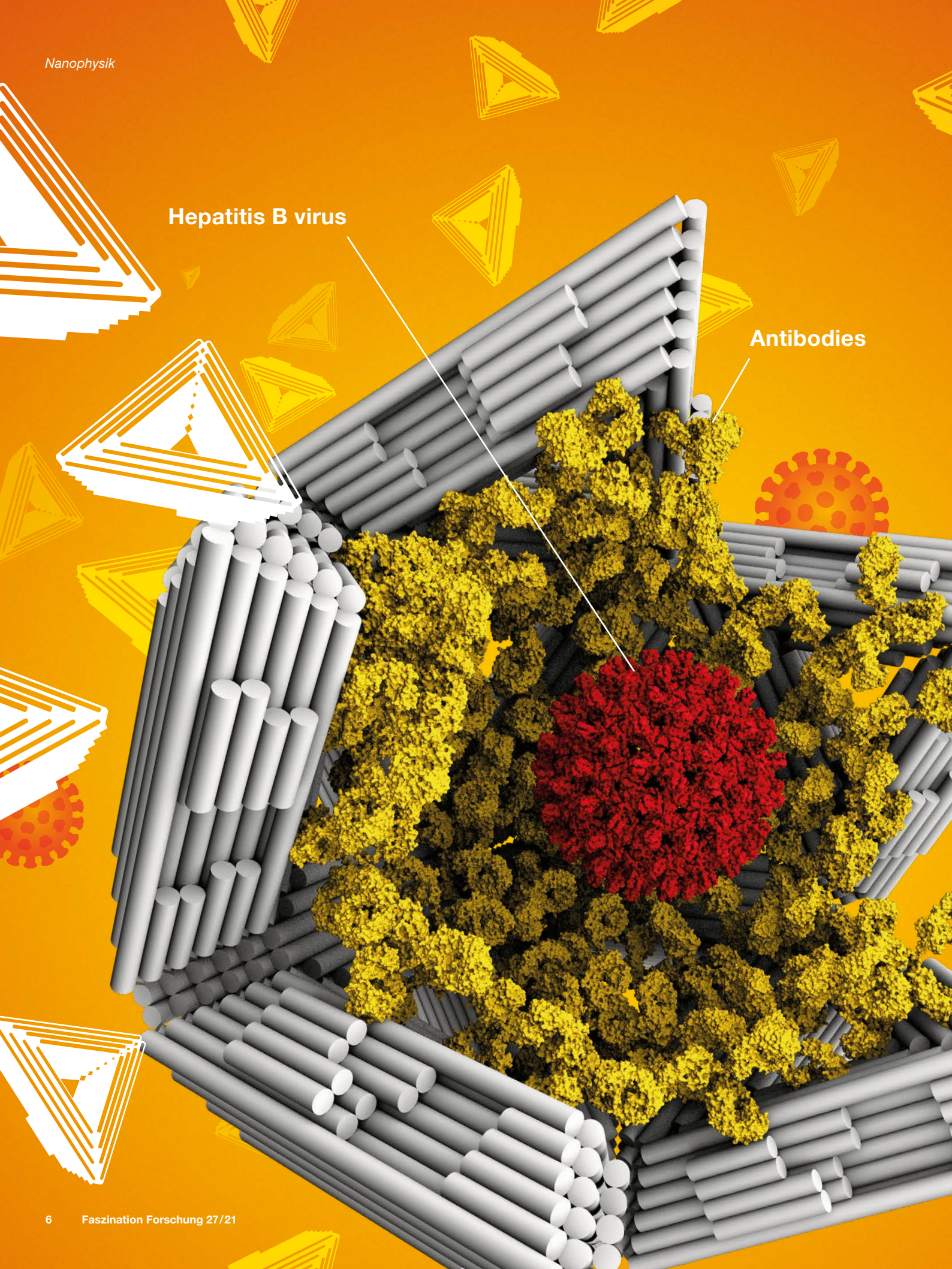
Hepatitis B virus
Antibodies

Link

www.dietzlab.org

Quarantine for Viruses

Though it might seem like an idea from the world of science fiction at first, it could soon become reality: Researchers hope to capture viruses in minute DNA capsules that prowl through the human body. TUM Professor Hendrik Dietz and his team have already laid the foundations for such a technology.

