Circuit Breaker for the Immune System

If the immune system is unable to get to grips with an infection or a tumor, it switches into a lower functional state. Prof. Dietmar Zehn has identified the molecular circuit breaker that causes the immune system to switch between active and reduced functional states. The ability to systematically and more effectively reactivate these exhausted immune cells could pave the way for new treatment approaches for both chronic infections and tumors.

Kurzfassung · Langfassung: www.tum.de/faszination-forschung-25

Schutzschalter für das Immunsystem

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Wenn das Immunsystem Infekte oder Tumore nicht in den Griff bekommt, schalten die zytotoxischen T-Zellen nach einer Weile in einen reduzierten Funktionszustand. Das beeinträchtigt zwar ihre Fähigkeit, ein Virus zu eliminieren, hat aber für den Körper auch Vorteile, denn eine dauerhaft starke Immunantwort belastet Zellen und Gewebe schwer. Bei Krebspatienten lässt das Herunterschalten der T-Zellen allerdings Tumore massiv weiterwachsen.

Prof. Dietmar Zehn identifizierte den molekularen Schalter, der den Wechsel zwischen aktivem und reduziertem Zustand des Immunsystems auslöst - das TOX-Protein. Es bindet an die DNA der T-Zelle und sorgt dafür, dass bestimmte Gene abgelesen werden, die notwendig sind, um den Erschöpfungszustand auszulösen, der Oberflächenrezeptor PD-1 etwa.

Die Forscher verwenden das Mausmodell und einen Transfer gentechnisch veränderter T-Zellen. Dabei konnten sie zeigen, dass T-Zellen ohne TOX-Gen ihre Aktivität nicht drosselten. Allerdings machten die Forscher eine andere wichtige Entdeckung: TOX schützt nicht nur das betroffene Gewebe, sondern auch die T-Zellen vor einem frühen Tod. Diese Erkenntnisse eröffnen neue, molekular zentrierte Ansätze um die Abschwächung von Immunantworten zu verhindern. Gleichzeitig liefern sie neue Einblicke, wie das Überleben von T-Zellen in chronischen Infektionen reguliert wird. Beides ist wegweisend hin zu effektiveren Immuntherapien gegen chronische Krankheiten und Krebs. □

