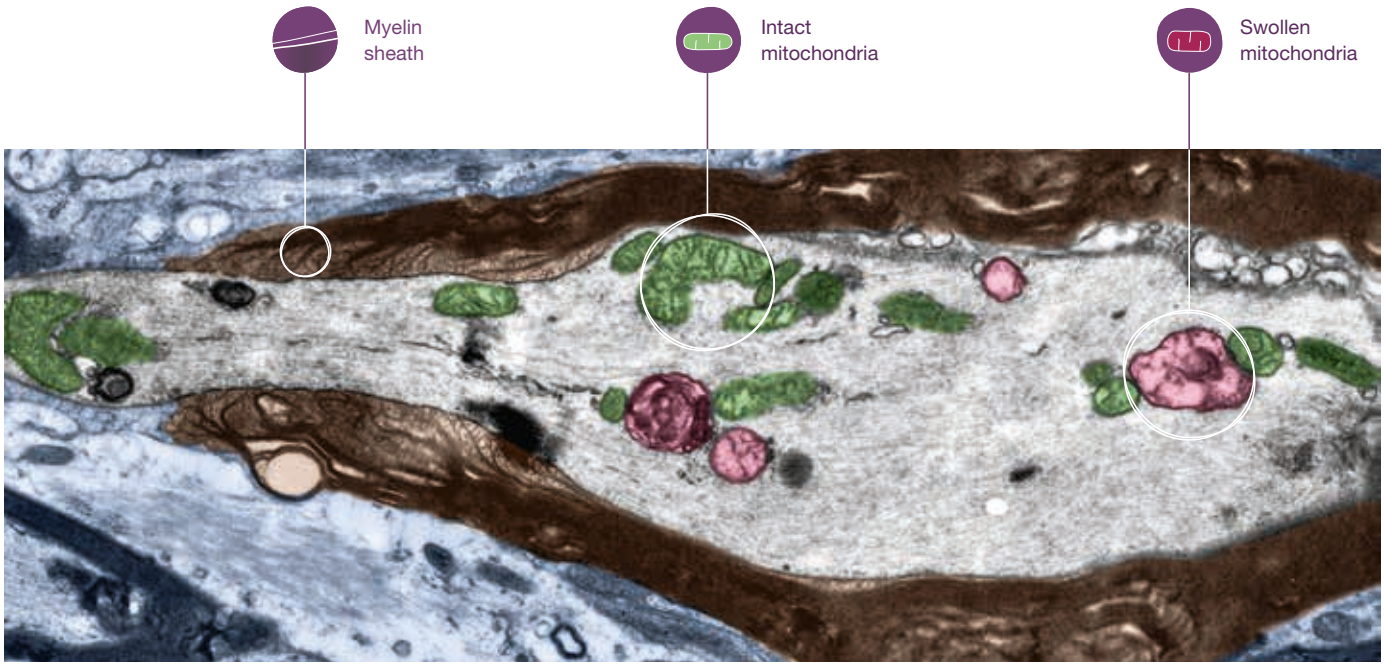
Nun lautet die Frage: Löst die Schädigung der Mitochondrien die FAD-Kaskade aus? Es gilt auch, die intrazellulären Signalwege aufzuklären, die die Radikale aktivieren. Die Wissenschaftler hoffen auf der Grundlage eines besseren Verständnisses, eines Tages mit entsprechenden Medikamenten auf diesen molekularen Prozess einwirken zu können. □

We still don't know enough about the molecular processes involved in multiple sclerosis, and about axonal degeneration mechanisms in this disease in particular," acknowledges Prof. Thomas Misgeld, director of TUM's Institute of Neuronal Cell Biology. Although researchers around the world have been focusing intensively on multiple sclerosis (MS) for many decades now, they still have not fully unraveled the complexities of this disease. Through painstaking research, they continue to piece the MS puzzle together.

