Flies stretch their cells to avoid a chromatin trap

Mar Carmena

Wellcome Trust Centre for Cell Biology, University of Edinburgh, EH9 3JR Edinburgh, Scotland, UK

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Before the final step of cytokinesis, termed abscission, dividing cells need to ensure that the cleavage plane is clear of chromatin. In this issue, Kotadia et al. (2012. *J. Cell Biol.* http://dx.doi.org/jcb.201208041) show that in *Drosophila melanogaster*, larval neuroblasts elongate to allow segregation of extra-long chromatids and clearance of the midzone, thereby avoiding cytokinesis failure and aneuploidy.

Successful mitosis requires the spatiotemporal coordination of chromosomal and cytoskeletal events. The mitotic cell has evolved a series of surveillance mechanisms to guarantee the fidelity of cell division and avoid aneuploidy (Holland and Cleveland, 2009). These mitotic checkpoints act at specific stages of the cell cycle to ensure that everything goes according to plan before proceeding to the next stage.

One of these crucial points is the moment in which the cell splits up in two through the process of cytokinesis, the final stage of cell division (Eggert et al., 2006). The initial step in cytokinesis is the localization of myosin at the equatorial cortex, followed by the formation of an actomyosin ring. When this contractile ring closes it also drives the furrowing of the cytoplasmic membrane at the cell equator, compressing the cytoplasm and the midzone microtubules into the midbody. But before the process goes any further, the cell has to check that chromosome segregation has occurred normally and that the midzone is clear of chromatin. If cytokinesis proceeded in the presence of a chromatin bridge, it could result in a negative outcome for the cell: chromosome breakage would generate aneuploidy and cytokinesis failure would lead to tetraploidy. In either case, the consequences would be potentially deleterious. For this reason, the mitotic cell delays abscission, the last stage of cytokinesis (Neto and Gould, 2011), until the chromatin bridges have been resolved: this is called the abscission checkpoint. In recent years, work in yeast, *Drosophila melanogaster*, and mammalian cells has contributed to a better understanding of the abscission checkpoint and its regulation by Aurora B kinase (Carmena, 2012). Aurora B is the enzymatically active component of the chromosomal passenger complex (CPC), an essential protein complex that performs multiple regulatory roles in mitosis and cytokinesis (Carmena et al., 2009; van der Waal et al., 2012)

During normal division, anaphase chromatid compaction and spindle elongation contribute to clear the midzone of chromatin. The presence of chromatin in the midzone can be a consequence of a defect in DNA replication or chromosome architecture. The mitotic cell can deal with unusually long chromosomes in anaphase to some extent, but studies in plants show that there is a limit to the extra length that can be tolerated (Schubert and Oud, 1997). Budding yeast carrying a recombinant version of its two longest chromosomes fused together can increase anaphase compaction of this extra-long chromosome in line with its extra length. This process requires Ipl1 (the yeast homologue of Aurora B) and involves phosphorylation of Histone 3 (Ser10; Neurohr et al., 2011).

Aurora B kinase is involved in the regulation of a pathway that delays abscission when chromatin is present at the midzone (Norden et al., 2006; Steigemann et al., 2009). Recent work has revealed more details of the link between Aurora B and the abscission machinery (Capalbo et al., 2012; Carlton et al., 2012; Carmena, 2012). The CPC component Borealin interacts with Shrb/CHMP4C, a subunit of ESCRT-III, a protein complex responsible for the scission activity. The pathway is not totally conserved: in human cells, Aurora B acts through phosphorylation CHMP4C, but in *Drosophila* it seems that Borealin acts in the regulation of abscission in an Aurora-independent way.

In this issue, Kotadia et al. describe a novel mechanism by which animal cells deal with exceptionally long chromatid arms in late mitosis. *Drosophila* larval neuroblasts elongate during anaphase and telophase to allow chromatin to clear the midzone before cytokinesis proceeds.

Drosophila larval neuroblasts divide asymmetrically into a smaller ganglion mother cell (GMC) and a neuroblast (NB). To generate long chromatids, the authors induced double-strand DNA breaks in the ribosomal DNA of the X chromosome by induction of the I-CreI endonuclease. The resulting chromosomal fragments remain linked by a DNA tether that increases chromatid length (Royou et al., 2010). Using this system, the authors observed that the asymmetrically dividing neuroblasts changed their usual spherical shape, becoming transiently elongated to accommodate the longer chromatin arms. Interestingly, the asymmetry of the division itself was not affected, although the shape change was more noticeable in the GMC, which became almost tubular compared with the neuroblast. The mitotic spindle appeared slightly more elongated and shifted toward the GMC. Most noticeably, although myosin localized to the equator, it did

