In This Issue

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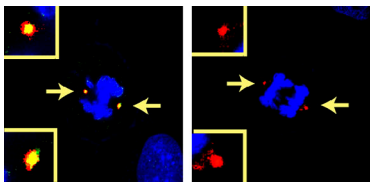
Bleach at the roots of mitotic progression



Phosphorylated peroxiredoxin I (green) is found near the centrosomes (red) early in mitosis (left) but not during anaphase (right).

Lim et al. show how hydrogen peroxide at the centrosome spurs cells to advance through mitosis.

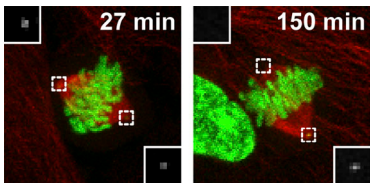
Cdk1 and regulatory proteins such as cyclin B1, Plk1, and Aurora A cooperate to initiate mitosis. To exit mitosis, cells destroy the regulatory proteins, an effect triggered by the APC/C when it is bound to its coactivator Cdh1. Cdk1 adds phosphates to Cdh1 that prevent it from attaching to the APC/C, but the phosphatase Cdc14B removes them. Cdh1, Cdc14B, and the components of the APC/C are present at the centrosome, but it is unclear how cells rein in Cdc14B activity during early mitosis to prevent premature activation of the APC/C-Cdh1 combo.

Lim et al. discovered that hydrogen peroxide helps control Cdc14B activity. The amount of hydrogen peroxide in a cell starts to rise as it enters G2, but the researchers found that an enzyme called peroxiredoxin I shelters the centrosome. Cells phophorylated and inactivated peroxiredoxin I near the beginning of mitosis, causing a surge in hydrogen peroxide around the centrosome that spurred mitotic progression. Targeting an enzyme that neutralizes hydrogen peroxide to the centrosome delayed cells' entry into mitosis.

The rise in hydrogen peroxide at the centrosome shut down phosphatases such as Cdc14B, allowing phosphorylation of Cdh1 and inactivation of the APC/C. Later in mitosis, phosphatases PP1 and PP2A stepped in to dephosphorylate and reactivate peroxiredoxin I, cutting hydrogen peroxide levels and allowing the APC/C to begin its work.

Lim, J.M., et al. 2015. *J. Cell Biol.* http://dx.doi.org/10.1083/jcb.201412068

p53 censuses centrosomes



Control cells (left) quickly arrange their chromosomes (green) during mitosis. The process is slower in cells missing a centrosome (right).

p53 guards the genome by preventing cells with abnormal numbers of centrosomes from dividing, Lambrus et al. show.

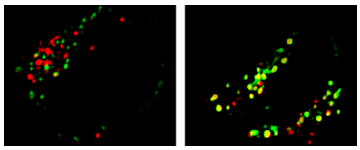
In each cell cycle, a cell duplicates its centrosome, producing a pair of the structures that can serve as the poles of the mitotic spindle. Cancer or developmental defects can stem from mistakes in duplication that result in cells carrying an incorrect number of centrosomes. However, researchers haven't been able to confirm that centrosomes help organize the mitotic spindle.

To address this question, Lambrus et al. used a technique that allowed them to remove centrosomes at precise times. They outfitted Polo-like kinase 4 (Plk4), a protein that controls centrosome duplication, with a segment known as a degron. Adding a plant hormone spurs the destruction of proteins carrying this segment, and the researchers found that the technique rapidly cut Plk4 levels and blocked centrosome duplication. When the researchers prolonged the depletion of Plk4, cells would divide a couple of times before the cell cycle halted permanently. However, the usual suspects to trigger this growth arrest—DNA damage, improper separation of chromosomes, and prolonged mitosis—weren't responsible. Lambrus et al. hypothesized that cells have a centrosome surveillance system to protect against harmful mitoses that arise from divisions with the wrong number of centrosomes.

The researchers found that if they depleted the tumor suppressor p53, cells that chronically lacked Plk4 could divide without centrosomes. These cells made more errors during mitosis, emphasizing the importance of the surveillance mechanism. The next step for researchers is to discover how a cell senses when it's short on centrosomes.

Lambrus, B.G., et al. 2015. *J. Cell Biol.* http://dx.doi.org/10.1083/jcb.201502089

Even killers recycle



VAMP8 (green) associates with recycling endosomes (red, right) but not early endosomes (red, left)

Recycling endosomes allow cytotoxic T cells to deploy their cell-slaying weapons, Marshall et al. reveal.

Cytotoxic T cells can dispatch tumor cells and cells infected by pathogens, killing their target within minutes by releasing the contents of specialized lysosomes known as cytotoxic granules. In general, exocytosis requires that R-SNARE proteins on vesicles interlock with Q-SNARE proteins on the plasma membrane, enabling the two membranes to fuse. One SNARE protein involved in granule release is the Q-SNARE syntaxin-11 (Stx11). Evidence from knockout mice also implicates VAMP8, which belongs to a family of R-SNARE proteins.

Marshall et al. found that VAMP8 built up at the immunological synapse, the junction between a cytotoxic T cell and its victim. To their surprise, the researchers found that VAMP8 did not associate with cytotoxic granules. Instead, it rode on recycling endosomes, organelles that normally return proteins to the plasma membrane. These endosomes fused with the plasma membrane at the immunological synapse.

Depleting VAMP8 not only prevented recycling endosomes from fusing with the plasma membrane, but it also abolished cytotoxic granule exocytosis. The team discovered that the VAMP8-carrying recycling endosomes arrived at the synapse before the cytotoxic granules and brought an important cargo—Stx11.

The study indicates that recycling endosomes outfitted with VAMP8 deliver Stx11 to the immunological synapse. The researchers hypothesize that Stx11 then latches onto an unidentified SNARE on cytotoxic granules, connecting them to the plasma membrane and allowing them to jettison their contents.

Marshall, M.R., et al. 2015. *J. Cell Biol.* http://dx.doi.org/10.1083/jcb.201411093