Graphics: ediundsepp (source: C. Sigl/TUM)

DNA capsule

Gesamter Artikel (PDF, DE): www.tum.de/faszination-forschung-27

Quarantäne für Viren

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In dem von der EU geförderten Projekt VIROFIGHT entwickelt Hendrik Dietz, TUM Professor für Biomolekulare Nanotechnologie, zusammen mit seinem Team und Fachleuten aus angrenzenden Gebieten eine radikal neue Technologie zur Bekämpfung von Viren. Das Team macht sich dabei die jüngsten Fortschritte in der supramolekularen Chemie, der molekularen Nanotechnik und der Virologie zunutze. „Unser Ansatz betrachtet ein völlig neues antivirales Konzept“, betont der Forscher. „Anstatt wie die derzeitigen antiviralen Medikamente auf die Funktion virusspezifischer Proteine oder Enzyme zu zielen, wird VIROFIGHT ganze Viren mit synthetischen Nanokapseln einschließen, um den Erreger effizient zu neutralisieren.“ Dies wäre eine absolut neue therapeutische Lösung, die verschiedene Viren auf derselben Plattform bekämpfen könnte, und würde im Erfolgsfall einen echten Durchbruch für die antivirale Medizin darstellen. Das innovative Konzept könnte viele Leben retten und enorme Gesundheitskosten für die Gesellschaft einsparen. □

Prof. Hendrik Dietz

was born in 1977, grew up in Berlin and studied physics at LMU Munich. After being awarded his doctorate in protein mechanics, in 2007 he moved to Harvard Medical School where he joined a group of researchers led by William Shih seeking to produce three-dimensional objects from DNA. Together with computer scientist Shawn Douglas and other colleagues, he become known as a pioneer of DNA origami. In 2009, accepted a position at TUM as Extraordinary Professor for Experimental Biophysics, and was made a full professor in 2014. Dietz has received numerous awards and grants for his work, including the Gottfried Wilhelm Leibniz Prize from the German Research Foundation (DFG) in 2015. He received a Starting Grant from the European Research Council (ERC) in 2010, followed by a Consolidator Grant in 2016 and an Advanced Grant in 2021. Dietz was made an honorary member of the North Rhine-Westphalian Academy of Sciences in 2019.

The coronavirus crisis has emphasized the point that while antibiotics are available to treat bacteria, we have far fewer drugs at hand to treat acute viral infections. While vaccinations have been created to counter some viruses, the process of developing such preventive measures is a tedious one and each vaccine has to be precisely tailored to the properties and characteristics of each virus.

What if we had a general-purpose weapon to fight and eliminate viruses, from COVID and influenza to hepatitis and HI viruses? Researchers are on track to make it a reality. Hendrik Dietz, Professor of Biomolecular Technology, and his team have developed minute traps that capture viruses in the body and “swallow” them. This is an utterly novel concept that sounds brilliantly simple. “As far as we know, what we are proposing has never been attempted before,” the researchers say, in prosaic language typical of scholars. “If successful, it would represent a disruptive advance.” ▷

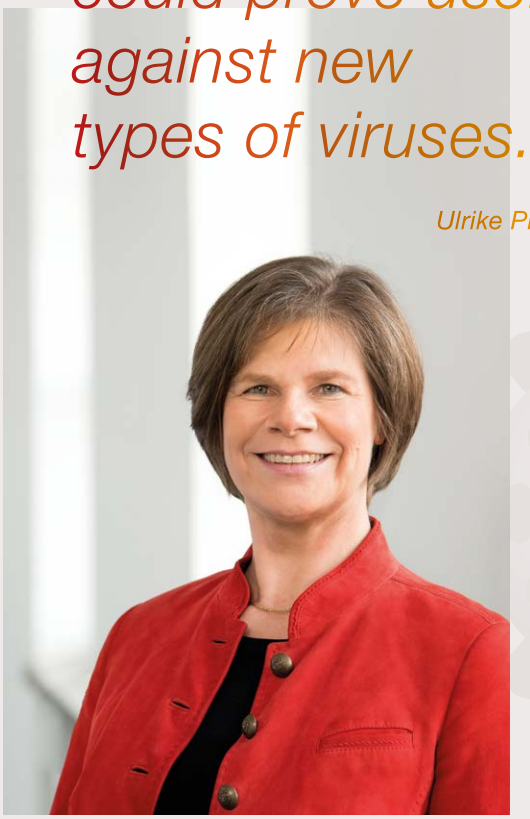
“As far as we know, what we are proposing has never been attempted before.”

