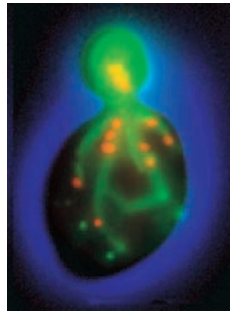


## Peroxisomes on the move

The cell may not be as lazy as we first thought. With an organelle that comes in multiple copies, there is always the option of leaving segregation of that organelle to random chance—in most cases each daughter cell will get at least one copy of the organelle. But budding yeast is not prepared to take that risk, especially when growing daughter cells are so much smaller than mature mothers. On page 979, Hoepfner et al. find that yeast cells actively segregate their peroxisomes.

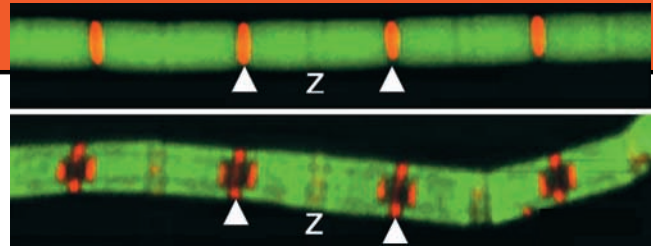


**Peroxisomes track along actin filaments.**

The segregation machinery includes actin cables and a specific motor, Myo2p, and the substrate for the segregation is provided by the dynamin Vps1p, which cleaves the peroxisomes into manageable pieces.

Hoepfner et al. came to these conclusions after observing the movement of individual GFP-labeled peroxisomes along the cell cortex to the bud neck, bud cortex, and bud tip. Peroxisomes localize along actin cables, and movement requires actin and the myosin Myo2p, and is directed toward actin patches. Somehow the segregation works even in the absence of Vps1p function, when there are only one to three peroxisomes per cell.

Peroxisome movement has been studied before in mammalian cells during interphase, but has been found to be microtubule-dependent. The authors believe that this movement may help the peroxisomes to find their substrates in larger mammalian cells, but that actin-dependent movements may dominate the longer-range segregation behavior in both yeast and mammalian cells. ■



**Tmod overexpression (red, bottom) arrests filament extension before it reaches the M line.**

## Tmod points the way

When it comes to actin dynamics, the pointed end of the actin filament is the forgotten cousin. The barbed end is in a state of constant flux in motile cells, pushing out the front of the cell, whereas the pointed end rarely rates a mention. But now, Mardahl-Dumesnil and Fowler report that in the actin-containing thin filaments of developing fly muscles, which grow at a more leisurely pace, the action is at the pointed end (page 1043).

The authors target the pointed end by transiently overexpressing the fly version of Tropomodulin (Tmod) in indirect flight muscles (IFM). If the Tmod is overexpressed during pupation when myofibrils are assembled, a central core of thin filaments is shorter than normal, which in the most serious cases results in adults that are unable to fly. The short filaments are permanently arrested—they do not resume their growth once Tmod levels decline to near normal—and a fraction of the overexpressed form of Tmod stays as a permanent cap on the pointed end.

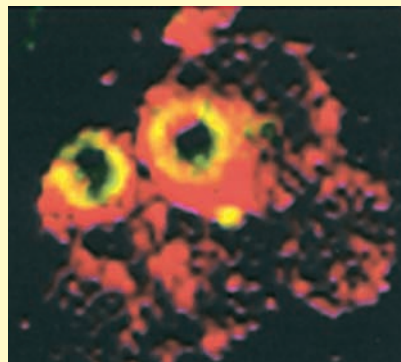
Fowler believes that the excess Tmod overwhelms a factor that normally makes Tmod capping of pointed ends dynamic. By the time the Tmod levels decline again, this factor may be far away in the central M line, conveyed from the foreshortened thin filaments by the continued elongation of the thick filaments of myosin. The outer thin filaments are not affected by the overexpression only because they are born later, after Tmod levels have returned to normal. ■

## Chopping and storing in one

A store is no good if its contents cannot be accessed. On page 991, Jiang et al. suggest that plant seeds might solve this problem by storing digestive enzymes in a membrane-bound structure called the globoid that is fully contained within the seed's protein storage vacuole (PSV). In theory, such a structure could protect the stored proteins from cleavage early in development, but then later allow access so the proteins can be mobilized for growth of the young seedling.

The globoid has been known for many years as a site where crystals of phytic acid (a phosphate source) are

stored. Jiang et al. use permanganate staining to reveal that the compartment has a limiting membrane surrounding it.



