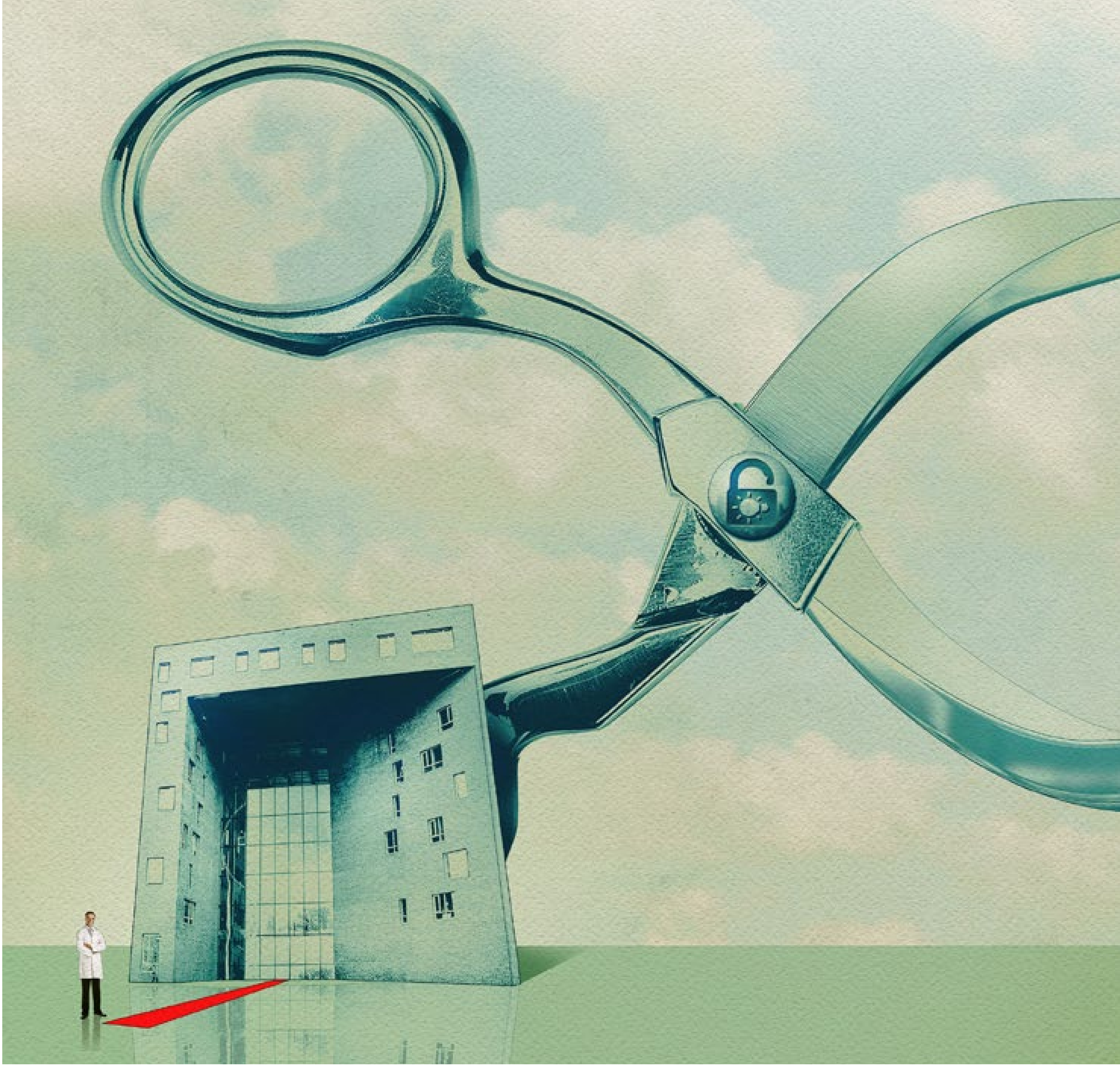
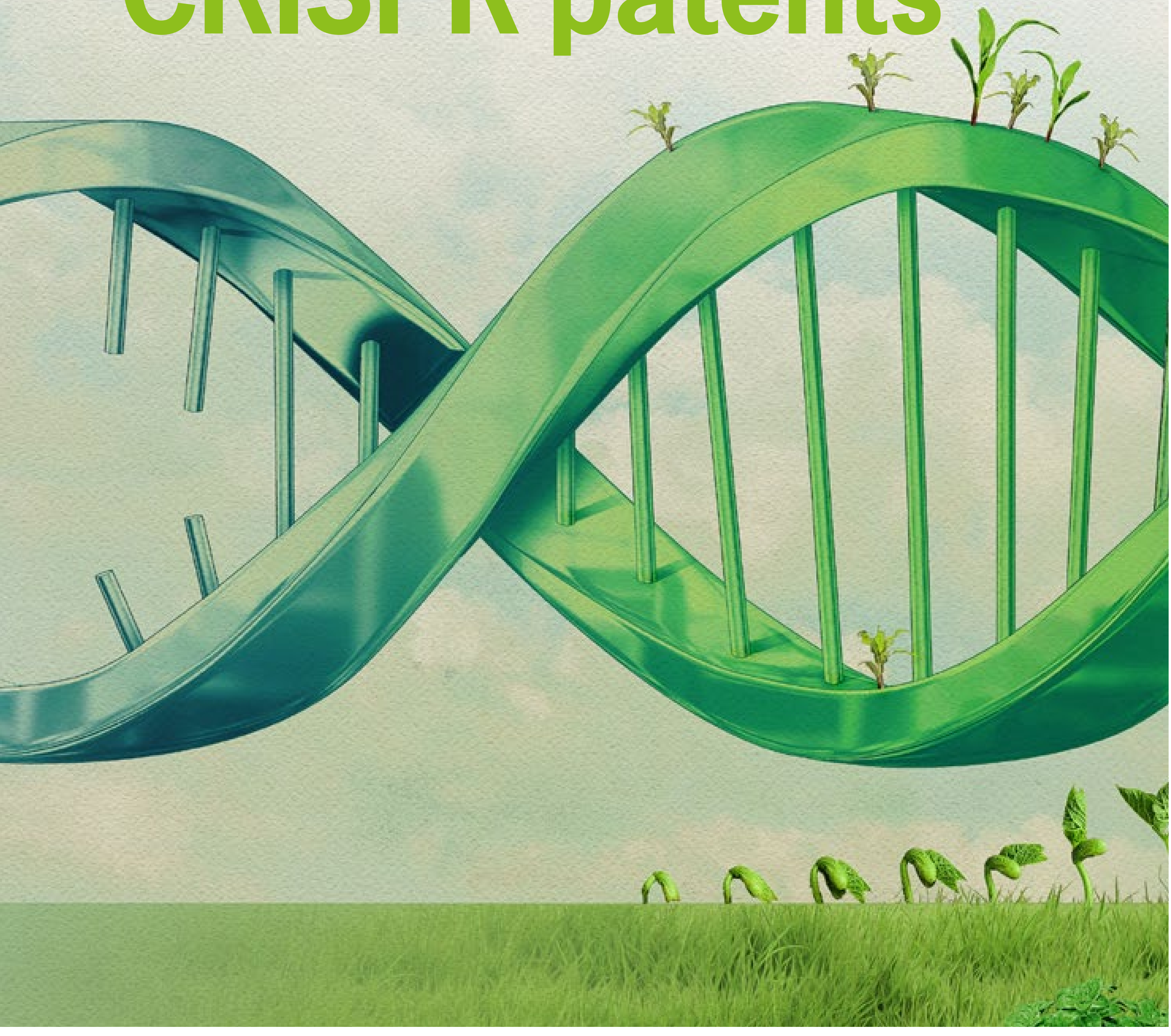
Wageningen is going to give non-profit organizations free access to a number of CRISPR-Cas patents as a stimulus for plant-breeding in emerging economies. But the technical and legal details are being worked out first. 'We do want to monitor how our licence is applied.'