Hendrik Dietz

Picture credit: Astrid Eckert

“The procedure could prove useful against new types of viruses.”

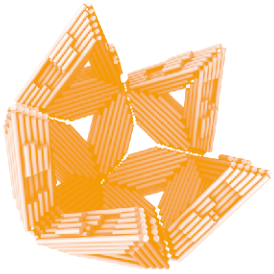
Ulrike Protzer



Prof. Ulrike Protzer has accompanied Hendrik Dietz’s project from the beginning. The virologist, who is Director of the Institute of Virology at TUM and the Helmholtz Center in Munich, provided inactivated hepatitis B viruses that enabled Dietz and his team to trial the process of capturing viruses in DNA capsules.

Professor Protzer, you must have been astonished when a physicist came to you with the concept of capturing viruses in tiny shells?

Ulrike Protzer: A little, yes. But I was already familiar with Hendrik Dietz and his work. That said, I was impressed by the highly complex objects he and his team had been able to construct; I had imagined they would be far more primitive. ▷



DNA triangles
assemble into shells

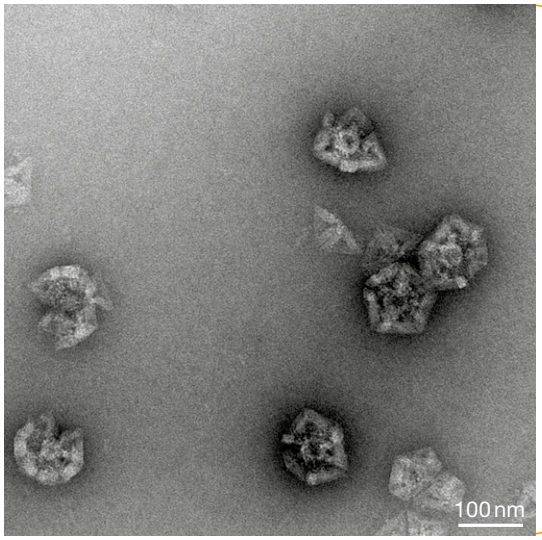
DNA origami triangles

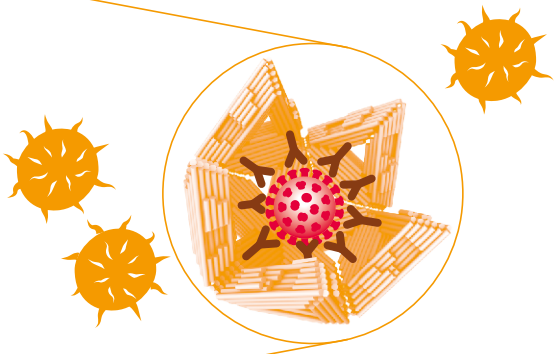
There are two main methods to build virus-trapping capsules. 1 One builds fully prepared half-shells that capture viruses. 2: Smaller building blocks bind to a virus particle across its entire surface, ultimately covering it to form a complete shell.

Antibodies in the shell's
interior bind to the viruses

The virus is captured
and neutralized

Transmission electron microscope image of
hepatitis B particles trapped in half shells