Correspondence to Mar Carmena: mar.carmena@ed.ac.uk

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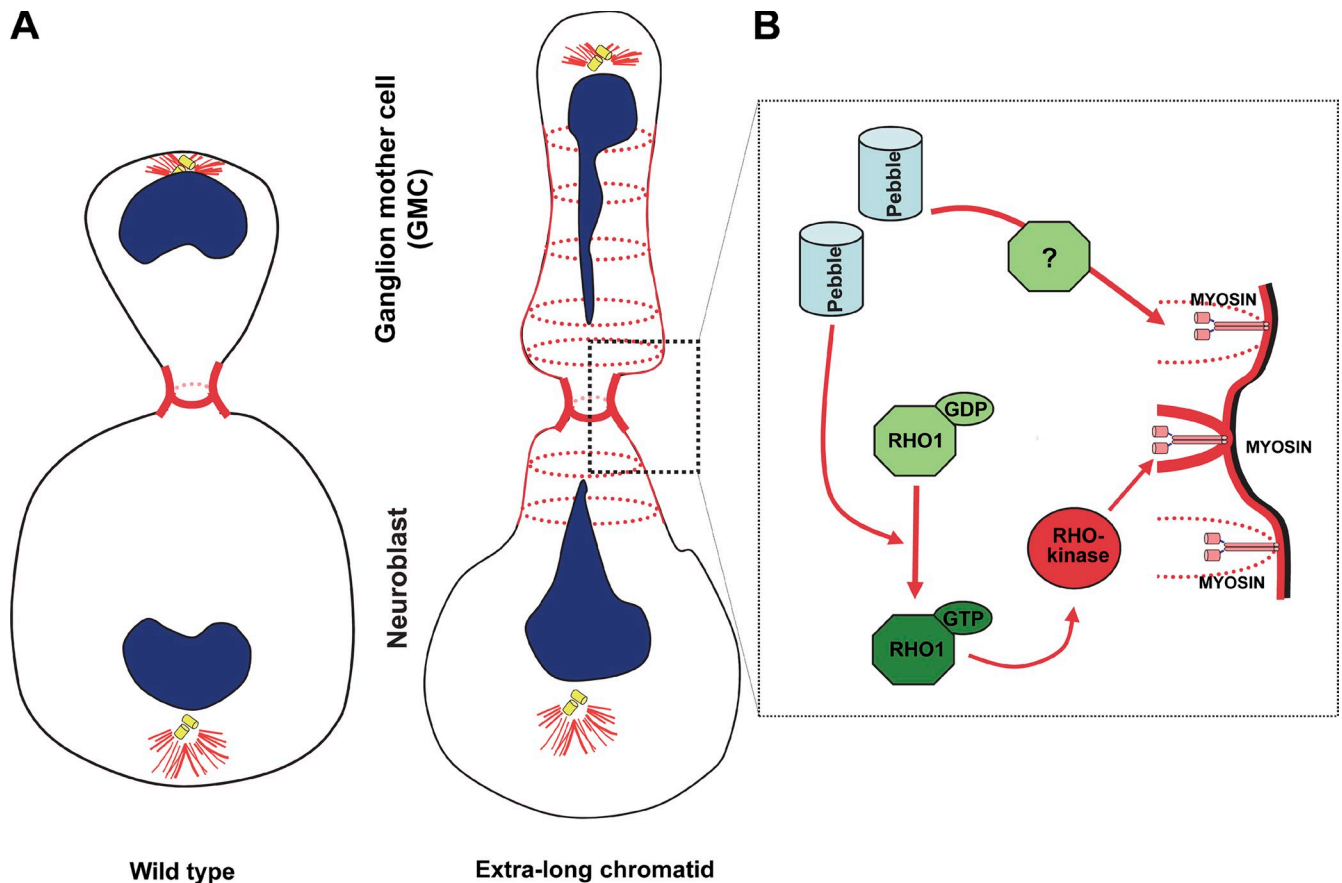www.jcb.org/cgi/doi/10.1083/jcb.201210135

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Figure 1. A novel mechanism that allows cells to clear chromatin from the midzone. (A) Asymmetrically dividing *Drosophila* neuroblasts elongate to allow segregation of an I-CreI–induced extra-long chromatid (right); the presence of extra myosin rings is visible on both sides of the main cleavage furrow. (B) Decreasing levels of the Rho1GEF Pebble abolishes cell elongation but has only a mild effect on cytokinesis. The mechanism by which Pebble regulates the extra myosin rings has yet to be clarified.

so in a much broader way, and several myosin rings appeared on both sides of the main one (Fig. 1). The authors propose that these extra rings constrict the cortex and drive the temporary deformation of the cell, allowing the segregation of the trailing chromatid. If this hypothesis were correct, inhibiting myosin activation by Rho kinase (which is in turn activated by the GTPase Rho1 in *Drosophila*) would inhibit elongation. In agreement with this, the authors showed that a decrease in the activity of Pebble (*pbl*; the Rho GEF that activates Rho1 GTPase) abolished elongation and the formation of extra contractile myosin rings in the presence of extra long chromatids, although cytokinesis was not affected.

This elegant description of a new mechanism used by cells to clear chromatin from the cleavage plane raises a wide range of new questions and opens exciting possibilities. It will be very interesting to find out to what extent this pathway is conserved and how it fits with what we know already about the regulation of abscission. We have only just begun to investigate the mechanisms that control the abscission checkpoint in *Drosophila* cells, but we already know that there are some important differences with other organisms (Capalbo et al., 2012; Carmena, 2012). All the main players involved in this newly described mechanism are conserved in other species, but, as the authors themselves mention, it remains possible that the pathway could be acting exclusively in asymmetrically dividing fly neuroblasts.

Conversely if the pathway is indeed conserved, it will be worth exploring how it is integrated with other mechanisms that regulate abscission. In particular a most intriguing question for future research will be how this new pathway is coordinated with the previously described Aurora B–dependent pathways. Aurora B kinase has not only been shown to have a key function in the regulation of abscission, but also has a role in the regulation of contractile ring assembly (Lewellyn et al., 2011) and furrow ingression. Aurora B regulates RhoA activity indirectly in different ways (Minoshima et al., 2003; Birkenfeld et al., 2007; Touré et al., 2008) and modulates the binding of myosin to the cytoskeleton (Ozlü et al., 2010), so it will be intriguing to investigate if this essential mitotic kinase contributes in any way to this new pathway.

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