TEXT ARNO VAN 'T HOOG **ILLUSTRATION** RHONALD BLOMMESTIJN



SHARING KNOWLEDGE IN A GOOD CAUSE

Free access to Wageningen CRISPR patents



‘We still have to invent the wheel somewhat; this hasn’t been done before’

‘Worldwide, thousands of CRISPR patents have been applied for. The nice thing is that we’ve got a few of them in our possession,’ says Wageningen professor of Microbiology John van der Oost. ‘That gives us freedom to operate. We can grant access to these patents free of charge. I hope that by doing so we can give research institutes in emerging countries a boost for the development of local crops, to produce higher yields for example. The very best thing would be for the improvement of crops to lead eventually to higher production and therefore to lower prices for consumers.’

In plant-breeding, CRISPR technology offers scope for improving crops faster than is possible by traditional breeding methods. But the forest of CRISPR patents poses a stumbling block, says Van der Oost. Anyone who uses this technique to create a new crop variety and then starts selling the seed is soon in breach of one of these patents. That usually means that licence money has to be paid to the patent holder.

STITCHED UP

The American patent-granting institution USPTO has already registered over 6000 CRISPR patent applications, and another 200 are added every month. Van der Oost: ‘This research field is completely stitched up. That is why it is interesting for other laboratories that we have found a very different variant. Our CRISPR-Cas system does the same work, but the molecular cutting tool looks very different. And it doesn’t matter which organism you take, in theory our system can modify every fragment of DNA in every organism. The target group we have in mind for the free licences to our patent are non-profit

organizations working on improving agricultural crops.'