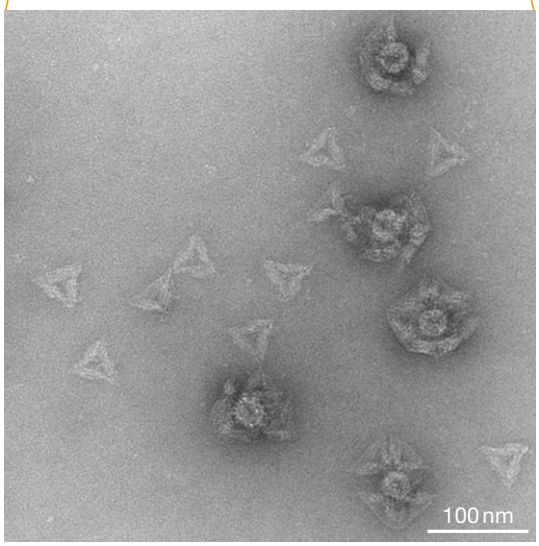


The body's own phago-
cytic cells eliminate the
shell containing the virus

DNA triangles modified with antibodies self-assemble around the virus

Ultimately, the triangles form a complete shell

Transmission electron microscope image of hepatitis B particles trapped by triangles self-assembling around them



Picture credit: Astrid Eckert; Graphics: ediundsepp (source: TUM)

How do you introduce the DNA capsules into the body?
There are different methods depending on where you want them to elicit an antiviral effect. One can inject them intravenously if an effect in the bloodstream is wanted. Alternatively, a patient can inhale them if the aim is to capture viruses in the airway mucosa or lungs. The best way to reach viruses in the gastrointestinal tract will probably be using a type of capsule that you swallow.

Assuming that you actually capture the viruses, what happens next?
The body's own phagocytic cells will eliminate them.

Are there any risks involved? Can the body reject these artificial structures or produce an allergic reaction?
We can't exclude that possibility, but I wouldn't expect so. The material that makes up the capsules is DNA, which is not foreign to the human body. However, it may activate the body's innate immune system. But that might actually even be helpful in fighting viruses. That's something to be investigated in further studies.

Do you believe the project has a realistic chance?
I think it's absolutely fascinating. We need to see whether it can win out over the antibody therapies currently in clinical trials. However, such antibodies always need to be developed specifically against a certain type of virus. In my view, this new approach has an advantage in that it is more universal. We could use these as ready-made kits to capture all kinds of viruses, including new viruses for which no antibodies have been developed yet.

Realistically, how long should we expect development of this treatment to take?
First of all, the project needs to complete preclinical studies, which take three to five years, followed by a clinical trial phase that will last approximately ten years on top. So, it will certainly take a lot of patience and a lot of funding. □

3D DNA

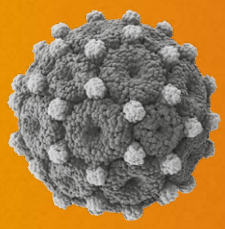
origami

uses a DNA building block to form highly complex, ultra-miniaturized objects.

