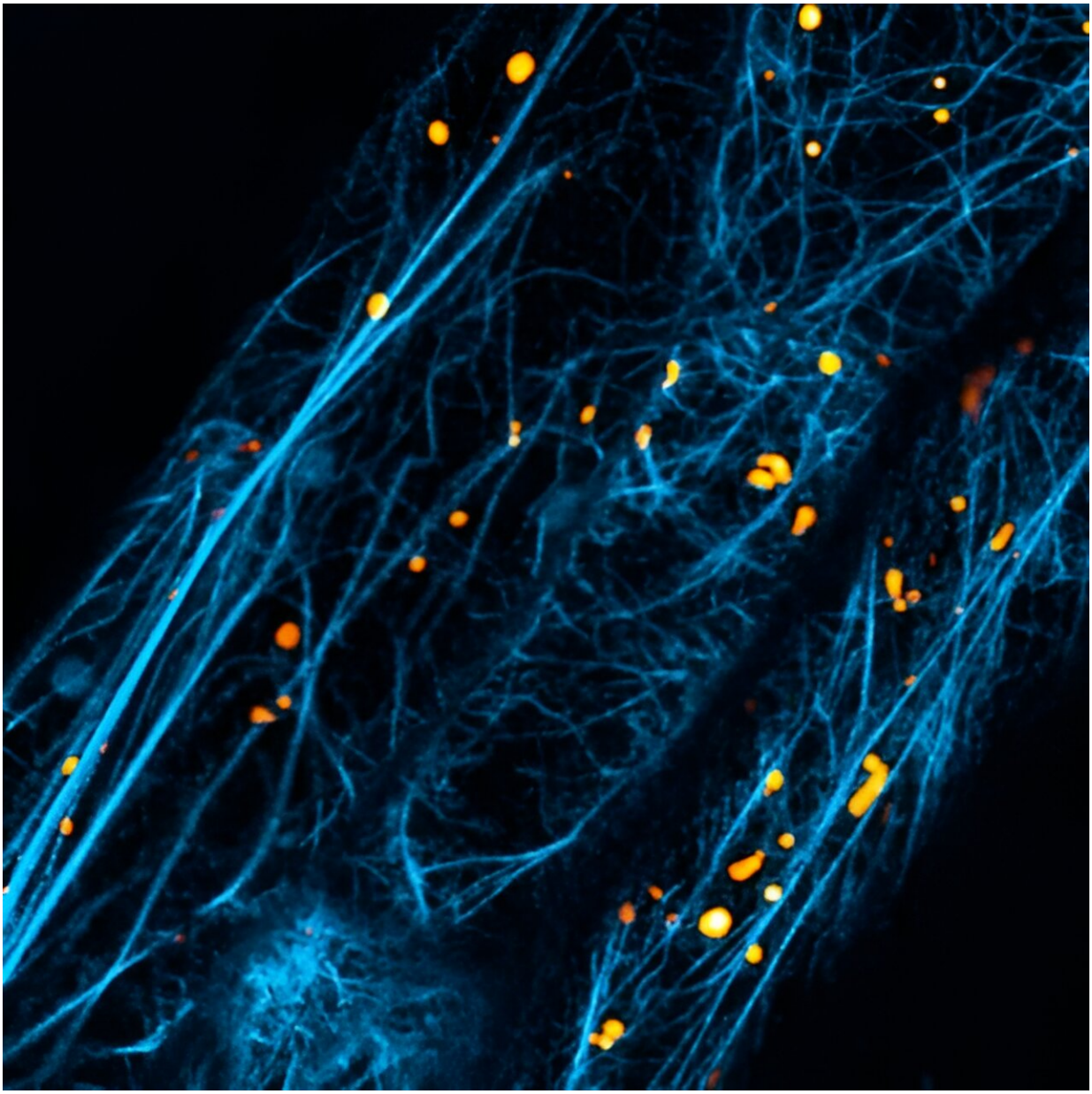
A protein that helps plant cells 'eat themselves'

October 25 2023



The actin cytoskeleton (blue) forms a dense network in plant cells that has many different functions. For example, peroxisomes (orange), like other organelles, use the molecular motors of myosins to move along actin filaments like a cellular "rail network". Credit: *Nature Plants* (2023). DOI: 10.1038/s41477-023-01542-6

Scientists from the Department of Experimental Plant Biology at the Faculty of Science of Charles University have published research in *Nature Plants* that brings new insights into the function of the ARP2/3 protein complex in plant cells. It turns out that plant cells can use this component in a very different way than animal cells—it is important for cellular recycling of peroxisomes.

