Nucleosomes get peeled and rewrapped

Remodeling by the SWI/SNF complex involves peeling and rewrapping of the DNA that winds around the histone octamer, according to Stefan Kassabov, Blaine Bartholomew, and colleagues (Southern Illinois University, Carbondale, IL).

Most models for SWI/SNF remodeling have been based on assays that measure either the accessibility of DNA to nucleases or the level of DNA supercoiling. The Illinois team, however, directly measured where specific sites on the histone octamer surface contact DNA before and after remodeling by attaching photoreactive groups to sites within the histone fold regions.

SWI/SNF displaced the histone octamers so that they were hanging on to only about 100 bp of DNA as compared to the normal 146 bp. “The fact that it can move an extra 50 bp beyond the end says that it is not just sliding by pushing the DNA into the nucleosome but it displaces large stretches of DNA off the surface,” says Bartholomew. The stable remodeled nucleosome also forms an unusual intranucleosomal loop in which the free DNA end is efficiently looped back onto the exposed surface of the octamer.

Using their direct readout, the team estimated that a remodeling event may require less than 1 s and the hydrolysis of fewer than 10 ATP molecules. This is a considerably lower energy estimate than those made by others, and provides further evidence that remodeling may involve a limited number of steps.

Endosomes make tracks

Early endosomes may help to create their own short-range tracks, according to a report from Stéphane Gasman, Yannis Kalaidzidis, and Marino Zerial (Max Planck Institute, Dresden, Germany). The results also suggest parallels between membrane trafficking, signaling, and regulation of the cytoskeleton.

Trafficking of early endosomes requires the small GTPase RhoD. Zerial’s group identified the first known downstream effector for RhoD, hDia2C, a relative of the actin-polymerizing protein formin. In vivo, RhoD recruited hDia2C onto endosomes, where the two proteins probably stabilize the association of endosomes with the actin network, as their overexpression caused the vesicles to align along actin filaments. In the overexpressing cells, endosomal motility was also slowed and long-range trafficking was inhibited, probably because the endosomes were unable to jump from actin to microtubules, where Rab5 stimulates long-range movements.

The results indicate a similarity between endosome motility and adhesion formation. Endosome-localized hDia2C recruited and activated c-Src kinase, which was required for the effects of RhoD and hDia2C on trafficking. Src is therefore another example of a signaling molecule required for organelle transport, although its relevant phosphorylation targets have not been identified. Actin remodeling during focal contact formation involves similar players, namely, RhoA, mDia1, and Src. Thus, although different molecules are used, the mechanisms underlying actin-dependent organelle trafficking and adhesion assembly appear to be conserved.

Reference: Gasman, B., et al. 2003. Nat. Cell Biol. 10.1038/ncb935.

Riding Kar9 into the bud

Alignment of the mitotic spindle with the axis of cell division in budding yeast ensures that one and only one chromatid set is pulled into the bud. Microtubule interactions with the cortex set this arrangement by capturing one spindle pole at the bud site. Now, Dimitris Liakopoulos, Justine Kusch, Yves Barral (Swiss Federal Institute of Technology, Zürich, Switzerland), and colleagues find that polarity is achieved because only one set of microtubules is marked for this interaction with the cortex.

The Swiss group identified a protein that distinguishes microtubules from the two spindle poles. This myosin-interacting protein, Kar9, localized specifically to the spindle pole destined for the bud (daughter-bound pole). Kar9 polarity depended on the cell cycle kinase Cdk1. Kar9 phosphorylation correlated with its decreased interaction with the microtubule- and spindle-pole–binding protein, Bim1, thus probably reducing its association with the mother-bound pole.

Meanwhile, microtubules at the daughter-bound pole were loaded with Kar9 and were guided to the bud in a myosin-dependent manner along the polarized actin network. Loss of Kar9 asymmetry (through inhibition of Cdk1) caused both sets of microtubules to be drawn to the bud, indicating that Kar9 distinguishes microtubules from the two poles. “For a long time, people were thinking microtubules were microtubules,” says Barral. “Now, we see they come in different flavors depending on which pole they come from.”

Reference: Liakopoulos, D., et al. 2003. Cell. 112:561–574.