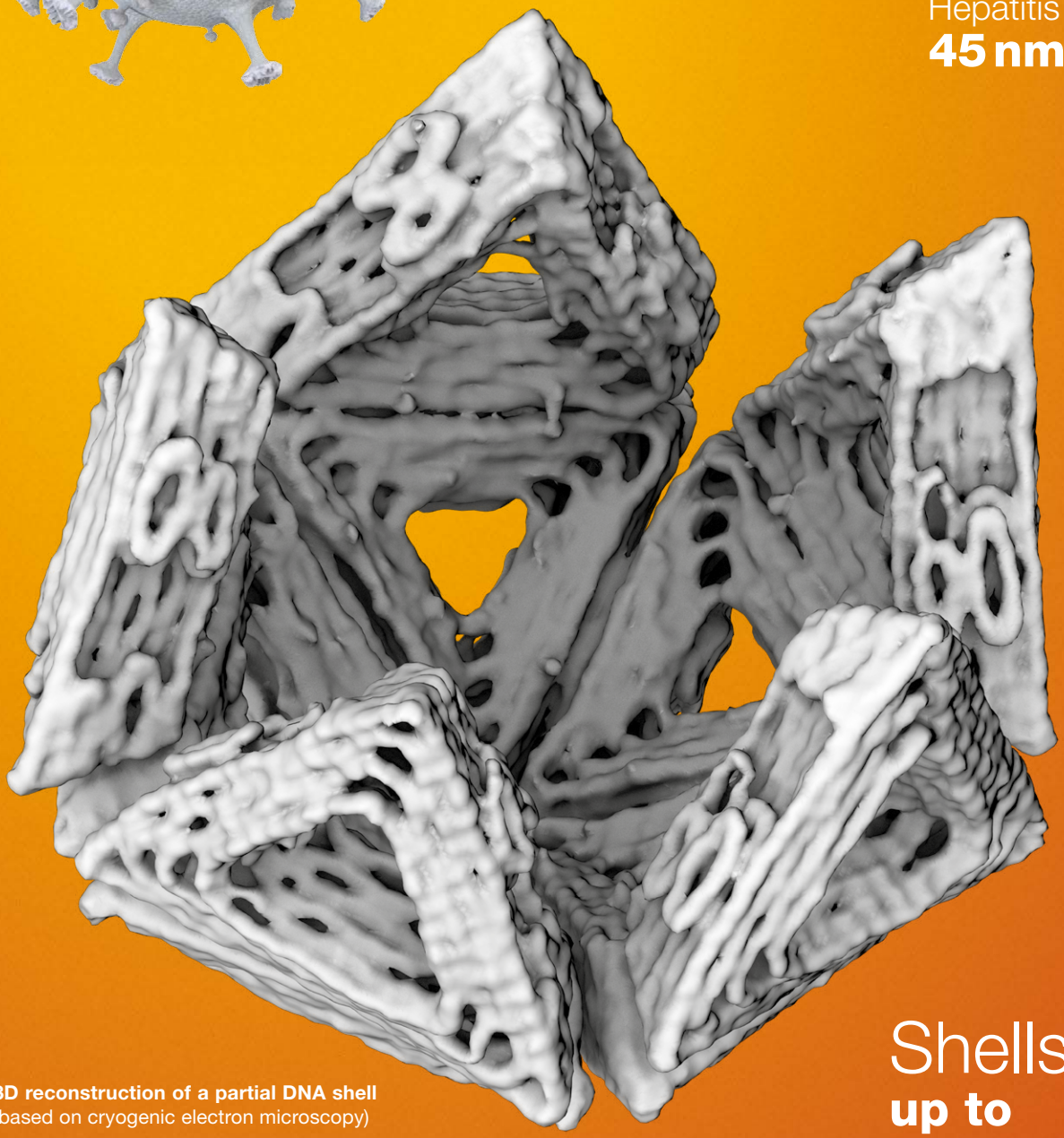
An epiphany in August

It began in summer 2019 – long before anyone was talking about the coronavirus – in Dietz's office at the north end of the Garching research campus. The biophysicist and his team had just managed to build nanoscale capsules from DNA material. The institute is specialized in this field: Over the last ten years, researchers there have refined technologies that use DNA building blocks to form highly complex yet ultra-miniaturized objects. These processes are further evolutions of a nanofabrication concept known as 3D DNA origami, a development Dietz was already involved in as a postdoc at Harvard. What initially had looked a bit like "a technology searching for an application" could now become a vital tool in combating viruses. The researchers based the design for their capsules on viruses and their shells. Back in 1962, the American biologist Donald Caspar published a paper with British biophysicist and Nobel Prize laureate Aaron Klug, outlining how the capsid – the outer shell of a virus – is made up of proteins. They identified that these shells are regular, hollow bodies that correspond to strict geometric rules. Hendrik Dietz and his colleagues Christian Sigl, Elena M. Willner and Wouter Engelen "copied" these geometric specifications together with colleagues Seth Fraden and Michael Hagan from Brandeis University to create their nanoshells and were thereby able to construct a series of stable capsules composed either of ▷

Coronavirus
80–140 nm



Hepatitis B virus
45 nm



3D reconstruction of a partial DNA shell
(based on cryogenic electron microscopy)

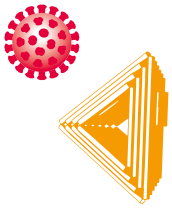
Shells
up to
280 nm

Picture credit: Astrid Eckert; Graphics: ediundsepp (source: C. Sigl/TUM, turbosquid)

Each of the

20 faces

forming the icosahedron is represented by a triangular piece of DNA.



half-shells or smaller pieces of shell. Similar to natural viral particles, these shells are hollow bodies with 20 sides (icosahedrons), but are made from triangular, multi-layered pieces of DNA. Each shell comprises many million individual atoms. By using a sort of click mechanism, the researchers managed to cause the shells' complex building blocks to combine of their own accord. This process of self-assembly thereby creates the spherical shells.