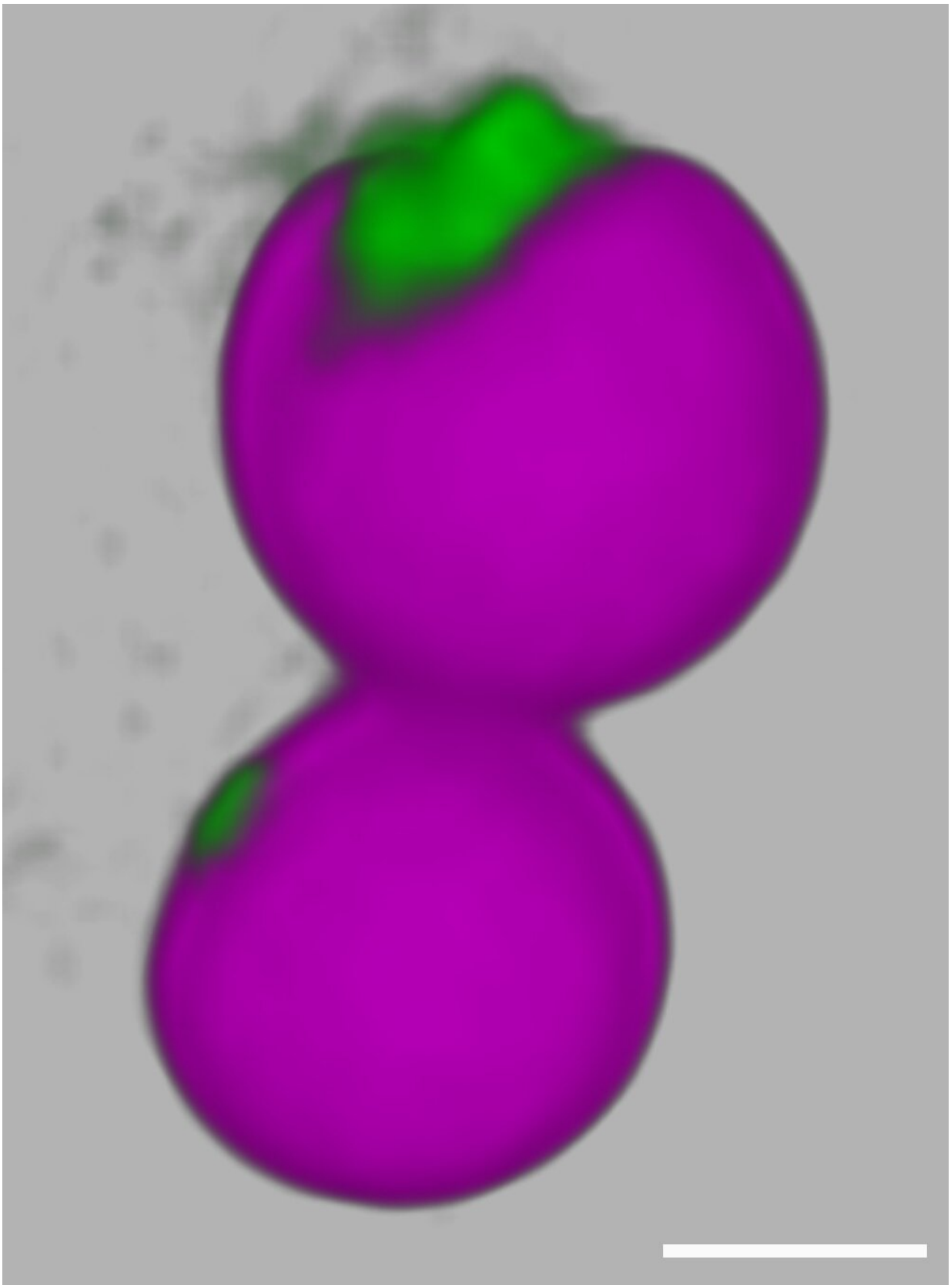
The protein complex under study consists of seven subunits and is important for the formation of the "cell scaffold", the actin cytoskeleton. In animal cells, ARP2/3 is responsible, for example, for the formation of membrane protrusions—lamellipodia—and is therefore essential for cell movement.