Graphics: ediundsepp

Link
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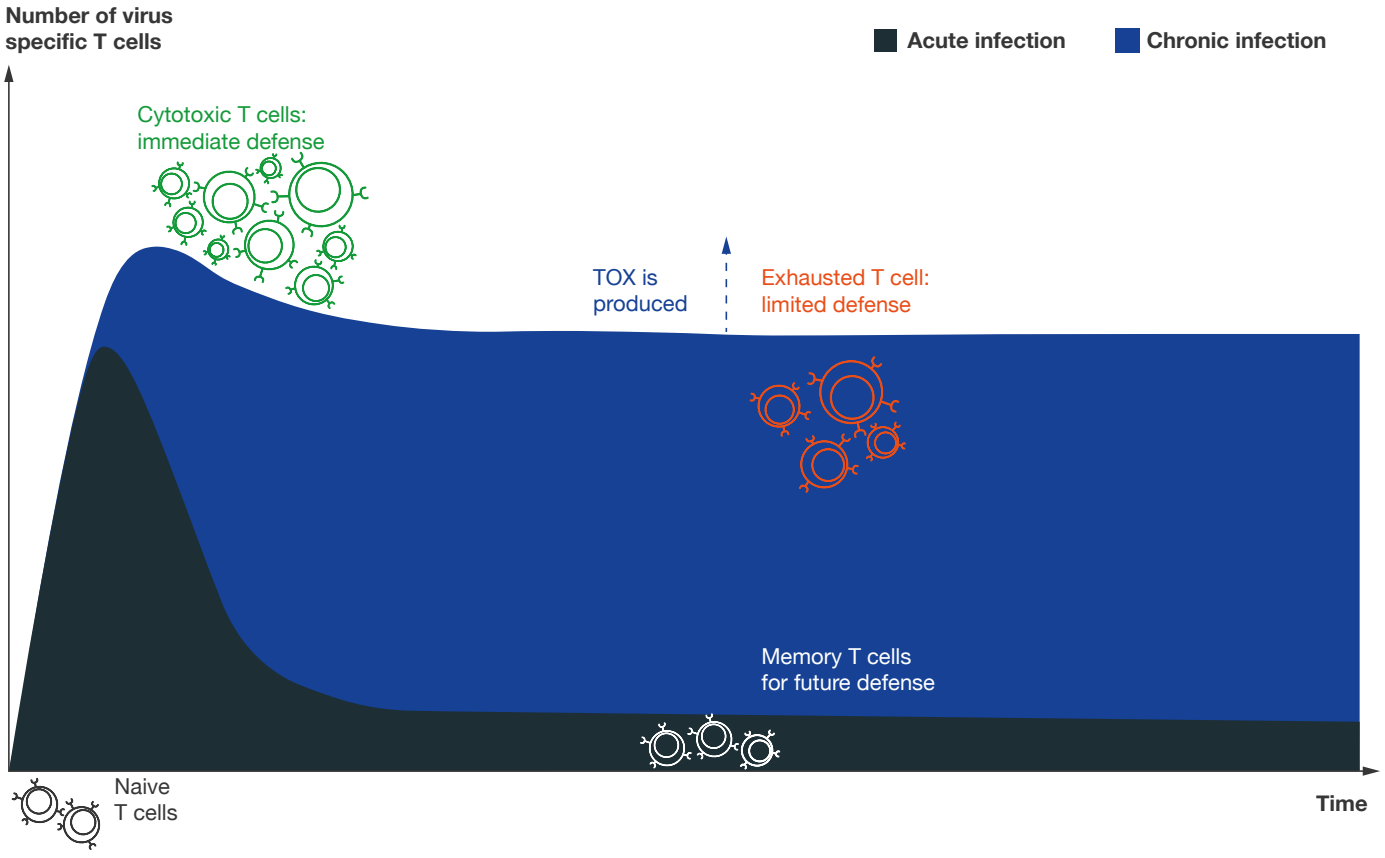
Whether it involves a virus, bacteria or single-cell parasites known as protozoa, when the body becomes infected with a pathogen it puts the immune system on high alert. While the intruders infect the somatic cells, immune cells such as T cells and B cells activate, expand, and aggressively fight to eradicate the pathogen. Cytotoxic T cells – specialized cells in the immune system – play a particularly important role, as they are responsible for eliminating infected cells or tumor cells. They are able to do so because affected cells normally have different proteins on their surface that act as a red flag, identifying them to cytotoxic T cells.

If T cells are unable to overcome the disease after several days or weeks, they often switch to a state in which their functions are significantly reduced – known as exhaustion. Although the cells can keep the pathogen or the tumor in check to some degree in this state, they will not be able to defeat it completely. An acute infection, such as hepatitis C or HIV in humans, can then transition from an acute form into a chronic state. Nevertheless, the immune cells' state of exhaustion also has its benefits. For one, the relentless onslaught of an immune system on high alert causes significant damage to the affected tissue. By reducing T cell activity, the body finds a compromise between the disease and the collateral damage that a prolonged aggressive immune response would enact. This is not the case for cancer patients. For them, the state of exhaustion is unequivocally negative, as the T cells switching to reduced functioning allows tumors to grow a great deal faster.

