Capturing Fast Pollen Tube Growth on Camera, Researchers Pin Down Plant Fertilization Process

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(PhysOrg.com) -- Studying pollen tubes, University of Massachusetts Amherst plant cell biologist Peter Hepler and colleagues have captured some of the fastest growing tissues known, on camera for the first time, to advance understanding of fertilization processes critical to development of all fruits, nuts, grains, rice, corn, wheat and other crops we depend on for food.

Growing at a rate of around 2 to 3 nanometers per second, or about 50 times faster than animal nerve cells, pollen tube tips are believed to travel near the upper limit of cell growth. Each pollen tube carries two plant sperm through the pistil to fertilize the egg and its food supply, the endosperm. Why the hurry? Because, as Hepler explains, the first pollen tube to reach the egg delivers its DNA to the next generation and wins the natural selection race.

Hepler was inducted as a Fellow of the American Association for the Advancement of Science this month for his contributions as “one of the most influential plant cell biologists, who has continuously and continues to achieve breakthroughs that have guided research directions of numerous plant scientists.”

The current work, a culmination of years of investigation by Hepler and colleagues at UMass Amherst, with others at Long Island University, Worcester Polytechnic University, Aberystwyth University, Wales, UK, and nearby Hampshire College is reported in two recent articles in the journals *Plant Cell* and *Plant Physiology*. It was supported by the National Science Foundation.

Using two different imaging methods, fluorescent dye marking and interference optical contrast, in the first study Hepler and colleagues report they can now detect building-block pectins being packaged in vesicles inside the tube, which are then transported by an acto-myosin system—like muscles—toward the tip. There the vesicles fuse with the tip membrane and secrete their contents to build up the new cell wall.

Movies made by Hepler and colleagues reveal that as this new material thickens, it grows top-heavy, like a gathering ocean wave, elongating until it suddenly lurches forward a few micrometers. Growth continues like this in oscillations, or waves, of thickening, buildup and lurching forward until the tube reaches its goal and can deliver the sperm inside to its destination. The plant physiologists often work with lily or tobacco pollen tubes to watch their growing tips “dash” for the egg.

“We’re gradually piecing together how this works and it’s very exciting because this oscillatory growth pattern in pollen tube tips has never been observed directly before. It’s now possible for us to understand new details about secretion of new cell wall materials from the internal vesicles,” Hepler observes.

Each of the two observation methods they use has its own strengths. For example, interference contrast allows the researchers to see the edges of the tube tip as it grows, while the fluorescent-labeled pectin enzyme and propidium iodide method illuminates the thickest tube parts as the brightest spots. The fluorescently labeled pectin enzyme, pectin methyl esterase, is particularly interesting because it is essential for regulating pectin structure and thus cell wall strength.

Overall, success in observing the minutest details of the growing tip’s cell wall weakening and thickening in predictable oscillations serves to revive interest in the role of turgor pressure, a biophysical property of plants examined in the

1960s by Jim Lockhart, also a UMass Amherst plant physiologist. The Lockhart equation established the relationship between the restraint or yielding of the cell wall against the outward forces of turgor pressure. New studies by Hepler and colleagues provide support for the idea that the pectin deposits are forced into the wall matrix by turgor pressure. Through this process, called intussusception, the newly inserted pectin molecules weaken the existing wall structure and allow the tip to extend and the pollen tube to grow.

As Hepler explains his team's breakthrough, "We think our observations support the idea that cell wall yielding depends upon insertion of new building materials, the pectins, into the existing matrix, which is enough to weaken existing cell wall bonds and allow it to stretch but not break. It's like a controlled dam break until the tip reaches the egg. In other words, it's going to break and is designed to break, but not too soon."

Their second recent study, led by post-doctoral fellow Caleb Rounds, explored the energy requirements for oscillatory pollen tube growth, which is not limited to using familiar oxidative metabolism. Rather, Rounds showed, by treating pollen tubes with growth inhibitors including cyanide, oligomycin and antimycin-A, that pollen tubes can withstand such toxins and after an adjustment period will begin to grow again but at a slower rate while still exhibiting growth oscillations described above.

Further the inhibited pollen tubes begin producing ethanol. Hepler says, "In brief, they have shifted from oxidative phosphorylation to aerobic fermentation as a way, albeit inefficient, to produce energy for growth."

More information:
www.plantcell.org/cgi/content/abstract/21/10/3026

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