



The MecanX team performs research on plant cell growth. (From left) back row: Tohnyui Ndinyanka Fabrice (IPMB, UZH), Hannes Vogler (IPMB, UZH), Chengzhi Hu (IRIS, ETHZ), Naveen Shamsudhin (IRIS, ETHZ) und Ueli Grossniklaus (IPMB, UZH); front row: Jan Burri (IRIS, ETHZ) und Gorka Santos Fernandez (IPMB, UZH)

Understanding the physics of plant growth (MecanX)

Growing under high pressure

Plant cells are surrounded by a stiff casing that withstands tremendous pressure from the cell's interior. According to current understanding of the subject, such entities should not be capable of growth, and yet grow they do. Scientists are trying to uncover their secrets in order to solve this enigma, which may help develop better crops for the future.

Every spring, grasses push their way up out of the ground, foliage sprouts from flowerbeds and buds come into bloom to form a collage of color, all within the space of a few days. In order to manage this rapid growth, plants employ a trick. In the previous year, they assemble many of the cells that go on to form the blossom, leaf or stem in miniature. When spring comes, all they have to do is enlarge these cells. But how the cells manage this without bursting or collapsing still remains mysterious.

Ueli Grossniklaus, professor in the Department of Plant and Microbial Biology at the University of Zurich, is trying to unravel these mysteries together with a highly interdisciplinary team of collaborators. As a start, he wants to measure the stiffness of the cell wall. This is the first step in an extensive research project with the aim of conclusively understanding the physics of plant growth. "There are so many theories floating about, but we don't have much concrete knowledge on the subject," says Grossniklaus.

A green cage

A plant cell's rigid cell wall can be likened to a cardboard box. Such structures are very sturdy, although when subject to external forces, they collapse easily. To counteract this, a plant cell has a large vacuole. This is like a big water balloon that fills the cell's interior, exerting high pressure – up to 10 bars – on the cell wall. This construction provides the plant cell with stability. "With this building system, plants are able to grow huge structures. For example, woodless tropical ferns can grow up to several meters long and are still very stable," says Grossniklaus.

The drawback of this approach is that in order for a cell to expand, the cell wall must be weakened in one or more places so that it can stretch out and new material can fill the gaps. This can be likened to trying to enlarge a house by opening up its outer walls, pulling them apart and then pouring fresh concrete in to fill the holes.

High pressure

In theory, a plant cell would have to reduce the pressure from the inside to avoid the cell exploding as a result of the instabilities in its wall during expansion. Yet the pressure evidently stays high. If this were not the case, all the leaves in your garden would constantly hang limp.

In order to get to the bottom of this paradox, the MecanX team is performing various measurements on pollen tubes from lilies, which are about 16 micrometers in diameter, and also much smaller ones from *Arabidopsis*, which are only 5 micrometers in diameter. These tubes grow out of pollen grains that have landed on the flower's stigma. Each tube is made up of a single cell and grows to several centimeters in length.

Measuring the stiffness of the pollen tube cell wall requires an appropriately fine sensor, which the researchers first had to develop. In essence, this force sensor is a silicon probe with a tungsten tip, much finer than a human hair (see picture, page 7). The probe is lowered perpendicularly onto the pollen tube's surface, indenting it. The depth of the indentation depends on the cell wall's stiffness and the applied force.

Measurements via capacitor

To quantify this resilience, or stiffness, the researchers attached the probe to a tiny capacitor. The capacitor is etched from a silicon wafer, a common process in computer chip production.

Capacitors store energy by separating charge, a bit like a battery. They are made up of two conducting surfaces separated by some distance. When the distance between these two plates decreases, the charge on the capacitor increases, and these changes in capacitance are measurable.

When the probe meets resilience from the cell wall, the resistive force causes the plates in the capacitor to be pushed closer together. The resulting change in capacitance is a measure of the force applied and, along with the indentation depth, is used to calculate the stiffness of the cell wall by way of a computer model. The model is based on the finite element method often used in mechanical engineering, and is simultaneously used to calculate the turgor pressure within the cell.