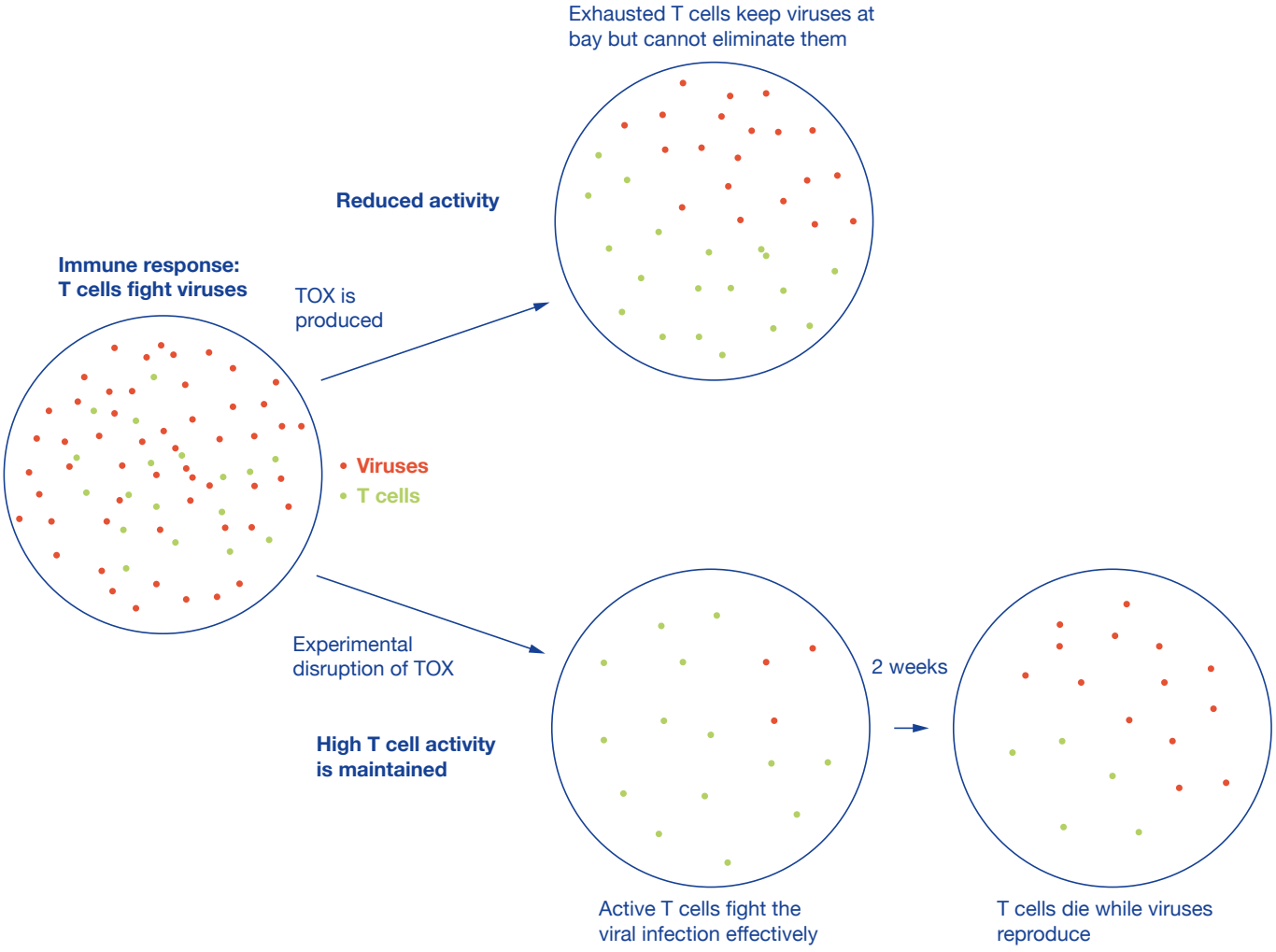


Progress of the immune response to viral infections. The immune system identifies cells infected by viruses as foreign structures. At the start of an infection, there are only few immune cells specific for cells afflicted by the virus. These virus specific, cytotoxic cells rapidly reproduce and destroy the cells which replicate the viruses. In the case of an acute infection (black area), this is successful. Subsequently, a few virus specific cells remain as memory T cells, forming a defense for future infections. For chronic infections (blue area), the cytotoxic T cells cannot completely eliminate the viruses. At some point they fall into an exhausted state. This state goes along with the production of the protein TOX. As a result, the body maintains a limited immune defense which holds the virus infection at bay but does not eliminate it.