Plant cells, however, are surrounded by a rigid cell wall and therefore cannot form membrane protrusion; they are immobile and remain in the same place in the plant body. Nevertheless, we find an evolutionarily conserved ARP2/3 complex in plant cells. It is therefore clear that it plays a role in different processes than in animals.

Previous studies have shown that the ARP2/3 complex has a role in coordinating the growth and shaping of plant cells, but now scientists have discovered a completely new role. This role is related to a specific form of autophagy, called pexophagy. What do these words mean?

Autophagy (Greek for "self-feeding") is the process by which a cell removes old and damaged organelles and recycles them into essential

chemical components that it can then reuse. Pexophagy is the name given to this recycling of peroxisomes—organelles that play a role in the detoxification of oxygen radicals, but also in the metabolism of fatty acids.



3D visualization of peroxisome (magenta) and ARP2/3 complex (green). The image was obtained by fluorescence microscopy using an Airyscan detector. The peroxisome is highlighted in purple and the ARP2/3 complex in the epidermal cells of the thale cress plant in green. Scale 1 μm. Credit: Kateřina Malínská, Institute of Experimental Botany of the CAS.

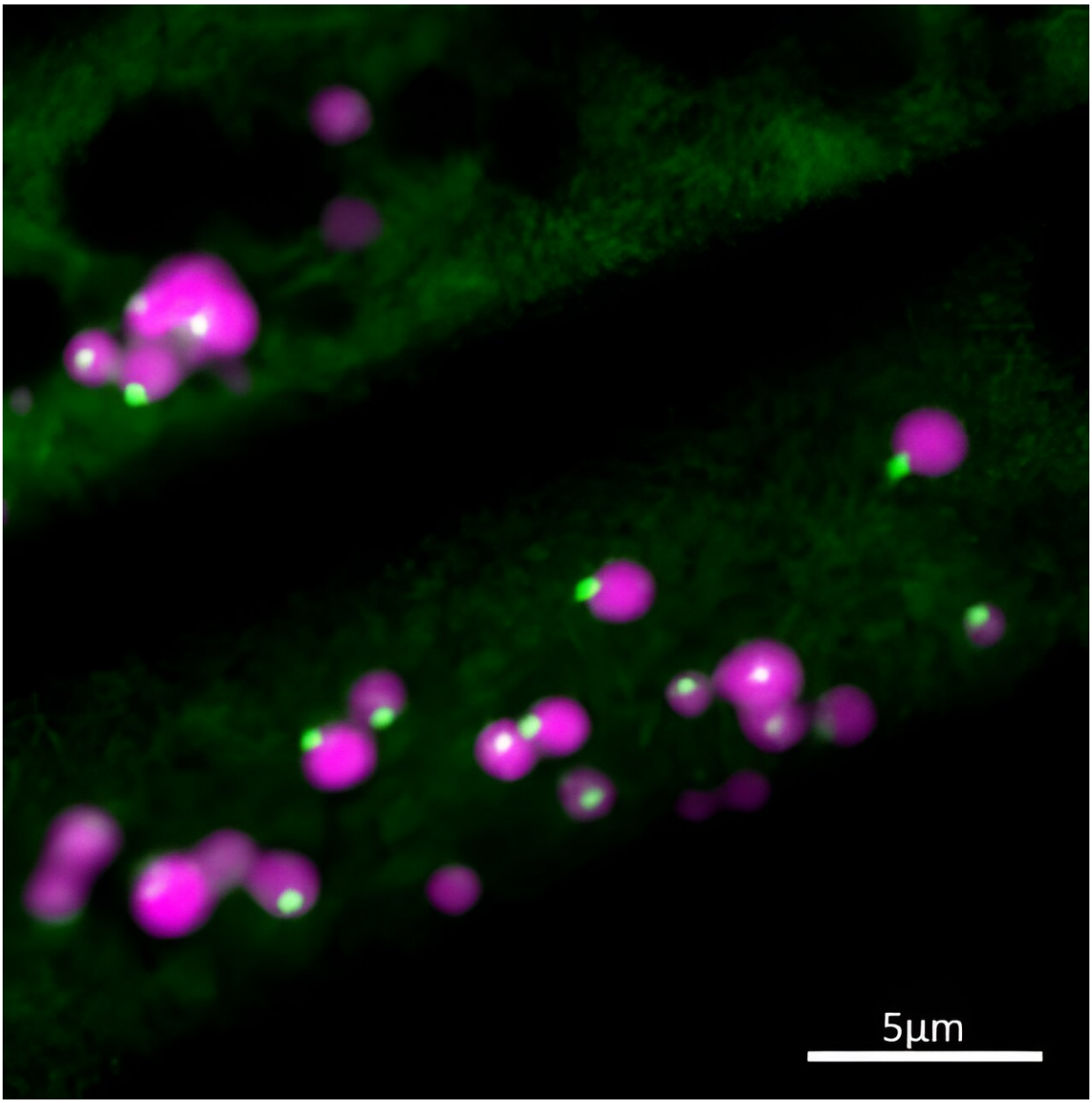
What does "recycling" look like in practice? If the peroxisome is already old and damaged, the cell encloses it in a double membrane, called an autophagosome. Thus wrapped, it moves it into a vacuole where it is dissolved in an acidic environment by its "digestive" enzymes.