"People have perhaps become a little tired of DNA nanotechnology," explains Dietz. "Although we have made magnificent advances in recent years, we still lacked real-world applications for our technology." Last year, however, a lucky coincidence that researchers like to call serendipity provided an answer. In August 2019, as he looked at his screens showing the newly formed hollow bodies on one side and the similarly structured virus on the other, the thought flashed through his mind that the virus should fit perfectly inside the artificial capsule.

Shells with adhesive interiors

Dietz and his team immediately set about exploring this idea. They created building blocks for shells of various sizes, procured inactivated hepatitis B particles from TUM virologist Ulrike Protzer to act as a model system, and tested whether these particles could be captured in their capsules. "In order for the viruses to stick to the inside of the shells, we equipped the interiors with antibodies that stick to particular features on the surface of the viruses," explains biophysicist Dietz. In principle, however, it should be possible to use any molecule that binds with virus particles, such as short peptide chains or aptamers. These are DNA sequences that can be obtained using a selection process in a test tube. "You then have two different strategies to choose from. First, you can build fully prepared half-shells that combine to form complete, closed capsules after they capture a virus. Or, as a second option, you can use smaller building blocks that dock with the virus particle across its entire surface, ultimately covering it to form a complete shell." Viruses encapsulated in this way would no longer be infectious and could be "disposed of" within the body. Currently, the researchers are working on "virucidal shells", shells whose interiors are coated with an enzyme that can digest virus surface molecules once encapsulated, similar to the way Venus flytraps "eat" the prey they trap.

With the help of their cryogenic electron microscope, the researchers have been able to demonstrate that these

Preparation of DNA nanostructures for imaging with transmission electron microscopy (TEM): The nanostructures are applied on a metal disk ("grid") and stained with fluid die (uranyl formate).

nanoscale virus traps work in test tubes. In other tests, they have proven that viruses captured in this way were prevented from interacting with other surfaces by up to 99%. Experiments with infectious Adeno-associated viruses were able to show the neutralizing effect of the shells with living cells.

These laboratory experiments are important initial steps. The technology, however, must now be tested in living organisms. In separate work, Dietz and his team have learned to produce their nano-objects in large volumes and at acceptable costs and can also stabilize them for use in physiological conditions. ▷

Up to

99%

of interactions between captured viruses and other surfaces were blocked.