**A proton pump (green) identifies a vacuole within the PSV (red).**

The membrane includes a vacuolar transmembrane protein, and the contents of the compartment include a lytic vacuolar protein. Thus, the authors suggest that this may be a degradative vacuole contained within the storage vacuole.

How this double vacuole structure might be generated remains a mystery. The lack of a double membrane suggests that the putative degradative compartment is probably not enveloped by the PSV. Choosing between other models for vacuolar formation will be easier once the relative orientation of globoid transmembrane proteins is known. ■

## Calcium-coated chromosomes

We know that cations associate with DNA and help to neutralize the negative charges of phosphate groups, but for the most part the analysis of cation effects has not gone any further. Now Strick et al. demonstrate that calcium is distributed on chromosomes in a particular pattern that may implicate it in regulating a chromosomal protein and thus chromosomal structure (page 899).

The author's method of choice is secondary ion mass spectrometry (SIMS). Unlike previous X-ray-based techniques, SIMS yields the depth and localization information characteristic of the results from a confocal microscope. It does so by first using a laser to displace a thin, localized layer of the material under study, and then using mass spectrometry to analyze the displaced material. Strick et al. refine this technique to measure isotopes of a number of cations, but the most interesting results come in the measurements of  $Mg^{2+}$  and  $Ca^{2+}$ .

Both are associated with mitotic chromatin, but  $Mg^{2+}$  is evenly distributed across the chromatin, whereas  $Ca^{2+}$  is concentrated at



Calcium (pseudocolored from blue to orange) localizes to mitotic chromosome axes.

the AT-rich axis of each chromosome. This mimics the localization of certain chromosome scaffold proteins, including topoisomerase II

(Topo II).

Leading up to mitosis, Topo II is needed to cleave and allow the untangling of DNA as it is condensed.

But later in mitosis the activity of Topo II is turned off, perhaps changing the protein into a structural component that helps to keep the center of the condensed chromosome together.

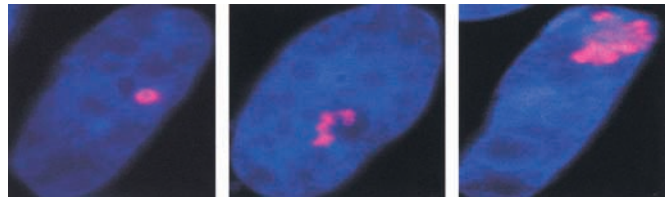
The basis of this inhibition was unknown. Now Strick et al. find that the altered ratio of  $Mg^{2+}$  and  $Ca^{2+}$  at the center of the chromosome is, at least in vitro, sufficient to shut off the cleavage activity of Topo II. Mitotic phosphorylation of Topo II may recruit Topo II to the chromosome but, when  $Ca^{2+}$  binds to the chromosomes during mitosis, this local increase in the concentration of  $Ca^{2+}$  may help to shut down Topo II activity. Although this model is appealing, it is yet to

be proven, and there may be many other things that the concentrated calcium is doing at the central axis of the chromosome. ■

## An unfolding breast cancer story

BRCA1 is one of the most intensely studied proteins in the history of cancer research, and it has more than its fair share of proposed protein partners and postulated activities. Rong Li hopes that he has made sense out of this mountain of information in a paper by Ye et al. starting on page 911. He and his coauthors suggest that BRCA1's effects on transcription and DNA repair have as their root cause a chromatin-unfolding activity of two BRCT repeats at the COOH terminus of the protein.

The assay for unfolding used here has been used with transcriptional activators. It involves targeting BRCA1 or a subset of the protein to multiple (probably several thousand) lacO repeats scattered over 90 Mb of heterochromatin. In 14% of cells expressing BRCA1 linked to the lac repressor there is decondensation of this focused spot of DNA. Either BRCT1 or BRCT2 alone cause an even greater extent of decondensation in a higher percentage of cells (60%), although a construct containing both BRCT repeats has no effect. This suggests that the intact protein may be inhibited for chromatin unfolding, either by the binding of another protein or because of an intramolecular interaction that is relieved by another protein.



DNA decondensation with wild-type (center) or mutant (right) BRCA1.

A candidate for that other factor is COBRA1, which Ye et al. isolate as a protein that interacts with BRCT1. COBRA1 by itself can mediate chromosome decondensation. A subset of cancer-causing BRCA1 mutations that result in greater unfolding activity also show greater COBRA1 binding activity, although Ye et al. do not yet know whether such mutations have dominant effects either in cells or people.

Chromatin unfolding could change both transcription and DNA repair by increasing DNA accessibility and helping to recruit other factors. Additional recruitment activities of BRCA1 still seem to be important, as BRCT1 by itself can cause unfolding but not transcription enhancement. ■