Switching the cell's state on purpose

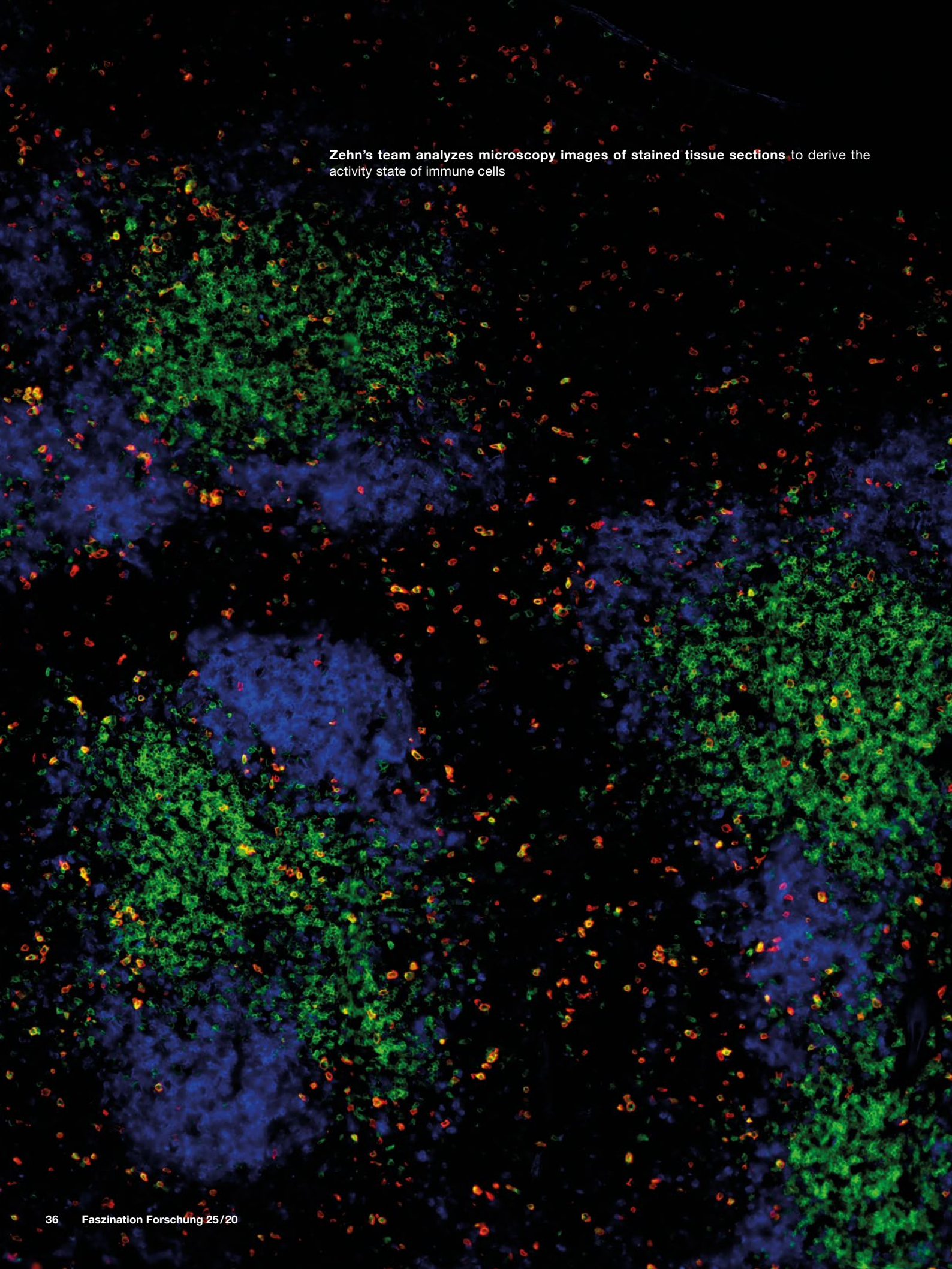
Prof. Dietmar Zehn's research team, supported by colleages from the University of Freiburg, Germany, the USA and Israel, recently identified the molecular switch that causes cells to transition from active to exhausted state. The Professor of Physiology and Immunology at TUM's Weihenstephan School of Life Sciences published the results of this study in renowned academic journal "Nature" – at the same time that two other working groups from the USA independently arrived at the same result. The projects backed each other up on key points and showed that the protein TOX functions as a molecular regulator of exhaustion.