Picture credit: Astrid Eckert

EU-funded project

VIROFIGHT

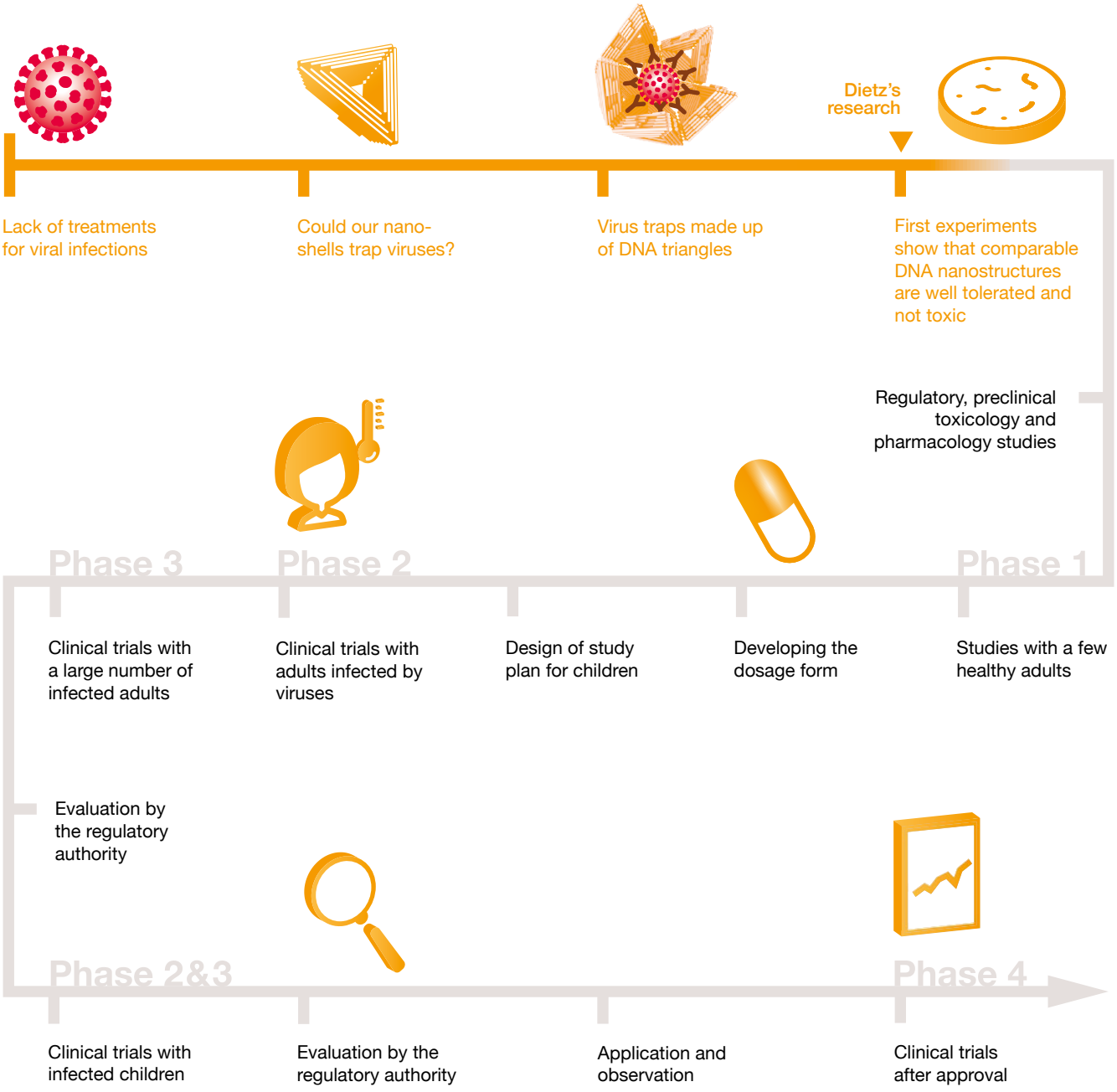
The interdisciplinary consortium integrates experts on supramolecular chemistry, molecular nanoengineering, and virology across Europe.

Experiments on mice soon

In light of the coronavirus pandemic, the research has suddenly taken on an unforeseen currency and urgency. Dietz and his team have been acutely focused on how their concept could be realized in practice; they have discussed it with numerous experts to ascertain whether the concept has a realistic chance as a treatment for viruses. "Of course, we're working to overcome some of the weaknesses and press ahead with practical testing," says Dietz. "There are many viral infections for which we have no therapy so far. To find new therapies we must explore avenues off the beaten path. Our concept is unusual but, in principle, resembles in part aspects of the adaptive immune response. As I understand it, we typically return to health when our body has managed to 'coat' viral pathogens with neutralizing antibodies."

Preliminary experiments on mice at the Helmholtz Center in Munich have already shown that comparable DNA nanostructures are well tolerated and not toxic. The EU has now committed to support the next stages of research as part of a project called VIROFIGHT. An interdisciplinary consortium of doctors, virologists, biophysicists, molecular biologists, chemists and experts in DNA origami and protein design will now get to grips with the concept.

Freezing viruses for long-term storage and preparing DNA nanostructures for gel electrophoresis analysis.



Picture credit: Astrid Eckert; Graphics: ediundsepp (source: VFA)

Their task will be to test the virus trapping process that successfully beat the hepatitis B virus and Adeno-assso-ciated viruses in test tubes with other viruses and in mice. "As soon as we see that encapsulation has positive ef-fects in the mouse model, it will probably make sense to explore setting up a company and look for investors," says Dietz, outlining his plans.

And then – if everything goes to plan – in a few years' time, there could be a universal family of medications capable of fighting viruses, created for the good of humanity. After all, this will not be the last pandemic humanity will face.

Brigitte Röthlein

How Hendrik Dietz's idea could eventually turn into medical treatment – if it proves itself in the many tests on the way. Cur-rently it is at stage 4.