Coping with the stress of morphogenesis

As a multicellular organism develops, its tissues are often subjected to powerful internal and external mechanical stresses. On page 757, Bosher et al. describe a set of critical reinforcements that allow *C. elegans* embryos to weather these forces rather than be torn apart. Besides illuminating a critical aspect of morphogenesis, the work establishes a new model for analyzing the coupling of tissues during development.

Using a genetic screen, the authors found that mutations in a locus called *vab-10*, which corresponds to spectraplakin, cause defects in the elongation of worm embryos. *C. elegans* *vab-10* encodes two protein isoforms. Mutations that affect VAB-10A isoforms disrupt fibrous organelles, which are molecular and functional homologues of vertebrate hemidesmosomes. These mutations cause epidermal cells to detach from the cuticle and muscles during elongation. When VAB-10B isoforms are disrupted, the epidermis instead becomes thicker.

The results suggest that VAB-10A proteins allow epidermal cells to resist external stresses, whereas VAB-10B proteins ensure that the basal and apical membranes of the cells are locked a fixed distance apart, even in the presence of powerful internal stresses. The *C. elegans* system should now provide a useful platform to study how spectraplakins modulate and direct these forces.

Glycosylation’s active site—finally

Beginning on page 715, Nilsson et al. drive home the lesson that persistence pays. Using established techniques on a system that has frustrated similar efforts for years, the authors identified the active site of the mammalian oligosaccharyltransferase (OST) complex. In the endoplasmic reticulum, OST carries out N-linked glycosylation, one of the most common and least understood eukaryotic protein modifications. Previous efforts to identify the active site of OST using peptide substrates, or even to determine which of the proteins in the OST complex contains the active site, have produced conflicting results.

The authors designed nascent polypeptide chains with cryptic glycosylation sites incorporating photoreactive probes. As the cryptic site translocates through the endoplasmic reticulum membrane, it can be cross-linked first to components of the translocon pore, and then to the OST. Strikingly, only one OST protein, STT3, is cross-linked in this way, providing strong evidence that the nascent chain portion of the OST active site lies largely or entirely within STT3. Probes placed immediately adjacent to the cryptic glycosylation sequence did not cross-link any OST components. Thus, the high specificity of OST for the glycosylation sequence, and the short residence time of incorrect and glycosylated sequences in the active site, probably doomed earlier efforts to cross-link nascent chains to OST.

Dance of the podosomes

Macrophages crawl across a substrate using podosomes, focal complex-like adhesions that form and disappear rapidly at the cell’s leading edge. Beginning on page 697, Evans et al. provide a high-resolution view of the dynamic turnover of these structures, revealing some surprising behavior and suggesting a novel mechanism of cell migration.

Using fluorescently labeled podosome components and quantitative 4-D microscopy, the authors show that the majority of leading edge podosomes either assemble from older podosomes or form through the dramatic fragmentation of a large podosome cluster precursor (PCP). In the first pathway, simple podosomes undergo both fission and fusion events. This often produces a sort of forward stepping movement, when a trailing podosome fuses with one closer to the leading edge. The other pathway begins with a podosome that grows to several times normal size to form a PCP. The PCP then fragments rapidly into a cluster of four to six individual podosomes. In contrast to focal adhesions, which stick to a substrate and allow a cell to pull itself forward, podosomes appear to step forward more or less continuously. This dynamic crawl may allow macrophages to adapt quickly while moving through complex tissues.

A PCP (arrowhead) turns into four podosomes (arrows) during migration.