Researchers investigating tumors and infections had long been searching for a molecule that triggered cells to change between active and exhausted states. Following this breakthrough, researchers hope it might be possible to deliberately alter the functional state of these cells in the near future. A more effective ability to systematically reactivate exhausted immune cells would pave the way for better treatment approaches for both chronic infections and tumors. Before that can happen, however, researchers need to understand the underlying mechanisms in greater detail. ▷



Graphics: ediundsepp (source: TUM)

Dietmar Zehn's team was able to show in experiments that T cells which can produce TOX reduce their activity in the course of the infection. The number of viruses drops but is not reduced to zero. A kind of balance develops. T cells without a TOX gene maintain their active state and are initially better able to fight viral infections. However, after some time, the number of T cells begins to fall and the virus increases again.



Zehn's team analyzes microscopy images of stained tissue sections to derive the activity state of immune cells

"The state of exhaustion actually appears to be useful because it protects the body in chronic infections from excessive and damaging immune responses."

Dietmar Zehn

The protein TOX normally bonds in the nucleus with genetic material (DNA) and regulates the transcription of certain genes that are needed to trigger a state of exhaustion in T cells. When researchers inactivated TOX by removing its DNA binding domain, the infection no longer triggered a state of exhaustion. However, this only applied in an early stage of infection. "TOX is only needed to switch the T cells to a state of exhaustion but not to maintain this state," says Zehn. Epigenetic changes in the T cells' DNA appear to play a role in this. Bonds to chemical compounds permanently activate these genes, after which time the bond with the regulator itself is no longer needed. The state of exhaustion remains highly stable.

Precisely which gene TOX directly activates is not currently known. "However, we have identified a strong correlation with the PD-1 receptor," says Zehn. Receptors are molecules on the cell surface, which allow other molecules to dock on and thus trigger signal processes in the cell. This receptor, named Programmed Death 1, was one of the discoveries for which the 2018 Nobel Prize in Medicine was awarded, after scientists in the USA and Japan showed that inhibiting receptors such as PD-1 enhances the efficacy of tumor-fighting treatments. Zehn points out that this underlines the relevance of his field of research. The mechanism of action of TOX remains unexplained; however, interpretation of the results suggests that TOX activates a genetic program in the nucleus of T cells, which in turn causes PD-1 receptors to emerge on the cells' surface. "That definitely isn't the entire mechanism yet, but it is an important aspect," says Zehn.

