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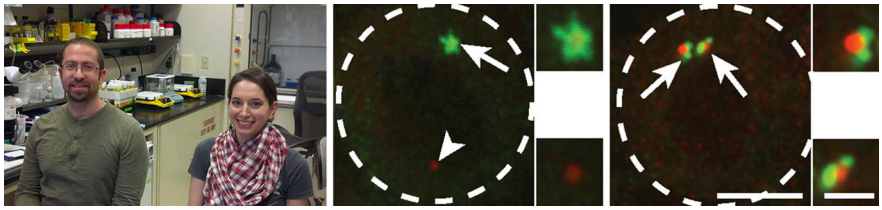
PLP helps the mother centrosome stay mum

Protein aids centrosome segregation by limiting centrosome activity in neural stem cells.

Focal Point



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Nasser Rusan (left) and Dorothy Lerit (right) explore how and why the centrosomes of *Drosophila* neuroblasts show asymmetric levels of microtubule nucleation and organizing activity. During interphase, the pericentrin orthologue PLP is enriched on the basal, mother centrosome, where it restricts the recruitment of Polo kinase, the master regulator of centrosome maturation and activity. In wild-type neuroblasts (center), the microtubule-nucleating protein γ-tubulin (green) only localizes to the active centrosome on the apical side of the cell. But in PLP-null cells (right), γ-tubulin is active at both centrosomes (red), which limits the ability of neuroblasts to separate their centrosomes and efficiently segregate them into different daughter cells.

Drosophila neuroblasts give rise to the fly's central nervous system by dividing asymmetrically to produce a daughter neuroblast and a ganglion mother cell (GMC). The neuroblast's centrosomes are themselves asymmetric in terms of their activity, localization, and pattern of inheritance. Lerit and Rusan describe how the centrosomal protein pericentrin controls this asymmetry to ensure that neuroblast centrosomes segregate correctly (1).

After a neuroblast's centrosome duplicates in S phase, the younger, "daughter" centrosome remains active throughout interphase, nucleating and organizing microtubules from the apical side of the cell. The older, "mother" centrosome, on the other hand, is inactivated and translocated to the basal side of the cell, only gaining the proteins it needs to nucleate microtubules as the neuroblast enters mitosis (2, 3). The positions of the two centrosomes help orient the mitotic spindle so that the mother centrosome segregates into the GMC while the apical daughter centrosome partitions into the new neuroblast.

Nasser Rusan, from the National Heart, Lung, and Blood Institute in Bethesda, Maryland, described the asymmetric activity of neuroblast centrosomes as a postdoc at the University of North Carolina (2). But the functional significance of this behavior remained unclear. "We thought this might be important to the function of neuroblasts," Rusan explains, "but we had no way to test this hypothesis until we understood how the centrosome's activity is regulated so that we could turn the inactive centrosome on."

> **"People don't usually think about negative regulation at the centrosome."**

Rusan and his postdoc, Dorothy Lerit, therefore set out to identify differences between the mother and daughter centrosomes that might explain their asymmetric activity levels. The researchers stained neuroblasts for a variety of centrosomal proteins and identified one, known as pericentrin-like protein (PLP), that was enriched on the inactive, basal centrosome during interphase. When neuroblasts entered mitosis, however, PLP localized to both centrosomes evenly, and, as the amount of PLP on the basal centrosome decreased, levels of the microtubule-nucleating protein γ-tubulin increased, indicating that the centrosome was active.

PLP is the *Drosophila* orthologue of pericentrin, a protein that organizes the pericentriolar material of centrosomes during mitosis to promote the organelle's microtubule-organizing activity. But PLP's localization pattern suggested that, in interphase cells, it might inhibit the activity of the neuroblast's basal centrosome. To investigate this possibility, Lerit and Rusan examined the neuroblasts of flies lacking PLP. "In about 50% of them, both centrosomes could recruit γ-tubulin and nucleate microtubules during interphase," Rusan says. "That's something we never see in wild-type neuroblasts."

PLP inhibits basal centrosomes by preventing the recruitment of Polo kinase, the master regulator of centrosome activity. In wild-type neuroblasts, Polo localizes to the active, apical centrosome during interphase and to both centrosomes during mitosis. In PLP-deficient neuroblasts, however, Polo localized to both centrosomes throughout the cell cycle. "People don't usually think about negative regulation at the centrosome, but PLP seems to limit Polo's access," Rusan says.

PLP itself is regulated by centrobin, a protein that specifically localizes to the apical centrosome of neuroblasts and is required for its activity during interphase (4). Targeting centrobin to the basal centrosome caused a loss of PLP and a corresponding increase in γ-tubulin levels.

Having identified the proteins that regulate the asymmetric activity of neuroblast centrosomes, Lerit and Rusan could now test the significance of this phenomenon. In PLP-null neuroblasts, abnormally active mother centrosomes often failed to migrate to the basal side of the cell. The cells still managed to form a correctly oriented mitotic spindle and divide asymmetrically to form a new neuroblast and a GMC. "But a small percentage of neuroblasts ended up with too many centrosomes," Rusan says. This phenotype was exacerbated if the flies were reared at lower temperatures to inhibit the dynamics of microtubules and motor proteins involved in centrosome migration and inheritance.

Asymmetric centrosome function therefore helps neuroblasts segregate their centrosomes to each of their daughter cells. Rusan now wants to investigate how PLP switches from inhibiting to promoting centrosome activity during mitosis.

1. Lerit, D.A., and N.M. Rusan. 2013. *J. Cell Biol.* http://dx.doi.org/10.1083/jcb.201303141.
2. Rusan, N.M., and M. Peifer. 2007. *J. Cell Biol.* 177:13–20.
3. Rebollo, E., et al. 2007. *Dev. Cell*. 12:467–474.
4. Januschke, J., et al. 2013. *Nat. Cell Biol.* 15:241–248.