Examining molecular components that are responsible for generating the action potential in the Venus flytrap

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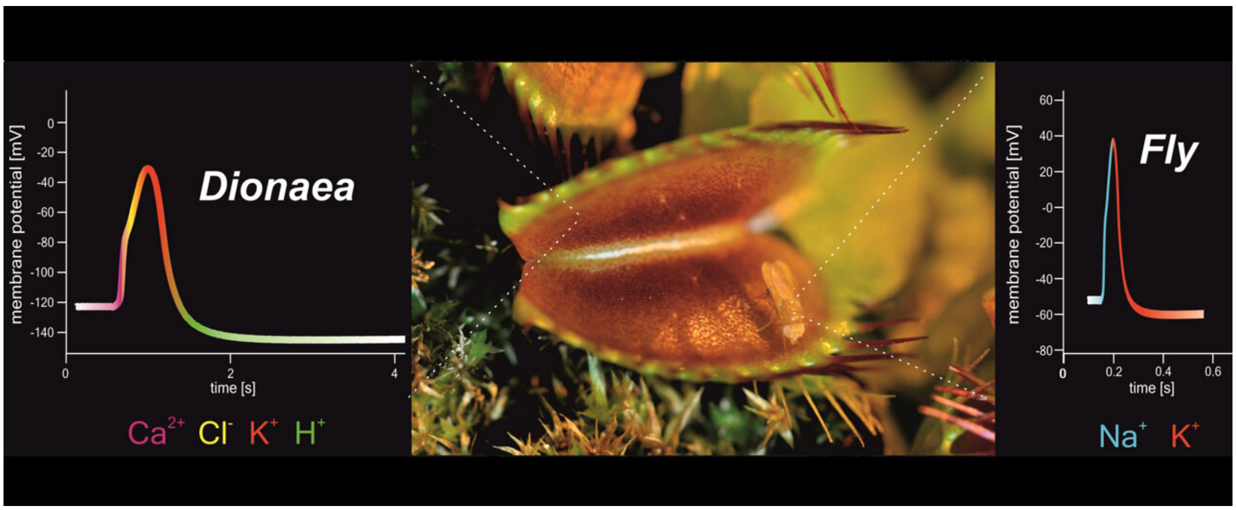

A Venus flytrap with its prey (center): When the fly comes close to the sensory hairs, an action potential (AP) is triggered in the Venus flytrap (left). Compared to the animal AP of the fly (right), distinctly different ion fluxes are involved in the different APs. Credit: Sönke Scherzer/Uni Würzburg

To hunt flies and other small animals, the Venus flytrap has to be faster than its prey. To do so, it has developed a catching organ that can snap shut in a fraction of a second and is controlled by the fastest signaling networks known in plants. An electrical signal known as the action potential is at the heart of this network. When a fly touches one of the six sensory hairs of the Venus flytrap, an action potential is generated, arming the snap trap. A second action potential finally triggers the capture organ.

The fact that electrical signals enable the Venus flytrap to catch prey has been known for more than 150 years. A team led by Professor Rainer Hedrich, a biophysicist at Julius Maximilians University (JMU) Würzburg, Germany, has now investigated the molecular components that are responsible for generating the action potential—an issue that was previously unexplored. In the current issue of the journal *Current Biology*, the scientists now present the results of their work. Their focus is on glutamate receptor channels and ion transport proteins that initiate action potential and keep it going.

When the Venus flytrap becomes electrically excitable

A fundamental question for the team was at what point in its development the Venus flytrap's capture organ becomes electrically excitable in the first place. The answer was given by first author Sönke Scherzer: "Only when the trap is fully developed and opens for the first time it fires its archetypal action potentials."

An action potential manifests as a transient deflection of a cell's membrane potential—the electrical voltage between the inside and outside of the cell. During an action potential the membrane potential typically drops rapidly during depolarization, only to rise again during the subsequent repolarization, initially above the original resting value before slowly approaching its original value again. The Venus flytrap's action potential usually lasts only one to two seconds and propagates as wave.

For communication within the cell as well as between cells, tissues and organs, plants additionally use calcium waves, which are mediated by positively charged Ca^{2+} ions, serving as secondary messengers. "Using flytraps that carried the gene for a calcium ion reporter protein, we were able to show that action potentials and calcium signals not only operate in a coordinated manner, but also propagate at the same speed," explains Rainer Hedrich.

Surprising discovery in the genetic makeup

With the expertise of Ines Kreuzer and Anda Iosip the team then identified the genes that encode this signaling pathway. "The Venus flytrap needs less than half a day to open its trapping organ for the first time," says Kreuzer. "We therefore looked at those genes that are differentially expressed when the trap enters its excitable stage."

Among the most highly expressed genes the Würzburg team pinpointed a glutamate receptor channel—a surprising observation, says co-author Manfred Heckmann, chair of physiology with a focus on neurophysiology at JMU. "Glutamate functions as a neurotransmitter in humans. If the plant channels actually function as glutamate receptor channels, too, stimulation with glutamate must trigger a calcium ion signal and an action potential," says Heckmann.

From gene expression profiles to the AP model

The new insights gained by the Würzburg research team allow only one conclusion: The influx of calcium ions initiates the action potential via the glutamate receptor channel. The question remains: How does the action potential pick up speed?

Upon closer examination of the genes, an anion channel, a potassium channel, and a proton pump caught the team's eye as potential actors in this process. With the help of Professor Ingo Dreyer, a former JMU fellow, now working as a biophysics bioinformatician at the University of Calca in Chile, they were able to describe the process in detail.

Accordingly, calcium ions that enter the trap cells via glutamate receptor channels represent the igniter. As second messengers, they initiate the opening of the anion channels. Anion efflux results in membrane potential depolarization. Depolarization in turn opens potassium ion channels, initiating the repolarization phase via potassium efflux. As repolarization progresses, the proton pump takes over to return the process to its initial state.

The complex action potential of the Venus flytrap

So compared to its victims, the action potential of the Venus flytrap is by far more complex. "While the action potential of humans and flies is based on only one sodium and one potassium channel, the Venus flytrap possesses two additional components," explains Rainer Hedrich.

Thus, a relative of the fly's potassium channel, together with the proton pump, warrants repolarization of the action potential in the fly trap. Sodium channels play no role in this process in plants. Instead, depolarization of the flytrap action potential is achieved by the concerted action of a glutamate receptor calcium channel and a calcium dependent anion channel.

Outlook and future research

Plants genomes encode for about 20 glutamate receptor channels but do not have synapses. What does the plant need so many receptors for? Where does the glutamate come from during stimulation and how is it maintained in the resting state? Hedrich's team plans to address these questions in upcoming studies. "We will soon be able to clarify this with the help of genetically encoded glutamate sensors in plants," Hedrich says.

"With regard to the structure, function and regulation of glutamate receptor channels and glutamate transporters, we currently have more questions than answers. It is possible that evolution is showing us the way here. In very early land plants, we find species with only one glutamate receptor channel. The question is whether there is a connection between the evolution of these channels and the excitability of plants. That's what we are determined to find out."

More information: Sönke Scherzer et al, A unique inventory of ion transporters poises the Venus flytrap to fast-propagating action potentials and calcium waves, *Current Biology* (2022). DOI: 10.1016/j.cub.2022.08.051

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