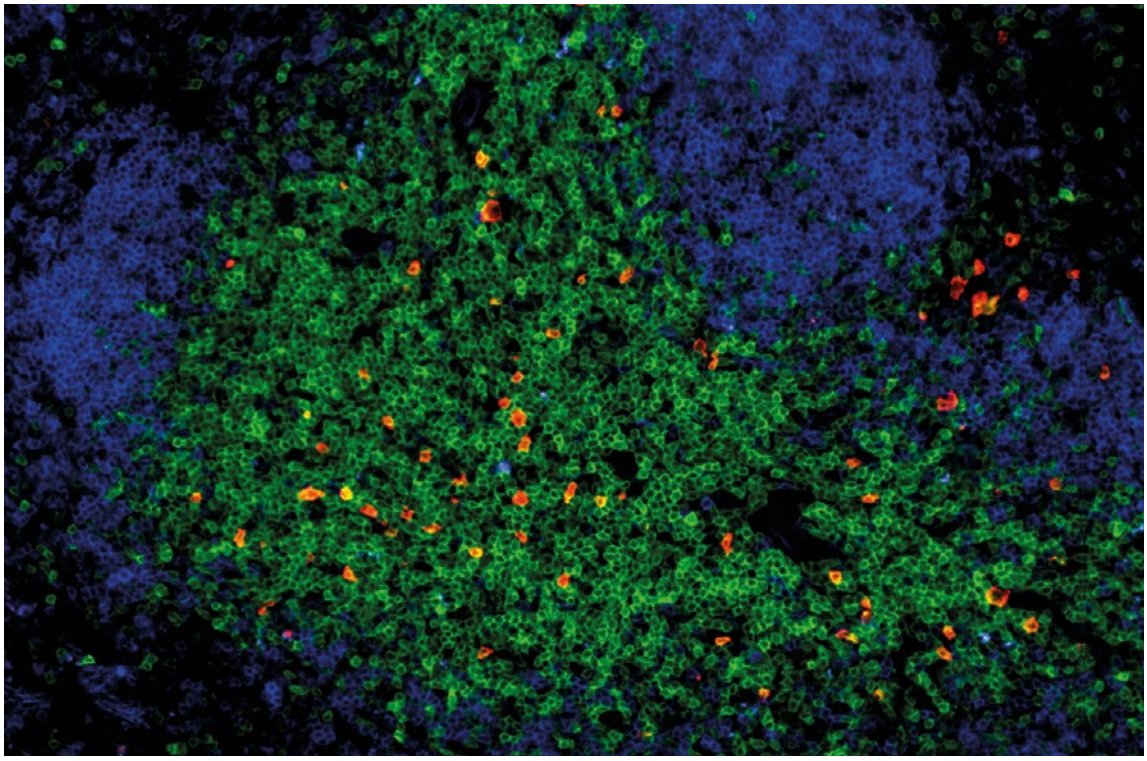
The protein TOX is the key

The researchers have been using laboratory mice infected with a virus and administering genetically modified T cells without a TOX gene. Using this method, they have been able to show that T cells without a TOX gene do not throttle back their activity. However, this change was associated with two major drawbacks. First, although mice with T cells without a TOX gene were initially better able to fight viral infections, their overall health was worse than that of their relations with unaltered T cells. This comes down to the fact that an unregulated immune system can actually cause greater damage than the infection itself. ▷

Picture credit: TUM

"We will only be able to influence the immune system in a targeted manner in emergency situations once we understand its mechanisms and how it works."