Silicone channels

That's in theory. In practice, measuring the stiffness of the cell wall poses still another technical challenge. Pollen tubes are somewhat difficult to work with. For example, when pollen grains are placed on a microscope slide, they germinate and the pollen tubes grow messily in all directions. It's not an ideal set-up for taking a large number of precise measurements.

The MekanX team had to find a way of restoring order to this chaos. The solution is a sort of micro-canal system for pollen tubes, a structure a bit like a computer chip and only a few millimeters across. It is, however, made of the optically transparent silicone polydimethylsiloxane (PDMS).

On this PDMS chip there is a row of channels emanating from a central basin. This is where the researcher places a few dozen pollen grains. After germination, the pollen tubes have no choice but to grow through the channels, where they can be easily probed and measured.

The team has developed an efficient way of producing these chips. First, they etch the negative of the chip into a silicon wafer.

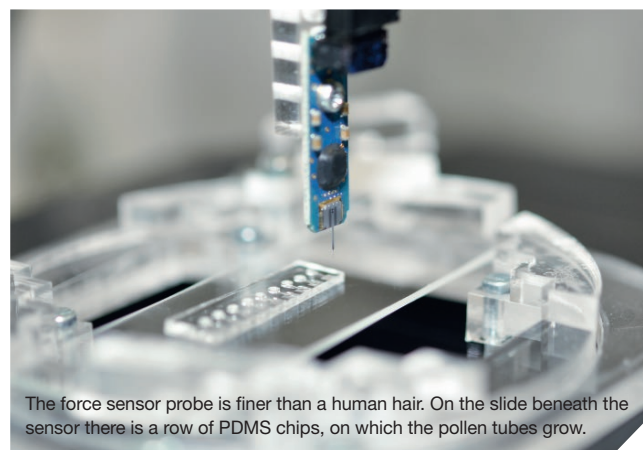
Liquid PDMS is then poured over this negative, and later the hardened chip is simply cut out and removed with tweezers.

Better crops

Initial results show that the pressure within lily pollen tubes amounts to about 3 bars, which is in agreement with measurements from other researchers using invasive methods. However, the stiffness of the tube casing has been measured to be roughly equal to that of rubber. A surprise, since it was previously thought to be much stiffer.

In order to solve the puzzle of plant cell growth, the MekanX team is turning to the tip of the pollen tube for answers. Until now, there have been no reliable measurements made at this location. Yet it is the most dynamic part of the pollen tube, since it is here that forward growth takes place.

The results might one day influence the development of new crop plants with improved cell wall properties. Even today there are forage maize varieties whose cell walls have a reduced lignin content. This makes them less stable, but at the same time makes the product much easier for cows to digest, leading to a 20 percent increase in milk production.



The force sensor probe is finer than a human hair. On the slide beneath the sensor there is a row of PDMS chips, on which the pollen tubes grow.

MekanX at a glance

Principal investigator: Prof. Ueli Grossniklaus

Research groups:

- Prof. Ueli Grossniklaus, Department of Plant and Microbial Biology (IPMB), University of Zurich – Biology, growth mechanics and physiology of pollen tubes
- Prof. Christoph Ringli, Department of Plant and Microbial Biology (IPMB), University of Zurich – Biology, biochemical composition of pollen tube cell walls
- Prof. Bradley Nelson, Institute of Robotics and Intelligent Systems (IRIS), ETH Zurich – Engineering, real-time cellular force microscopy, force sensors, control software, lab-on-a-chip devices
- Prof. Hans Jürgen Herrmann, Institute for Building Materials, ETH Zurich – Engineering, mathematical modeling of pollen tubes
- Dr. Abu Sebastian, IBM Research – Zurich – Engineering, applications of atomic force microscopy to pollen tubes, lab-on-a-chip devices
- Dr. Felix Beyeler, FemtoTools AG – Engineering, 2-D force sensors

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