CRISPR-Cas is applicable in plant-breeding in a variety of ways. A few years ago, there were successful efforts to make cucumber plants resistant to several kinds of plant virus, and to increase salt tolerance in rice. A variety of oil-seed rape was created that makes more omega-3 fatty acids. There are not very many commercial applications yet. Seed producer Dupont Pioneer produces a new hybrid maize variety in the US that creates a kind of starch by switching off a single gene. The starch is of interest to the paper and food industries as a binding agent.

GMO OR NOT?

Does breeding with the aid of using CRISPR-Cas produce a genetically modified organism (GMO)? This question has led to totally different outcomes in Europe and the United States. The American government does not see genome editing as genetic modification, so its use is permitted there. Since a ruling by the European Court of Justice in 2018, however, Europe has adopted the opposite position: CRISPR-Cas crops come under existing GMO legislation. Consequently, stringent approval studies are required and labelling is compulsory for access to the market, to safeguard freedom of choice.

In April 2021, however, the European Commission published a report on new genomic techniques in plant breeding. One of the conclusions was that old GMO legislation on transgene crops is not really appropriate for evaluating genome editing. This report is a starting point for discussions in European countries about revising the legislation for new breeding techniques – a process that could take years.

CUTTING TO SHREDS

Around 2008, Van der Oost's lab was involved in describing the building blocks of CRISPR-Cas, a natural immune system with which many microbes defend themselves against bacterial viruses. The system recognizes the genetic material of viruses and cuts it to shreds. A few years later – around 2011 – several research groups realized that you could use the same system, with a few adjustments, on the DNA in the cells of other organisms. That way you could use genome editing to switch off genes or introduce mutations.

It is a relatively straightforward job to get genome editing with CRISPR-Cas off the ground in a laboratory. Researchers can ask a fellow researcher for the basic materials, or order them online, and methods are described in all sorts of publications. As a result, the technology has spread widely, resulting in a tsunami of publications and patent applications since 2013. The applications are in areas ranging from medicine to microbiology, horticulture and even Covid-19 diagnostics. The first patents spawned dozens of new biotech companies. In other words, CRISPR-Cas is big business.

The patents in the name of John van der Oost and his colleagues look just like any others: page after page of lists of technical claims and descriptions of amino acid sequences and methods. Altogether, they describe the ingredients and the method for use in genome editing. The big difference, however, lies in the origins of the CRISPR-Cas system.

FROM THE COMPOST HEAP

Van der Oost's patents are the product of a discovery his lab made around 2015 in a steaming compost heap in Ede. During the composting process, the temperature goes up to more than 65 degrees, a temperature at which the heat-loving bacterium *Geobacillus thermodenitrificans* flourishes. Van der Oost and his colleagues decided to decipher the genetic material of the bacterium to see whether the species makes use of CRISPR too. This put them on the trail of a thermostable CRISPR-Cas9 which was clearly different to previously identified Cas9 variants.

Van der Oost: 'This new CRISPR system works well, and the study eventually led to five different patent applications. When you apply for a patent, you never know whether it will be granted. It's hard to predict that in advance. But the first patent has since been granted in the United States, and the second and third are on their way.'

Because the CRISPR-Cas comes from a thermophile bacterium species, it does best in warm conditions, says Van der Oost. And that makes it particularly handy for genome editing in other heat-loving >

‘We go one step further with our licences than other institutes’

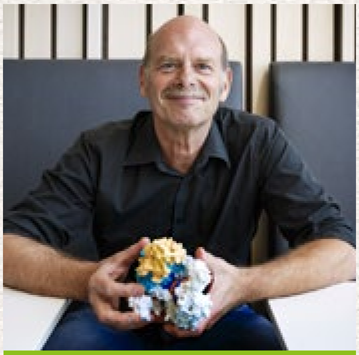


PHOTO ANP

JOHN VAN DER OOST

John van der Oost, Professor of Bacterial Genetics in Wageningen since 2005, is considered a pioneer of the CRISPR revolution. He has been researching the genetic material of micro-organisms for over 25 years. In 2008, he and his chair group were the first to clarify the mechanism behind the CRISPR-Cas defence system in bacteria. This allowed researchers to develop new tools with which they can make rapid and very precise genetic modifications.

micro-organisms. ‘It works optimally at somewhere between 50 and 60 degrees. DNA in a cell gets cut at around 30 degrees too, but less efficiently than it is by other CRISPR systems. What we say about any licence we currently make available to non-profit organizations is: it works, so go ahead and get started. You can already get things done with it but be aware that its efficiency is not yet very great.’
According to Van der Oost, a 2.0 version is on its way that will work better at lower temperatures, hopefully around room temperature. ‘We want to further optimize this CRISPR system for applications in plants, or in animal and human cells. We are working on that. I’m optimistic because we have succeeded previously in reducing the optimum temperature for other enzymes. I expect that we shall have such mutants within two years. Non-profit labs can make use of these too without paying, as long as they share their discoveries with local farmers on a non-profit basis.’