Dietmar Zehn



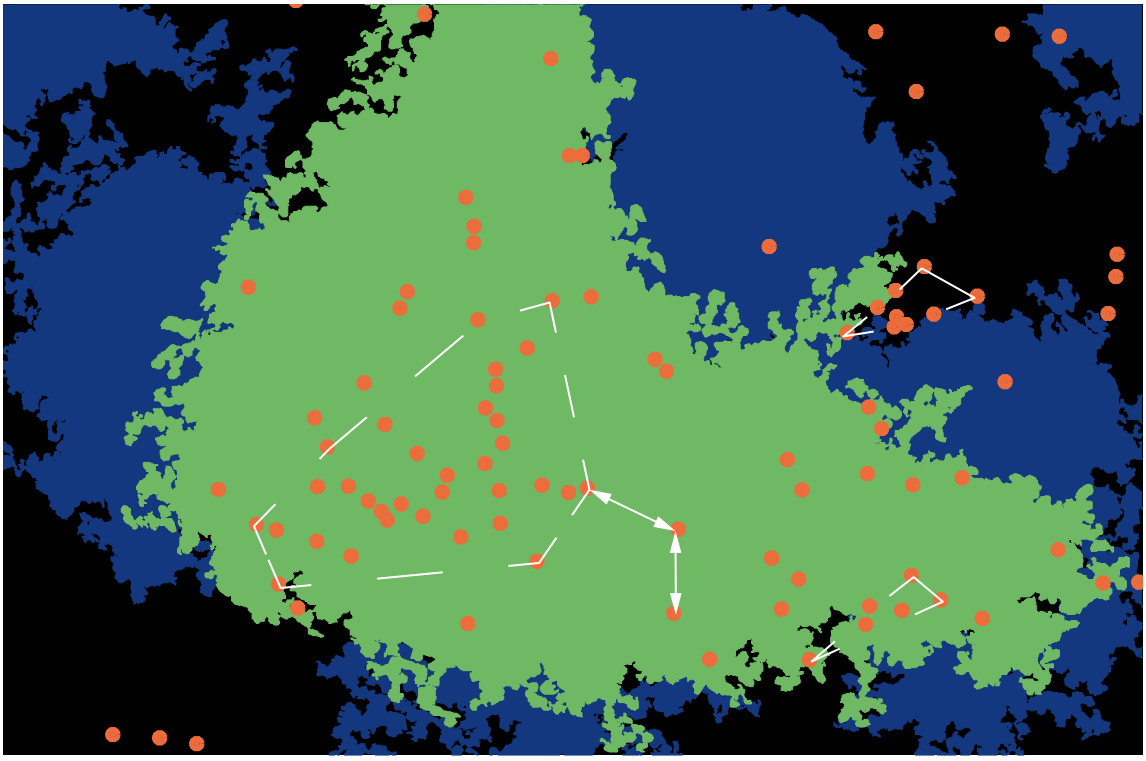
The position and accumulation density of cells helps researchers to learn about their state of activity. The picture shows a microscopy image of stained tissue sections of the spleen. Blue: B cell zone; green: T cell zone; red: pathogen specific model T cells that were activated during an infection.

Without TOX, cells maintain their active state

Second, even without TOX, the T cells only remained active for two weeks. After that point, the virus continued to reproduce while the number of T cells without the TOX protein began to fall. “The cells probably die when they are strongly activated for long periods and cannot enter a state of exhaustion,” continues Zehn. TOX therefore not only protects the affected tissue from excess damage, but also saves the T cells from an early death.

Activated T cells are short-lived. “However, in the case of chronic diseases and tumors, a small subfraction emerges that constitutes extremely long-living T cells and a sort of stem cell population,” says Zehn. These stem cells maintain the immune response and continue to produce new virus specific cells. The more of these stem cells are present, the more likely it is that a patient will respond well to different tumor therapies. “We are currently working with colleagues on a way to apply this to develop a treatment for tumor patients,” says Zehn.

His next objective is to find molecules that make it possible to manipulate cells’ functional state. Switching in both directions could play an important role in developing new therapeutic approaches. Patients with chronic infections such as hepatitis C, HIV or tumors would benefit from an increase in cell activity. However, it is possible that reducing T cell activity could benefit patients with other illnesses, such as autoimmune disorders and even some infections. “For instance, in the context of COVID-19, caused by the coronavirus, it is conceivable that patients with severe symptoms might have immune systems that are overshooting the mark, with TOX playing a role in this,” says Zehn. The question of why most people handle the virus well while others struggle is one that interests Zehn. ▷



The microscopy image is computerized into a vector graphic. The dotted lines indicate local accumulations, while arrows denote distance measurements. The researchers use these to calculate accumulations in specific areas and measure cell-cell distances.

Graphics: TUM; Picture credit: TUM

Is there some kind of dysregulation – and, if so, why? Have patients who present with more serious symptoms perhaps previously experienced an infection with a similar or different pathogen? The immune system protects us against re-infections with the same pathogen. In some cases, however, a second infection with a similar but not identical virus takes a more severe course than the initial infection. It is still not clear whether this is the case for COVID-19. “What is certain is that we will only be able to influence the immune system in a targeted manner in emergency situations once we understand its mechanisms and how it works,” says Zehn.

