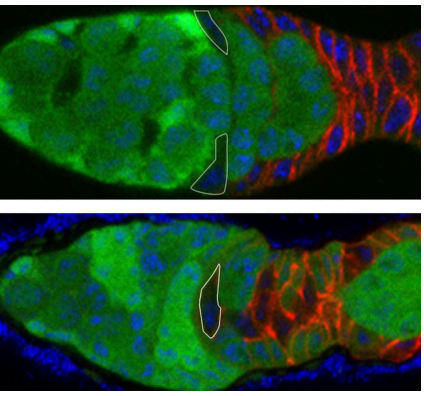
In This Issue



Follicle stem cells (outline) in a normal fly (top) and in one with mutant integrin (bottom).

Stem cells stand up for themselves

Adult stem cells are not pampered pushovers. O'Reilly et al. report that certain stem cells take charge of their surroundings, molding their environment to control their division and differentiation.

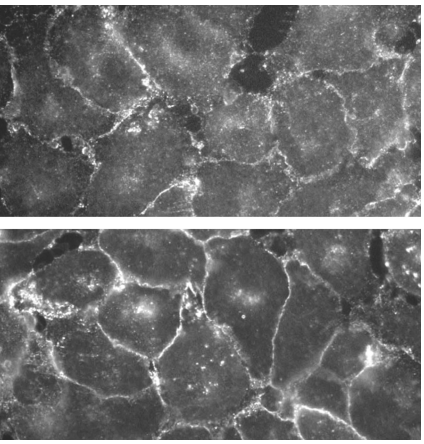
Some stem cells are cosseted like newborns. Neighboring cells cradle them in a structure called the niche. The niche not only nurtures its charges, it also dictates their behavior, determining whether they reproduce and specialize. The standard view is that the niche shapes stem cells, not vice versa.

O'Reilly et al. found evidence for more active stem cells while studying how the cells anchor themselves in the *Drosophila* ovary. Previous work indicated that ovary stem cells attach to the niche through the protein E-cadherin. O'Reilly et al. tested whether the stem cells also depend on integrins, cell surface proteins that link molecules in the extracellular matrix to the cytoskeleton.

They found that follicle stem cells (FSC)—one type of ovary stem cell—drifted away from their niche when they carried mutant integrins. These breakaway cells were abnormally shaped, divided more slowly than normal, and displayed some cancer-like characteristics, such as refusing to stop crawling even after contacting another cell.

Integrins hook onto an extracellular matrix protein called laminin A. FSCs pump out laminin A, and the scientists found that mutant cells that were unable to make the protein broke their moorings and reproduced sluggishly. Two other kinds of stem cells in the ovary—germline stem cells, which spawn the egg, and escort stem cells, which travel along with it—didn't rely on integrins for anchoring, the researchers showed. The team concluded that the interaction between laminin A and integrins ensures that FSCs remain in place, primed to divide. By laying down laminin A, therefore, FSCs help build their own niche.

O'Reilly, A.M., et al. 2008. *J. Cell Biol.* doi:10.1083/jcb.200710141.



The brighter margins of the cells at the bottom indicate these cells are being stretched.

Feeling the pull

Chiu et al. reveal the first step in the pathway that allows endothelial cells to determine when they are under mechanical stress.

Cells throughout the body are constantly being bent, tugged, pushed, and twisted. With each beat of the heart, for instance, cells that line blood vessels stretch and are exposed to shear stress. Such deformations prod a plasma membrane protein called PECAM-1, which latches endothelial cells together. In turn, PECAM-1 forwards the signal to other proteins in the cell. Scientists knew that the activation of PECAM-1 involves its phosphorylation. The mystery was what affixed the phosphate to the protein.

Researchers suspect that PECAM-1 is part of a multi-protein sensor that clings to the cytoskeleton. To search for PECAM-1's activator, Chiu et al. soaked endothelial cells in detergent to remove most of the membrane and the cytoplasmic proteins, leaving just the insoluble components, including the cytoskeleton. Stretching these bare-bones cells fired up PECAM-1, meaning that the activating enzyme remained. By adding different compounds that inhibit phosphate-attaching kinases, Chiu et al. narrowed down the list of enzymes to three candidates: Fyn, Src, and Yes. Blocking each with RNAi indicated that only Fyn sticks phosphates onto PECAM-1. Fyn might belong to the same sensing complex as PECAM-1, the researchers conclude. They now want to determine how mechanical stress jolts Fyn into action. One possibility is that it nudges Fyn closer to PECAM-1.

Chiu, Y.-J., et al. 2008. *J. Cell Biol.* doi:10.1083/jcb.200801062.

Young proteins cut down in their prime

"Last hired, first fired" is the rule when businesses are laying off workers. As Medicherla and Goldberg show, cells follow a similar strategy when weeding out damaged proteins, eliminating the freshly manufactured molecules.

Defective proteins, such as those induced by heat or reactive oxygen species, are a threat. Clumps of them build up in Parkinson's disease, Alzheimer's disease, and other illnesses. One way that cells get rid of faulty proteins is by feeding them into a molecular garbage disposal system called the ubiquitin–proteasome pathway. What researchers didn't know was whether certain types of proteins were more susceptible to damage and more likely to get trashed.

To find out, Medicherla and Goldberg warmed yeast cells to deform their proteins. In these cells protein breakdown surged, but surprisingly it tailed off after about an hour. The researchers determined that during this time the cells were demolishing newly made proteins, not established ones. The scientists

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saw similar discrimination against youthful proteins when they treated the cells to spur production of reactive oxygen species. Older proteins escaped destruction even though they incurred damage.

