## Bridging the gap between atlastin conformations



Compared with a control cell (leff), the ER network is unbranched in cells expressing an atlastin mutant lacking a key intramolecular salt bridge (right).
 orin-Leisk et al. describe how a salt bridge drives a conformational change in the atlastin GTPase to promote membrane fusion and ER branching.

Atlastins are membraneanchored members of the dynamin family of GTPases that tether and fuse ER tubules together in order to maintain the organelle's branched morphology. Crystal structures suggest that atlastin molecules in opposing ER membranes initially dimerize head-on to tether the membranes together. GTP hydrolysis is then thought to trigger a rotation in the dimer's conformation that brings the two membranes close enough to fuse with each other.

Morin-Leisk et al. identified several mutations in atlastin that prevented the GTPase from maintaining a branched ER network.

## For Myo4p, two heads are better



Myo4p and She3p form elongated, single-headed structures on their own (left), but they assemble into two-headed, V-shaped complexes in the presence of She2p (right).

The mRNA-binding protein She2p couples two myosin motors together in order to transport mRNAs through yeast cells, Krementsova et al. report.

Myo4p is a class V myosin that carries mRNAs along actin cables into the buds of S. cerevisiae. Myo4p links to its cargo by pairing up with the adaptor protein She3p, which, in turn, binds the mRNA-binding protein She 2 p. In vitro, however, Myo4p and She3p fail to move continuously along actin tracks because, unlike many other class V myosins, the motor doesn't homodimerize and therefore only has one ATPase head domain, which can't take processive steps along the filament on its own.

## A new spin on radial spokes



A pair of radial spokes (red) project from an outer microtubule doublet (gray) in a Chlamydomonas flagellum.

Pigino et al. describe the three-dimensional structure of radial spokes, key regulatory complexes that determine how cilia and flagella move.

Radial spokes connect the central pair of microtubules in cilia and flagella axonemes to the nine outer microtubule doublets and are thought to regulate how dynein motors slide these microtubules to generate movement. Each spoke contains at least 23 proteins, with two or three spokes (depending on the species) clustering together at regular intervals along the axoneme. How the spokes function is unclear, however, so Pigino et al. used cryoelectron tomography to observe their structure in various cilia and flagella.

Two of these mutations affected either the glutamate or the lysine residue of an ionic salt bridge that forms when atlastin assumes its "postfusion" conformation. Altering the charge on either of these polar amino acids had no effect on GTP binding or hydrolysis but inhibited the assembly of "postfusion" atlastin dimers. Restoring the salt bridge by reversing the charge on both residues rescued atlastin dimerization and ER branching.

The salt bridge therefore promotes ER tubule fusion by stabilizing atlastin's postfusion conformation. Surprisingly, MorinLeisk et al. found that GTP hydrolysis wasn't required for the transition to this conformation, at least for soluble versions of atlastin lacking the GTPase's transmembrane domain. Senior author Tina Lee now wants to determine whether the same is true for full-length atlastin and, if so, to investigate which part of atlastin's fusion mechanism is dependent on nucleotide hydrolysis.
Morin-Leisk, J., et al. 201 1. J. Cell Biol. http://dx.doi.org/10.1083/ ¡cb. 201105006.

## than one

Krementsova et al. found that Myo4p-She3p complexes gained the ability to move processively along in vitro actin filaments in the presence of She2p. The mRNA-binding protein formed tetramers in solution, each of which recruited two Myo4pShe3p dimers. The resulting complex walked along actin cables like vertebrate myosin Vs, taking similarly sized steps even though the two Myo4 motors are linked via a series of adaptor proteins instead of through a direct interaction.

Senior author Kathleen Trybus thinks that She2p's ability to couple Myo4p motors is an elegant way of regulating mRNA transport, because the myosin will only move along actin cables when linked to its cargo. Trybus now wants to investigate whether mRNAs themselves alter the activity of the Myo4p-She3pShe2p complex.
Krementsova, E.B., et al. 201 1. J. Cell Biol. http://dx.doi.org/10.1083/ jcb. 201106146.

Pigino et al.'s reconstructions of flagella from the alga Chlamydomonas revealed that spokes project from outer microtubule doublets and split at their necks to form two closely apposed heads near the center of the axoneme. The symmetry of these structures supported the idea-previously suggested by biochemical experiments-that radial spokes assemble through the dimerization of smaller, precursor structures. In addition, by comparing the structures of radial spokes in Chlamydomonas mutants lacking particular spoke proteins, the researchers were able to assign many of these components to specific locations within the complex.

One surprise was that Chlamydomonas, whose radial spokes are thought to exist in pairs, actually has a short protrusion next to each pair, exactly where the third spoke exists in the cilia of species like the protozoan Tetrahymena. Senior author Takashi Ishikawa says it's unclear whether Chlamydomonas lost, or Tetrahymena gained, a full third spoke during evolution.
Pigino, G., et al. 2011. J. Cell Biol. http://dx.doi.org/10.1083/icb. 201106125.