Karoline Stürmer

Prof. Dietmar Zehn

After studying medicine at Charité in Berlin, Dietmar Zehn spent five years conducting postdoctoral research at a renowned immunological laboratory in the USA at the University of Washington. He devoted his time there to basic research into infectious and autoimmune diseases. Subsequently, Zehn managed a laboratory in Switzerland for six years, during which time he was awarded a sponsored professorship by the Swiss National Science Foundation (SNF). In 2015, he accepted a position at TUM and assumed the Chair of Physiology and Immunology at the Weihenstephan School of Life Sciences. His field of expertise comprises molecular and cellular mechanisms in T-cell-induced immune responses to acute and chronic infections and tumors, as well as immune tolerance and autoimmune diseases. Dietmar Zehn has received numerous awards and fellowships in the course of his career. In 2015, he received the European ACTERIA Prize for Immunology. In 2018, he successfully applied for an ERC Consolidator Grant, which directly followed the ERC Starting Grant he had obtained in 2013. Both grants run for five-year terms and are aimed at elite researchers distinguished by their excellent research. In 2020, he acquired a new Center for Integrated Infection Prevention for TUM, co-financed by the federal state and the federal government, in a competitive procedure.

EUR 40 Million for **New TUM Institute**

TUM is going to bundle its competencies for the development of innovative strategies for preventing, combatting and avoiding the spread of resistant pathogens in humans and animals.

The marked increase in resistant bacteria and the associated massive rise in the danger of infections in both humans and animals which cannot be treated with antibiotics is, in the long term, one of the biggest scientific, medical and social challenges of our time.

"Without innovations, we are at risk of regressing to the pre-antibiotic era in which simple injuries could develop into deadly threats," state TUM professors Dietmar Zehn, Percy Knolle and Bernhard Küster. They represent the team of researchers who supported the application to found the institute. "The number of deaths caused by infections, which is just under one million per year, could rise to about ten million by 2050."

TUM therefore combines its competencies for the development of innovative strategies for preventing, combatting and avoiding the spread of resistant pathogens within a new research facility, the Center for Integrated Infection Prevention (ZIP).

The federal government and the Free State of Bavaria will support the new construction project at the Weihenstephan campus equally to the tune of roughly EUR 40 million in total.

Fast transfer of research into practice

The new center's research work is divided into three program pillars: modulation and dynamics of the microbiome, strengthening of the local immunity on microbially populated boundary surfaces, and innovative technologies.

With cross-species observation of resistant bacteria in humans and farm animals, the ZIP lays emphasis on the intersection of medicine, life sciences, microbiology, bioanalytics and information sciences, an emphasis that is unique in the world.

One of the most important goals pursued by the institute is to put new prevention strategies into practice as fast as possible. The idea is to largely avoid the use of antibiotics in livestock farming, to better control existing infections and to suppress transmission paths between animals and humans.

Positioned in a unique research environment

Bernd Sibler, Minister of Science, said about the funding of the ZIP: "Through their multidisciplinary research, TUM offers a nationally and internationally outstanding and stimulating research environment for the pressing future topic of bacterial resistance. It is therefore the right place for the Center for Integrated Infection Prevention, a state-of-the-art research facility in which their research competencies are combined to facilitate interdisciplinary approaches at the intersection of health research and big data, from the agricultural and nutritional sciences to biomedicine to computer sciences, and strengthened at an international level."

"Infection prevention as a central objective of the ZIP is more relevant than ever and requires completely new research approaches," says TUM president Prof. Thomas F. Hofmann. "In order to activate the enormous potential of the ZIP, we have already created the critical, interdisciplinary environment through strategic appointments for top-level positions, pooling of financial resources and successful efforts in the promotion of young researchers, equal opportunities, diversity and technology transfer."

■ *Andreas Battenberg (TUM)*

"Infection prevention as a central objective of the ZIP is more relevant than ever and requires completely new research approaches."

Prof. Thomas F. Hofmann

Picture credit: Astrid Eckert