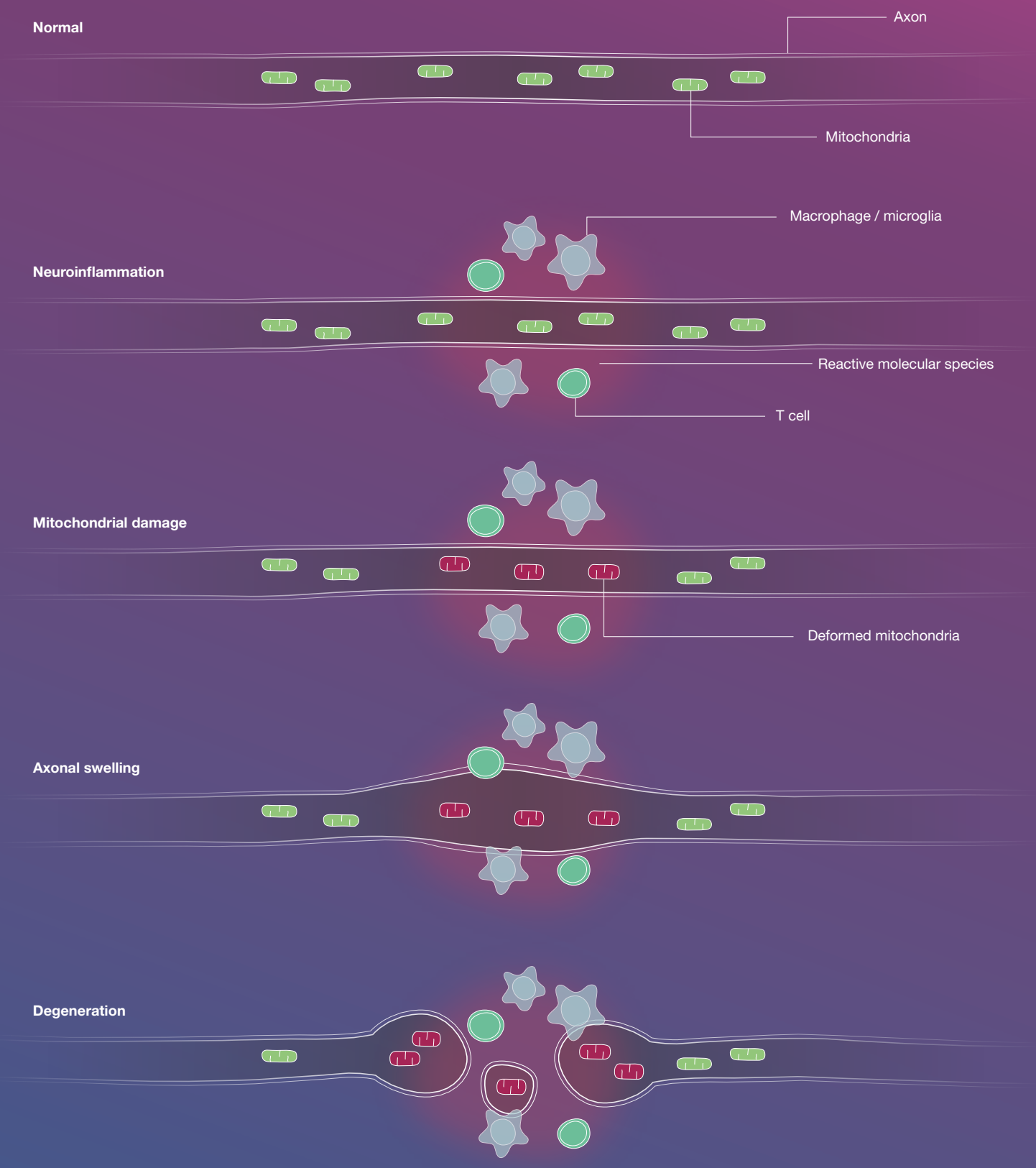
This autoimmune disease is characterized by local inflammation sites in the brain and spinal cord at its onset, as well as by episodic disruption of signal (action potential) conduction in the nerve cells, or neurons. The resulting symptoms include visual impairment, numbness or paralysis. A particularly striking aspect is damage to the myelin sheath – the fatty protective layers that surround and electrically insulate the long projection fibers of neurons, known as axons. Numerous axons also degenerate over the course of the disease – a process that can lead to irreversible loss of neuronal function. "There is a clear correlation between permanent neurological problems in MS patients and the extent of axonal damage," stresses Misgeld.