CHARGING A FEE

Meanwhile paid licences for the same Wageningen CRISPR patent have been granted to two companies: for genome editing in micro-organisms in one case, and in human cells in the other. Van der Oost: ‘It’s not a bad idea, after years of research and financing, to charge companies a fee for commercial applications. Sometimes that works out. Most of the licence money is channelled into new research projects. The costs of licences vary: for a small company we’re talking about a few tens of thousands of euros a year; for bigger companies sometimes a few hundred thousand. If a successful product is launched that is based on the technique, those amounts go even higher.’
The announcement of the free CRISPR licences by Van der Oost and WUR president Louise Fresco in *Nature* journal has already drawn questions and requests to send research material. ‘We are working on that now, drawing up a kind of basic contract in which each party can fill in further details,’ says Van der Oost. ‘We still have to invent that wheel a bit, because this has never been done before. I expect we shall publish an account of the nuts and bolts of it, such as the principles behind the free licences and the rules of the game on using our technology.’

ACADEMIC USE

For academic labs and fundamental research, CRISPR-Cas technology is not completely under lock and key. The Broad Institute, for example, which is related to Massachusetts Institute of Technology, grants access to patented CRISPR-Cas technology. Researchers can order a complete CRISPR-Cas system through Addgene. ‘For academic and non-profit research use, no written licence is necessary. For these communities we make CRISPR tools, knowledge, methods and other intellectual property for genome editing freely available for research,’ states the institute on its website
‘It’s true that other parties make this technology available as well, but everyone does it in a slightly different way,’ says the Wageningen patent expert Paul van Helvert. ‘We have had the occasional licence from the Broad Institute ourselves, and the limits on it are quite strict. They restrict your free

access to internal scientific research. Even if a research institute makes a modified wheat variety freely available on a non-profit basis, that can be seen as a commercial activity because the party that takes it further stands to make a profit. The licences we want to grant do offer the possibility of developing products and trading in them without us charging a fee. So as far as we can see, we really are going one step further with our licences than other institutions.'

A licencing agreement is a document of 15 to 20 pages, says Van Helvert, and a contract like that covers a range of rights and obligations. One of these is that the user should tell the patent holder what they do with the technology. 'Our licences offer several possibilities, but we are keen to monitor how and where they are applied. Agriculture knows no borders and crops can be sold to people in other countries. We want to make agreements on that, and keep an eye on it, since we may have granted commercial licences to institutes in other countries. There could be a conflict between these things.'

In the coming years, Wageningen could be taken to court because someone claims that another licence is being breached. Van Helvert: 'That doesn't mean you are in the wrong, but then legal enforcement comes into play. Then the issue is who has the greatest stamina, because those kinds of cases go on for years. They say success has many fathers, and that's true of technology too. It is a commercial game to some extent. The more successful a technology is, and the more you can earn from it, the sooner people go to court about it. My guess is that when this technology really takes off, not everyone will be really pleased that an institute in Wageningen is granting free licences.'

SHARING TECHNOLOGY

If a judge in that kind of lawsuit should find against Wageningen, non-profit research institutes could still get a bill for licencing fees from a third party. Van der Oost: 'We can't guarantee anything, and we do say that to the organizations who want to start working with our technology. But I do wonder whether that will actually happen, because any company that takes legal action puts itself in the media spotlight, and the attention they get is not positive. My main hope is that others will follow in our footsteps. What we want to achieve by doing this is to bring about a change in the way we share knowledge and technology. Hopefully other parties will do their bit towards that too.' ■

www.wur.eu/crispr-licenses

CUTTING WITH CRISPR-CAS

CRISPR-Cas is relatively easy to use. Researchers order something called a plasmid which contains the code for a DNA-cutting enzyme (Cas9) and a guide RNA (CRISPR). The guide RNA will lead the Cas9 to the right destination in the cell, which might be a gene that the researchers want to switch off. After injection, the cell translates the information on the plasmid into a working CRISPR-Cas complex that goes in search of a target. If the guide RNA comes across the right DNA sequence, the cutting is done. The cell quickly repairs the damage to the DNA, and this often gives rise to small mutations which cause the targeted gene to be switched off.

Researchers can also help the cell with the repairs by injecting a template, such as a short piece of DNA that serves as an example. This enables the cell to do the repair work while the researchers can introduce mutations they have thought up in advance.