Randall Platt: Hijacking the CRISPR system to create 'living diagnostics'

CREDIT: ETH Zurich

Randall Platt. More about his work at bsse.ethz.ch/platt

Randall Platt is the 2020 Eppendorf Young European Investigator. He is an assistant professor in both the Department of Biosystems Science and Engineering at ETH Zurich, and the Department of Chemistry at the University of Basel. Here he talks to science writer Geoff Marsh about his award-winning work on creating a 'molecular recorder'.

Interview also available as a podcast at: go.nature.com/eppendorf2020

What is a molecular recorder?

A molecular recorder is an emerging class of molecular technology. It's basically a collection of genes that, when put inside cells, endows them with a synthetic form of memory. A molecular recorder could, for example, take a biological signal — a chemical, a molecule or a transcription factor — and transduce that into changes in a DNA storage medium. Then, through sequencing, we can reconstruct that cellular memory and identify what the biological signal was.

What is its relationship to the CRISPR system?

Most people are familiar with CRISPR as gene editing technology, which relies on an enzyme called Cas9. The enzyme acts as molecular scissors to correct a genetic mutation and cure a disease, for instance. But CRISPR is a whole toolbox of proteins.

In nature, CRISPR serves as an adaptive immune system for microbes. In humans, the adaptive immune system remembers the pathogens it has encountered and provides resistance against future infections. In a bacterial cell, when it's infected with a virus, the CRISPR system responds using its army of proteins.

Some CRISPR proteins take a piece of the virus and insert it in the host genome, where it serves as a molecular memory of that invader; at a later point in time, the CRISPR system can recall that memory in the form of a small piece of RNA. This so-called 'guide RNA' directs CRISPR-associated enzymes back to destroy viral invaders by cutting them using Cas9, and thus provides the bacterium with immunity.

Our advance specifically focuses on that first step of the CRISPR system: the recording part, where a piece of the virus's genome is inserted into the bacteria's. We're leveraging that recording process to endow other cells with the ability to record RNA, which is a technique we call Record-seq.

How does Record-seq work?

In bacterial cells, the CRISPR proteins form a big multi-protein complex. They grab RNAs, proportional to their abundance, and take a little snippet of them — about 40 nucleotides — that they integrate in a CRISPR array. The array is simply a DNA sequence of repetitive records that are formed iteratively and sequentially. After multiple recordings, it's like a fossil record. The records are aligned topologically. So we're not only able to record whether a gene was expressed, but also when it was expressed.

How do these records provide information about the environment outside the cell?

We're quite fortunate in that evolution endowed bacteria with a plethora of sensors for a range of molecules. They are single celled organisms that need to survive in tremendously diverse settings. Compared to us, bacteria are masters of different environments. *Escherichia coli* for example, can live in the soil, in a culture tube in your lab, or inside your intestines. With this natural arsenal of sensors, it gives us a lot to work with. And as we already have a good understanding of *E. coli* gene expression, we should be able to predict what their outside environment looked like.

How might this technique be applied?

We think it can have immediate value in the area of 'living diagnostics'. For example, people over a certain age routinely have colonoscopies to check for colorectal cancer. This is an invasive procedure that involves insertion of an

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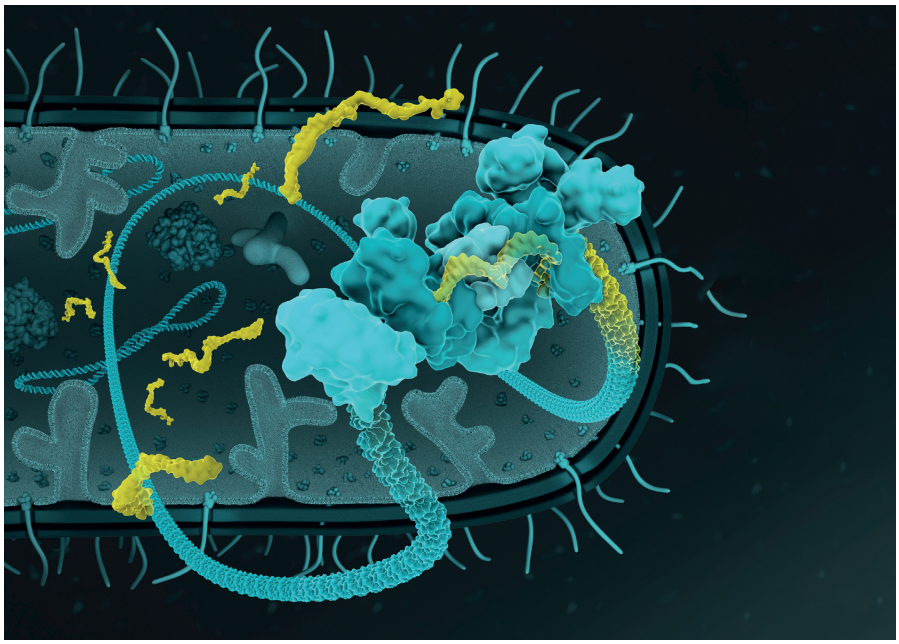

Record-seq works through acquisition of intracellular RNAs stored in a CRISPR array.

endoscope to look for small masses. Imagine an alternative example where a physician could instead give you a yoghurt containing molecular recording bacteria. You could drink this at home and send your stool sample into the lab. The cells could tell the clinician all sorts, from whether you have an early stage polyp to inflammatory bowel disease. Our goal is to evaluate to what extent these recording cells can be applied to disease pathology via the gastrointestinal tract.

We're already looking at mice on different diets and under different disease settings. For example, if we induce colitis (inflammation of the large intestine) we expect an environment rich in reactive oxygen species to develop. These molecules act on the *E. coli* in a specific way: upregulating the expression of protective factors. And when we look in our 'transcriptional records', as we call them, we do indeed find this stress response. We could use other bacteria with a different arsenal of sensors; there are huge open areas for future developments.

How close is Record-seq to the clinic?

With any radically new technology, the timeline between development and implementation is a long one, so this isn't happening tomorrow. It might not even happen in this decade. It depends on a number of factors, including how well the system works and how specific it is for a given disorder. Also, the regulatory path, to use genetically modified bacteria inside people, is unclear. So this is a big unknown. We still have a lot of development to do in the lab. The experiments in mice are incredibly promising but there are a number of engineering, quality control and clinical development steps that we need to do.

What other applications might there be for Record-seq?

Bacteria and other microbes cover every surface of this planet. If you embed them with recording devices, you could create sentinels of any environment. We're interested in the body, but you could potentially put this in bacteria in the soil, the ocean or the air, and monitor environmental contaminants.

What's next for you and your lab?

We will continue to develop Record-seq in the context of human health. My lab also develops other molecular technologies with different goals — understanding how genetic disorders arise and coming up with genomic-based treatments. We've created a few different technologies based on CRISPR enzymes that allow us to massively change genomes and transcriptomes towards correcting complex genetic disorders. We're also developing other techniques in functional genomics to increase the scale at which we perturb and understand gene function.