In This Issue

The sound of one foot walking

Cells often extend pseudopodia to migrate, but it has been difficult to characterize this process biochemically because intact pseudopodia could not be isolated from cell bodies. Now, on page 725, Cho and Klemke describe a clever method for purifying pseudopodia that have been induced to grow or retract, and identify a signaling complex whose assembly and disassembly controls these processes.

Using a chemoattractant, the authors induced cultured mammalian cells to extend pseudopodia through 3.0-μm pores in a membrane. Under these conditions, over 90% of the cells extend a single leading pseudopodium, allowing Cho and Klemke to scrape away the cell bodies and extract the remaining pseudopodia with detergent. Because removal of the chemoattractant causes the pseudopodia to begin retracting, the system permits the purification of either growing or retracting pseudopodia for direct comparisons.

Analysis of the purified pseudopodia shows that the assembly of a p130Crk-associated substrate (CAS)/c-CrkII (Crk) scaffold leads to Rac1 translocation and activation during pseudopod growth, and Rac1 activation provides a positive feedback loop to maintain the CAS/Crk scaffold. Disassembly of the CAS/Crk scaffold is required for a decrease in Rac1 activity, which in turn induces pseudopod retraction.

The new work simultaneously defines new aspects of signaling in pseudopod growth and retraction, and describes a technique that should be broadly useful in future studies of cell migration. In addition to allowing the purification of pseudopodia for biochemical analysis, the system is quantitative and permits time-lapse imaging of pseudopodia in living cells.

Activating actin

On page 677, Hudson and Cooley characterize Drosophila melanogaster strains with defects in the evolutionarily conserved actin nucleation factor Arp2/3. The work provides significant new insights into the specificity of Arp2/3 activity.

During Drosophila egg development, intercellular connections called ring canals form between nurse cells and developing oocytes, allowing cytoplasm from the nurse cells to flow into the oocytes. These processes require at least three distinct types of actin structures: contractile actin to squeeze cytoplasmic contents, actin to keep ring canals open, and bundles of actin filaments that position the nucleus and stop it from clogging the ring canals. Similar actin bundles are also seen during metamorphosis, when the growth of bristles on the pupal epithelium requires hexagonally packed bundles of actin filaments.

In the new work, Hudson and Cooley found that flies with defective Arp2/3 complexes exhibit specific flaws in the formation of the loosely organized actin filaments in ring canals, and that Arp2/3 components are localized to the ring canals. However, the mutants are still able to form parallel actin filament bundles in nurse cells and epithelial bristles, suggesting that other factors are required for actin nucleation in these structures. The authors are now using their Drosophila system to try to identify these factors.

In addition to defects in oogenesis, Arp2/3-deficient flies exhibit several other developmental phenotypes, and on page 689, Zallen et al. build on the results of Hudson and Cooley to dissect the activities of two Arp2/3 activators. Previous work has shown that members of the WASp-Scar/WAVE family of proteins can regulate Arp2/3 activity, and that the two main branches of this protein family, WASp and Scar/WAVE, are highly conserved. Mutations in SCAR, which encodes the only Drosophila Scar/WAVE subfamily protein, cause flaws in oogenesis and several other developmental processes that are strikingly similar to some of the phenotypes seen in Arp2/3 complex mutants. Mutants in the single WASp homologue in the fly undergo apparently normal oogenesis, but display defects in the Arp2/3-dependent process of asymmetric cell division in neural lineages, indicating that SCAR and Wasp induce distinct cellular activities of Arp2/3 during development.