The results indicate that young proteins pass through a vulnerable stage that lasts around an hour. Folding into shape would probably take a few minutes at most, say the authors. But the proteins might also have to join with other proteins to form complexes, undergo structural modifications, or move to their home in the cell, during which time they are prone to damage and thus to destruction.

Medicherla, B., and A.L. Goldberg. 2008. *J. Cell Biol.* doi:10.1083/jcb.200803022.

How a cell puts itself on the menu

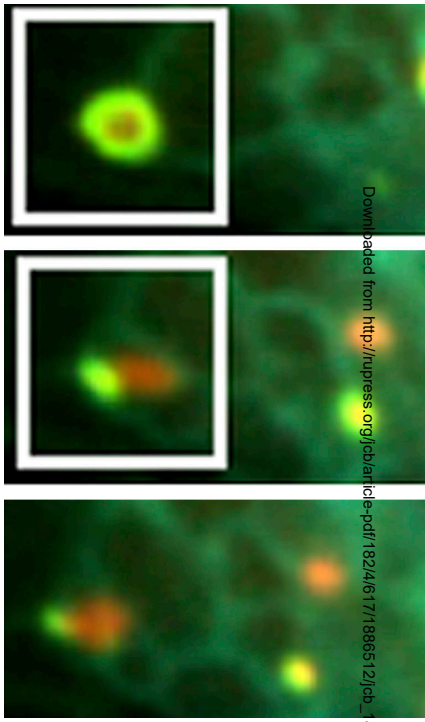
It could be starving or just cleaning up, but a cell sometimes devours a portion of its own cytoplasm, a process called autophagy. Two groups help clarify the workings of this form of self-eating. Axe et al. identify a cellular nursery for the membrane pouches that perform autophagy. Cao et al. reveal a method for pinning down the activity of autophagy control proteins.

During autophagy, a membrane container called an autophagosome scoops up some of the cell's contents, which the lysosome then digests. This cannibalism not only recycles nutrients for famished cells, it helps purge marred proteins. Too little or too much autophagy might cause illnesses such as Alzheimer's disease and certain types of cancer.

A stubborn mystery is where the autophagosome membrane comes from. Does it derive from the endoplasmic reticulum, as many scientists think? Or does it form when lipids in the cytoplasm rendezvous? Axe et al. attacked the question by tracking a phospholipid named PI(3)P that's essential for making an autophagosome. In starving cells a bud rich in PI(3)P bulged from the ER. This outgrowth, which the researchers dubbed an omegasome, spawned autophagosomes. The team observed new autophagosomes appearing inside the omegasome and then breaking free. Whether the omegasome separates from the ER or remains attached is unclear, the researchers say. But they conclude that the results support an ER origin for the autophagosome.

More than 20 proteins collaborate to orchestrate autophagy, but researchers are still trying to work out each protein's job. Instead of following the typical strategy of eliminating two or three of these proteins, Cao et al. went whole hog, deleting all 24 of the known yeast autophagy proteins. The team then added back different combinations of proteins to test hypotheses about their functions. For example, previous work suggested that the protein Atg17 is a pioneer that draws other proteins to the developing autophagosome. However, the researchers found that two other proteins are also necessary to instigate autophagosome formation.

Axe, E.L., et al. 2008. *J. Cell Biol.* doi:10.1083/jcb.200803137.
Cao, Y., et al. 2008. *J. Cell Biol.* doi:10.1083/jcb.200801035.



A newborn autophagosome (red) emerges from an omegasome (green circle).

Arp2/3 phosphorylation kickstarts cells

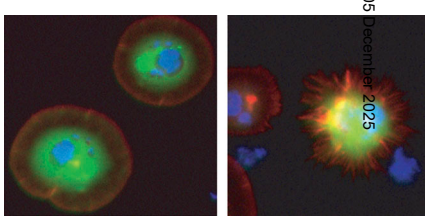
Crawl or stay put—it seems like a simple choice. But LeClaire et al. show that the decision is more complex than previously thought.

A cell slithers by extending a membrane protrusion, or lamellipodium, driven by newly assembled actin filaments. A seven-part protein complex called Arp2/3 controls the lengthening and branching of these filaments. Researchers thought that binding of a pair of so-called nucleation promoting factors, WASP and Scar, was sufficient to switch on Arp2/3. However, LeClaire et al. determined that phosphorylation of Arp2/3's subunits also was essential for the complex's activation—and thus for actin elongation and lamellipodia extension.

Arp2/3 works by attaching to the side of an existing actin filament and then spurring a branch to grow. To nail down which action phosphorylation controls, the team used several methods to dislodge phosphates. After treatment Arp2/3 could still grab hold of an actin fiber, but it couldn't perform capping, an essential step that prevents the newly formed actin filament from breaking apart. The cells also could not extend lamellipodia.

The work suggests that Arp2/3 serves as a command center, receiving input not just from WASP and Scar, but also from phosphate-adding enzymes. The researchers think that this complexity provides the cell with more precise control over its movement.

LeClaire, L.L., et al. 2008. *J. Cell Biol.* doi:10.1083/jcb.200802145.



In controls, lamellipodia spread out evenly around the cells (left). But cells lacking part of the Arp2/3 complex sprout abnormal, spiky extensions (right).