Until recently, many in the research community believed that destruction of the myelin sheath was the cause of subsequent axonal degeneration and neuronal atrophy. Now, however, there is a new take on this. Using a mouse model, research teams working with Misgeld and his colleague Martin Kerschensteiner (LMU Munich) were able to demonstrate that axons with intact protective coating also die off. The two researchers thus now consider it unlikely that myelin sheath destruction could be the sole cause of axonal degeneration.

As yet poorly understood, this mechanism – known as focal axonal degeneration (FAD) – involves several steps. First, the axons swell up at certain points, before later disintegrating into individual pieces. To picture this, imagine pearls forming on a string, detaching themselves and then dropping off. However, the breaking-away stage does not happen immediately. Many axons hang on in a swollen condition for a few days before the disintegration process takes hold – and some spontaneously repair themselves. "Interestingly, such interim stages of the FAD process are also found in brain biopsies from MS patients," comments Misgeld. ▷



Electron micrograph of an axon located in the spinal cord of a mouse model with multiple sclerosis. The axon is the first stage of degeneration which is characterized by swelling mitochondria. It contains intact looking mitochondria (green) and swollen ones (red). The myelin around the axon (colored brown) is intact.



Picture credit: Nikic et al., Nature Medicine 2011; Graphics: ediundsepp (source: TUM)

Focal axonal degeneration is a process which can lead to axon degeneration in inflammatory lesions in the spinal cord. A normal nerve fiber – characterized by elongated mitochondria – swells and fragments, often despite an intact myelin sheath. Even before a nerve fiber swells, its mitochondria show local damage. It is notable that in the early stages of this process some axons recover spontaneously.

“There is a clear correlation between permanent neurological problems in MS patients and the extent of axonal damage.”

Thomas Misgeld

Prof. Thomas Misgeld

Homing in on the brain

Following medical training in Munich, at TUM and the Max Planck Institute for Neurobiology, Thomas Misgeld did postdoctoral research in the United States. At Washington University in St. Louis and at Harvard, he acquired expertise in cutting-edge microscopy techniques for in vivo imaging that continue to shed new light on the life of individual nerve cells.
Returning to TUM in 2006 as a Kovalevskaja group leader within the Institute of Neuroscience, he became a fellow of the TUM Institute for Advanced Study and one of the university’s first tenure track professors, as well as a principal investigator in the Excellence Cluster CIPS-M (Center for Integrated Protein Science Munich).

Now a full professor, he is director at the TUM Institute of Neuronal Cell Biology and an associate member of the German Center for Neurodegenerative Diseases (DZNE). In 2012, Misgeld and a number of collaborators at TUM, DZNE and LMU established the Munich Cluster for Systems Neurology (SyNergy), an Excellence Cluster dedicated to investigating the mechanistic basis of neurological diseases. Misgeld and Prof. Christian Haass, a renowned Alzheimer’s researcher at DZNE and LMU, are co-spokespersons of SyNergy.

Misgeld’s team uses confocal and two-photon microscopy to carry out in vivo measurements on nerve fibers in model organisms. The image on page 26 was recorded in this way.

Picture credits: eduundsepp

And that is not the only peculiarity. FAD typically also involves deformation of the mitochondria. These act as a cell's power houses, moving back and forth along the axons and supplying the neurons with energy – and have also been found to swell up in FAD. Thus, mitochondria are damaged very early on in MS, and Misgeld and his colleagues are also aware that: "Oxygen and nitrogen radicals produced by the immune cells play a major role here." Using molecular imaging as a read-out, the researchers were able to conduct pharmacological experiments in mice that demonstrate that these two free radical types can damage the mitochondria and trigger FAD. If the aggressive molecules were neutralized by appropriate substances, the affected axons were able to recover. So the researchers have reason to believe that FAD could also be reversible in MS.

According to Misgeld, whether patients stand to benefit from these findings also depends on resolving a series of questions: What exactly happens inside the mitochondria? Is their damage a trigger in the FAD cascade? Which intracellular signaling pathways are activated by the radicals? What is the role of the spike in axonal calcium levels observed in the early stages of FAD?

There is a chance that it might become possible to influence this molecular process with the right medication – always assuming that it can be completely decoded at some point. FAD could then be a potential treatment target. However, as Misgeld cautions: What works in a mouse model is by no means guaranteed to work in patients. And it is not yet known whether structural recovery of axons goes hand in hand with full recovery of their function. Similarly, it is still unclear whether FAD is the sole mechanism behind axonal degeneration in MS.

