Shaking *Arabidopsis thaliana*

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Potassium has a special role in plants because a high intracellular concentration is required to maintain turgor pressure, the force that keeps celery from becoming limp and lettuce from wilting. In searching for the protein that transports potassium into the cytoplasm of such cells, two groups of investigators have cloned genes from the plant *Arabidopsis thaliana* that are unlike any known ion pumps or carriers. In fact, the two genes encode proteins with a surprising resemblance to a family of potassium channels found in animal nerve cells—the *Shaker* potassium channels from *Drosophila melanogaster*.

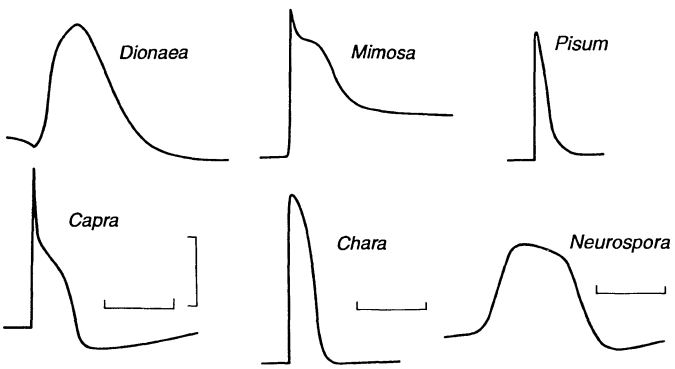
Plant and fungal cells accumulate potassium to high concentrations. This steep gradient of potassium across the cell membrane is indirectly maintained by a proton pump [the proton adenosine triphosphatase (H^+-ATPase)], which generates a large electrochemical gradient (negative inside the cell). This allows positively charged ions to flow passively inward because the interior of the cell is maintained at −160 to −240 mV. Unlike animal cells, plants and microbes can survive in media with little nutritional value. This ability may be explained by their rather substantial resting potential and their reliance on ever-present protons rather than sodium, the ion pumped by the analogous animal ATPase.

To elucidate the molecular structure of potassium transporters in plant cells, H. Sentenac and co-workers and R. F. Gaber and co-workers used mutants of yeast that are defective in potassium accumulation. They report their results simultaneously in this week's issue of *Science* (*1*) and in the current issue of the *Proceedings of the National Academy of Science* (*2*). Wild-type yeast grows normally on media containing micromolar concentrations of potassium, but a yeast mutant that is defective in potassium uptake requires millimolar concentrations for normal growth. The mutant genes responsible for this defect have been cloned and sequenced and encode putative potassium transporters, one high-affinity (*TRK1*) and the other low-affinity (*TRK2*). *TRK1* and *TRK2* encode structurally related proteins with 12 putative membrane-spanning domains, but there is no sequence similarity with known ion channel proteins or other transporters in the GenBank database (*3*).

Gaber's group at Northwestern University and Sentenac and colleagues in Montpellier, France, inserted various cDNAs made from the green plant *A. thaliana* into these mutant yeast strains. Each group identified a cDNA that complements the mutant phenotype and allows these defective cells to grow. Although the two cDNA clones encode proteins of different size, 78,000 and 95,400 daltons, both of the predicted proteins contain sequence domains with structural features characteristic of the *Shaker* family of potassium channels. Like the *Shaker* protein, the clones contain putative membrane-spanning domains, of which one is a presumed "voltage-sensing" domain within the fourth transmembrane region and another is a highly conserved "pore-forming" domain between the fifth and sixth transmembrane regions. When the Gaber potassium channel gene is expressed in yeast, cell growth can be inhibited by the potassium channel blockers, tetraethylammonium and barium ions. Of particular interest is the fact that Sentenac's clone contains an additional hydrophilic carboxyl-terminal domain of six imperfect repeating sequences with homology to the 33-residue ankyrin repeat—a sequence found in animal cell proteins that serves as a protein-protein interaction surface. This ankyrin repeat–containing isoform may have unique functions requiring its interaction with the plant's cytoskeleton.

The existence of two isoforms of these plant proteins has precedence in the *A. thaliana* plasma membrane H^+-ATPase, which has ten isoforms (*4*). These pump isoforms may be differentially expressed in tissues with unique transport functions (*5*). Phloem cells, responsible for proton-coupled movement of sugar, and root hairs, which capture soil nutrients, appear to utilize different forms of the pump to accomplish their transport needs. Because proton and potassium fluxes are so tightly coupled in the plant cell, the newly cloned potassium transport proteins may also be distributed according to the particular function of the tissue. The expression of these proteins in yeast offers the exciting opportunity to use a facile genetic system for exploration of functional questions as well as for exhaustive site-directed mutagenesis.

Do these *A. thaliana* proteins that function as a potassium uptake system in yeast perform similar functions in their native environment, green plants? Perhaps in plants, as in yeast, channels can be responsible for the accumulation of potassium. Indeed, light-induced opening of stomata via an increase in the turgor of guard cells is caused by opening of an inward-rectifying potassium channel and flow of potassium into the cell (*6*). Patch-clamp electrophysiological experiments document the presence of many types of plasma membrane ion channels in plants and fungi (*7*). The rapid movements of touch-sensitive plants such as *Mimosa pudica* and the Venus flytrap are well-known examples in which electrical events direct behavior, but action potentials are also seen in other less animated plants and fungi (see figure). The molecular cloning of genes encoding other plant and fungal ion channels may point to functions and structures for these proteins that are unrecognized in other organisms.



Action potentials in plants, fungi, and algae—*Dionaea*, the carnivorous Venus flytrap; *Mimosa pudica*, a touch-sensitive plant; *Pisum*, the pea; *Chara*, an inanimate giant alga; *Neurospora*, a mycelial fungus—and an animal cell, goat (*Capra*) Purkinje fibers; 50 mV; 0.5 s (left), 5 s (middle), 50 s (right). Adapted with permission from C. L. Slayman, in *Biological Century*, R. Barlow, G. Weissman, J. Dowling, Eds. (Harvard University Press, Cambridge, MA, in press).

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