Systems analysis of cell-cell communication in plant roots (RootBook)

Spatial single-cell expression map of the plant root

The team from the ERASysAPP project RootBook has not only set out to investigate cell-cell communication in the plant root tip. The scientists also want to find out what role intercellular communication plays under stress conditions. Innovative technologies and methods, as well as the international composition of the research team, constitute the ideal conditions for this endeavor.

A technical innovation stands at the center of the international RootBook project. At first glance, it looks like an inconspicuous transparent plastic block, whose interior is covered in a network of fine, geometrically arranged lines. Upon closer inspection, these lines are revealed to be channels, through which any medium can be made to flow thanks to an ingenious valve system. "This type of apparatus is known as a microfluidic device. Their use is becoming increasingly popular in biological research for the observation of miniaturized biological systems," explains project coordinator Manfred Claassen. "Based on this, our project partner Matthias Meier has developed the 'RootChip'", he adds. Its special feature is the inclusion of tiny drill holes, allowing the researchers to plant seedlings in the plastic block. Their roots then grow along the holes into the nutrient-filled channel system below. The transparent material of the block allows this process to be microscopically monitored.

Communication between root cells

Whereas the first version of the RootChip (see picture) had the capacity to grow eight seedlings, the team of researchers is now working on a development that would allow the simultaneous cultivation of 48 plants. Claassen succinctly summarizes the main advantage of the new RootChip: "This device will allow us to measure root development in a spatial context and determine the molecular composition of individual cells." Here, the scientists are particularly interested in how the cells of the root tip communicate with each other to coordinate root growth.

Although some of the genes and mechanisms involved in this intercellular communication are already known, the RootBook team wants to be able to characterize the involved activities and correlations between individual molecular structures comprehensively and in a spatially accurate way. To this end, the scientists are making use of yet another innovative technique.

Painting an expression map with 64 colors

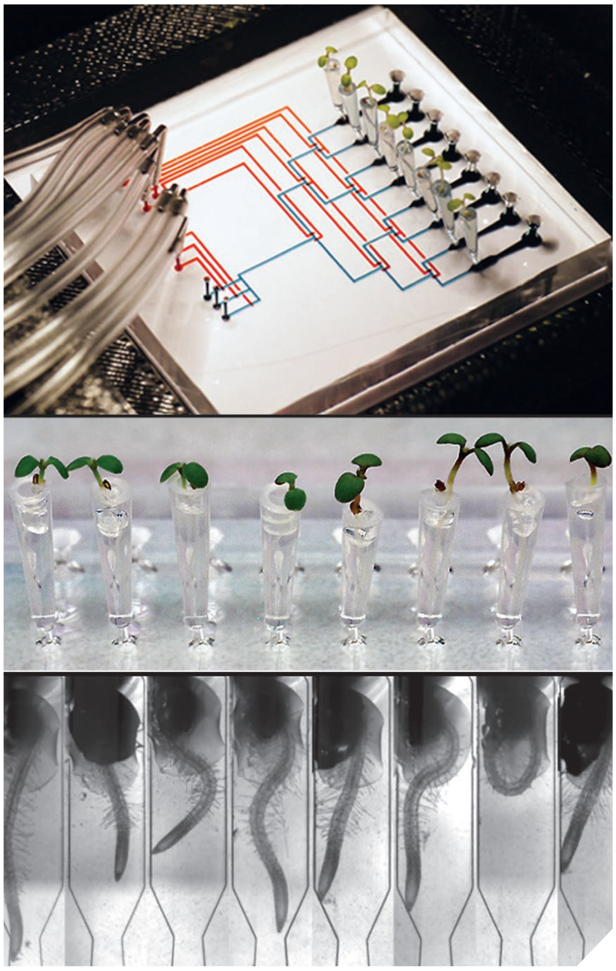
"In the first step, we perforate the cells of the root tip," explains Claassen. This is an intervention that requires extreme caution and precision, as the holes must be just big enough so that no molecular material leaks out of the cells. Then, colored dyes that bind specifically to previously selected protein structures are injected into the prepared cells. "In this way, we're able to mark up to 64 different mRNAs, each with a different color," says Claassen. Since an mRNA is the transcript of a particular gene and its immediate surroundings on the DNA, its concentration correlates with the activity level of the corresponding gene.

"After staining the cells, we obtain an individual picture for each cell with different numbers of colored spots in 64 different shades. These are counted using fluorescence microscopy, and from this we can ascertain which genes are expressed, and how strongly, in a particular cell at a particular point in time," explains the scientist. Although the pictures taken using the RootChip are two-dimensional, they can be extrapolated into the third dimension with

A

B

C



A The RootChip mounted with eight live plants.
B Top view of the eight plants in transparent tubes filled with agar and mounted onto the chip.
C Side view of the microchannels containing seedling roots 7 days after germination.
Illustration: Matthias Meier

Matthias Scholer

the right computational and mathematical analysis. Using this method, the researchers can create what is known as a three-dimensional, single-cell expression map of the entire root tip, on which the measurements from each individual cell are recorded spatially accurately. “These transcription patterns will hopefully allow us to build mathematical models and understand the cell-cell communication in detail,” summarizes Manfred Claassen.

Behavior under stress conditions

If this step is successful, the RootBook team would like to examine the communication behavior under stress conditions. The growth of roots is heavily dependent on the nutrient content of the soil. “Changes in phosphate levels or salt concentration cause abiotic stress in plants, potentially resulting in the withering of the roots and subsequently the whole plant,” describes the project coordinator.

The third member of the RootBook team, plant molecular geneticist Reidunn Aalen, will take advantage of the RootChip to selectively expose the plants to differing stress conditions and systematically measure the resulting effects on intercellular communication. Claassen contemplates future applications of their findings: “This might lead us to novel solutions for maintaining plant growth even under adverse environmental conditions,” he muses.

International cooperation a great opportunity

RootBook is still very much in the conception stage, the project having only been launched at the beginning of the year. The whole team is highly motivated. “RootBook is a very exciting project. We’re investigating a fascinating, cell-transcending biological phenomenon, enabled by the innovative integration of microfluidic technologies and customized computational biology solutions,” says Manfred Claassen. The project coordinator is confident that the project will meet its ambitious goals, not least thanks to the international composition of the consortium. “A transnational collaboration has the great advantage that the involved scientists contribute to the diversity in research culture.” This results in correspondingly complementary approaches being brought together to innovatively address the research questions, which ultimately also contributes to uncovering the complex relationships of intercellular communication in the root tip.

More information on RootBook:
www.erasysapp.eu > Calls > 2nd Call > RootBook

Manfred Claassen coordinates the international RootBook project.

RootBook at a glance

Research groups:

- Prof. Manfred Claassen (project coordinator), Institute of Molecular Systems Biology, ETH Zurich – Computational biology
- Dr. Matthias Meier, Department of Microsystems Engineering, University of Freiburg, Germany – Biotechnology
- Dr. Reidunn Aalen, Department of Biosciences, University of Oslo, Norway – Plant biology

Total budget (2016–2018): EUR 1 million, including EUR 215,500 from SystemsX.ch

Project type: International Project – As a partner in the European research network ERASysAPP, SystemsX.ch has co-funded six international application-oriented projects in which Swiss consortium partners are involved.

Further reading: Grossmann G. et al. (2011) The RootChip: An Integrated Microfluidic Chip for Plant Science. The Plant Cell 23(12):4234-4240.