Other questions preoccupying the neurobiology specialist include: Are the mechanisms of axonal degeneration in MS unique to that condition, or do similar processes also play a role in other neurological disorders, for instance in dementia or following trauma? And are the disease-induced mechanisms related to the physiological processes involved in the development of the nervous system, during which numerous axons are dismantled and remodeled?

As it stands, nobody knows the answers – since, according to Misgeld, there are still huge gaps in our basic understanding of the nervous system. "We must now focus intensively on the structural and developmental dynamics of the brain."

Evdoxía Tsakiridou

Cluster for Systems Neurology (SyNergy): **Investigating the Mechanisms Underlying Neurological Disease**

"It is becoming increasingly clear that, in all neurological disorders, the main pathomechanism is always accompanied by other pathological processes that influence the course of disease. In stroke patients, for instance, blockage of a blood vessel triggers inflammation. And in multiple sclerosis, which is primarily an immune system disease, inflammation leads to degeneration of the axons. Certain immune cells play a central role in Alzheimer's too," underscores Thomas Misgeld, Professor at TUM's Institute of Neuronal Cell Biology and a researcher at the German Center for Neurodegenerative Diseases (DZNE).

So the progression of many neurological disorders might be determined by mechanisms common to multiple conditions – an analytical level that has not played a central role in research efforts to date, since scientists have typically considered each pathology within its own limited context. However, for some time now, researchers and clinicians have been striving to link up traditionally separate specializations within neurology (inflammation, degeneration, vascular disorders and glial cell dysfunction) and develop Munich into the European center for systems neurology – still a young scientific field.

To achieve this, Munich's two universities joined forces with the German Center for Neurodegenerative Diseases (DZNE), the Max Planck Institutes of Biochemistry, Neurobiology and Psychiatry, and the research center Helmholtz Zentrum München to establish the SyNergy excellence cluster. This research alliance has received around 30 million euros in funding from Germany's Excellence Initiative since 2012 and is coordinated by Alzheimer's researcher Christian Haass (LMU) and Thomas Misgeld (TUM) as co-spokespersons.

Around fifty scientists and their teams are investigating the joint mechanisms of inflammatory, degenerative and vascular diseases of the nervous system within the SyNergy cluster. This is accomplished through so-called "Tandem Projects", which involve collaboration between at least two SyNergy researchers from different fields within the areas mentioned. "The idea is to explore the overlaps between the various areas of specialization – for instance between neuroinflammation and the glio-vascular system," specifies Misgeld. This broader focus across traditional boundaries is what sets the cluster apart. The initiators are looking to gain more comprehensive insights into neurological pathomechanisms with an eye on the bigger picture. They also want to give the members of the cluster the opportunity to enter new areas of research and collaborate across disciplines.

However, it is not just a question of joint research, but also of intensive collaboration between institutions. "We have the Institute for Stroke and Dementia Research under the same roof as DZNE – and soon also the MS research center at TUM as a similarly collaborative institute. The interplay and collaboration between these areas is what we are striving to reinforce. We have already cleared the first hurdle by establishing the cluster as a joint initiative. The partnership between the two Munich universities, and especially with Prof. Haass and the other colleagues at DZNE, is the cluster's major strength. And now we intend to tighten the cooperation still further," reports Misgeld.

Applications for the next round of the Excellence Initiative are now in the preparation stage, with participants seeking to secure funding of 20 to 50 million euros for the next seven-year period. "It has a huge lever effect. Because it also motivates other faculties and institutions to join forces and invest in this area," Misgeld concludes. "In the coming years, we hope to build the necessary infrastructure to totally transcend the boundaries between institutions and research topics – as we have already started to achieve at DZNE. The MS center is another good example of this. On the one hand, its outpatient department will treat MS patients and pursue clinical research. And on the other, the floors above will house research labs – not just for clinicians, neurologists and immunologists, but also for neuro- and developmental biologists tackling the basic issues underlying MS. We believe that integrating basic research is absolutely essential to gaining a better understanding of the pathomechanism of neurological disorders."

Evdoxía Tsakiridou

Picture credit: ediundsepp