It is the ARP2/3 complex that plays a role in the process of peroxisome envelopment by the autophagosome membrane. Scientists have shown that in mutants that have the ARP2/3 complex non-functional (lacking any of its seven subunits) or inactive (lacking its activator), peroxisomes accumulate in the cells because the cell is unable to degrade them properly. They also used confocal microscopy to show how the ARP2/3 complex forms a domain on the peroxisome at the very point where the autophagosome sits to engulf the peroxisome.

This discovery, which also involved scientists from the Institute of Experimental Botany of the CAS and scientists from the UK, not only expands our understanding of how the ARP2/3 complex and autophagy function in plant cells, but also reveals how different groups of organisms can use the ARP2/3 complex for their different needs.

For example, whereas in animals membrane remodeling by the ARP2/3 complex is important for movement, in immobile plant cells it has found a role in membrane remodeling during the process of autophagy. This shows the amazing ability of nature to use existing tools that have arisen during evolution for different purposes.

During the research, the team of Kateřina Schwarzerová, Ph.D. used several advanced microscopy methods. Due to the very small size of the peroxisomes, which are about 1 micrometer in size, and the speed at which peroxisomes move in plant cells, a special microscope with TIRF imaging mode was used to observe the ARP2/3 complex on peroxisomes, which allows to observe very fast processes in living cells.



ARP2/3 complex on peroxisomes in epidermal cells of Arabidopsis thaliana plants. To visualize the structures, they are labeled with fluorescent proteins. These serve as markers to allow detection of the structures in living cells. In the confocal microscope image, the ARP2/3 complex is highlighted in green and the peroxisomes in purple. Credit: Jan Martinek, Department of Experimental Plant Biology, Charles University.

In collaboration with Kateřina Malínská, Ph.D. from the Microscopy Department of the Institute of Experimental Botany of the CAS, v.v.i., the scientists also used a microscope with an Airyscan detector, which allows better resolution than is achievable with conventional light microscopy.

"When we first saw the dots that the ARP2/3 complex makes in cells under the microscope, we expected them to mark the branching sites of the actin cytoskeleton. So we were surprised when a colocalization study showed that they were actually associated with peroxisomes. This was quite different from what we know about the localization of the complex in animals, for example. At the same time, it did not make sense given the known function of the ARP2/3 complex in shaping plant cells."

"It was only after several years of detective work that we were able to discover that this surprising localization of the ARP2/3 complex on peroxisomes was necessary for its completely new function in peroxisome degradation by autophagy", says Jan Martinek, first author of the publication in *Nature Plants*.

More information: Jan Martinek et al, ARP2/3 complex associates with peroxisomes to participate in pexophagy in plants, *Nature Plants* (2023). DOI: 10.1038/s41477-023-01542-6

Provided by Charles University