CYTOSKELETON

Keeping dynamic

The actin-severing and -depolymerizing activities of cofilin help to keep the actin cytoskeleton dynamic, which is crucial for cellular processes such as cell division and motility. Phosphorylation of cofilin on Ser3 by LIM kinase (LIMK) inhibits its activity, but a broadly expressed, counteractive phosphatase has remained elusive. Gary Bokoch's group, though, has unearthed chronophin (CIN), a haloacid dehydrogenase (HAD)-type phosphatase that dephosphorylates cofilin.

CIN was isolated from bovine brain cytosol on the basis of its ability to dephosphorylate Ser3-phosphocofilin, and sequence homology showed the presence of orthologues from bacteria to humans. CIN protein is widely expressed throughout human tissues; at the subcellular level, it colocalizes with F-actin and cofilin in membrane ruffles and lamellipodia, and mirrors the dynamic distribution of cofilin to regions of actin dynamics during mitosis.

Having confirmed that CIN had intrinsic phosphatase activity, which was abolished by the mutation of a crucial aspartate in the N-terminal HAD motif DXDXT, the authors then verified that recombinant CIN could dephosphorylate cofilin. CIN showed no activity towards tyrosine-phosphorylated peptide substrates. Next, Bokoch's group showed that transfected wild-type CIN decreased the steady-state levels of phosphocofilin in HeLa cells by ~50%, and markedly induced cofilin dephosphorylation during mitosis. Conversely, when CIN was depleted from HeLa cells using RNA inhibition (RNAi), the levels of phosphorylated cofilin increased.

But how does CIN actually affect cells? Expressing recombinant CIN for prolonged periods of time caused cells to lose their cortical actin cytoskeletons, decrease their amount of stress fibres, and eventually round up and detach from the substrate. Over a shorter time period, membrane ruffling increased. A phosphatase-dead CIN construct increased actin polymerization, which is similar to the effect of increasing LIMK activity.

As interfering with CIN levels also perturbed cell division (causing multinucleate cells to form), the authors studied the cofilin phosphorylation profile in response to CIN during mitosis. As noted previously, wild-type CIN reduced cofilin phosphorylation during mitosis. Phosphatase-dead CIN caused levels of phosphorylated cofilin to accumulate, which resulted in prolonged mitotic progression and impaired cytokinesis — in particular, contractile-ring defects. Such phenotypes are similar to those that arise from defective cofilin function in lower organisms.

So CIN is required to control cofilin activity during cell division. On the basis of the authors' other observations and the expression patterns of CIN, this unusual HAD-type phosphatase probably influences motility, polarity and membrane dynamics as well.

Katrin Bussell

References and links

ORIGINAL RESEARCH PAPER Gohla, A., Birkenfeld, J. & Bokoch, G. M. Chronophin, a novel HAD-type serine protein phosphatase, regulates cofilin-dependent actin dynamics. *Nature Cell Biol.* 5 Dec 2004 (doi:10.1038/ncb1201)

WEB SITE

Gary Bokoch's laboratory: http://www.scripps.edu/imm/bokoch/

IN BRIEF

PROTEIN EVOLUTION

Components of coated vesicles and nuclear pore complexes share a common molecular architecture.

Devos, D. *et al. PloS Biol.* **2**, e380 (2004)

How did the nuclear pore complex (NPC) evolve when prokaryotic organisms lack a comparable transport system? These authors characterized the structures of seven proteins that form a core building block of the NPC, and found that they are structurally similar to protein components of the main types of vesicle-coat complex that maintain vesicle-trafficking pathways. So NPC and coat proteins evolved from an early, common membrane-curving module that led to the development of the complex intracellular membrane system of modern eukaryotes.

CELL FATE

A critical role for Cyclin E in cell fate determination in the central nervous system of *Drosophila melanogaster*.

Berger, C. *et al. Nature Cell Biol.* 5 Dec 2004 (doi:10.1038/ncb1203)

Structural and functional diversity in the central nervous system of *Drosophila melanogaster* is generated by different cell lineages that arise from some of the serially homologous neuroblasts within the thoracic and abdominal segments. Berger *et al.* showed that the cell-cycle protein cyclin E is required to specify cell fate in the thoracic neuroblast NB6-4 lineage. It is expressed asymmetrically after the first division of NB6-4t cells and functions upstream of *prospero* and *glial cell missing* to generate neuronal cells.

CHROMATIN

Acetylation by Tip60 is required for selective histone variant exchange at DNA lesions.

Kusch *et al. Science* 4 Nov 2004 (doi:10.1126/science.1103455)

These authors showed that the *Drosophila melanogaster* Tip60 complex uses both its chromatin-remodelling and acetylation activities to catalyse the exchange of the histone variant H2Av (the fly homologue of human H2AX). By mimicking the phosphorylation of H2Av in a manner analogous to the DNA-damage-induced phosphorylation of mammalian H2AX, Kusch *et al.* found that histone exchange is enhanced by Tip60-mediated acetylation of phosphorylated H2Av. This could explain how phosphorylated H2AX is removed from the damage site.

AGEING

The AMP-activated protein kinase AAK-2 links energy levels and insulin-like signals to lifespan in *C. elegans*.

Apfeld, J. *et al. Genes Dev.* 1 Dec 2004 (doi:10.1101/gad.1255404)

Limiting energy availability is thought to extend lifespan in some organisms. Apfeld and colleagues have now found that the cellular ratio of AMP:ATP, which is a measure of energy levels, increases with age in *Caenorhabditis elegans*. They also identified an enzyme, AMP-activated protein kinase α-subunit (AAK-2), that acts as a sensor of high levels of AMP and functions to extend lifespan.