Cell fate decisions - do peroxisomes have some skin in the game?

Ulrike Gruneberg[1] and Francis Barr[2]

[1]Sir William Dunn School of Pathology and [2]Department of Biochemistry, University of Oxford.

francis.barr@bioch.ox.ac.uk

ulrike.gruneberg@path.ox.ac.uk

The concept of a checkpoint monitoring chromosome attachment to microtubules, and the discovery of the spindle assembly checkpoint proteins constituted a major milestone in understanding cell division (*1*). Chromosomes, however, are not the only cellular constituents that have to be partitioned. Membrane bounded organelles such as the endoplasmic reticulum, Golgi apparatus, endosomes and mitochondria are required for cell function, and since they are typically copied from existing structures it is necessary that both daughter cells receive a share of these components. Fuchs and colleagues provide an exciting new view of the problem of organelle inheritance. In an elegant screen for factors affecting the balance between basal keratinocyte progenitor cells and differentiated suprabasal cells in the mouse epidermis, Asare et al. identify the peroxisomal protein PEX11b as a key determinant for the normal balance between proliferating and differentiating cells. Of particular interest, knock-down of PEX11b resulted in a thinning of the epidermis. This was due to a decrease in dividing keratinocyte progenitor cells and an increase in the population of early differentiating epidermal cells that were still dividing, abnormal features often associated with cancer.

Through careful analysis of peroxisomal protein levels and enzymatic activity after PEX11b depletion, Asare et al. conclude that the alterations in keratinocyte proliferative characteristics are unlikely to be caused by a metabolic defect. Previous work also indicates that loss of PEX11b does not perturb peroxisome functions such as beta-oxidation of fatty acid or ether lipid biosynthesis but rather is required for peroxisome division (*2*). Asare et al focus on a role of PEX11b in organizing peroxisomes on the microtubules of the mitotic spindle. Peroxisomes normally associate with the spindle poles of mitotic cells and this distribution was disturbed in PEX11b depleted cells, resulting in uneven inheritance of these organelles during anaphase. Remarkably, PEX11b depletion appeared not only to affect peroxisome inheritance as such but more importantly had significant effects on cell cycle progression and cell division plane selection, providing a novel link between organelle inheritance and different aspects of mitosis. Asymmetric cell divisions with

the division planes perpendicular to the basement membrane are known to be critical for the stratification of the epidermis, and PEX11b depleted cells displayed a significantly increased number of cells with parallel instead of perpendicular divisions in epidermal progenitor cells, resulting in the observed thinner epidermis in Pex11b depleted mouse skin. The altered division plane selection in Pex11b depleted skin cells coincided with a changed distribution of the spindle pole and cortex-localized large coiled-coil protein NuMA. This is a highly significant observation since NuMa is one of the key factors determining division plane selection (*3*). NuMa anchors the dynein complexes at the cortex which generate pulling forces on astral microtubules and hence position the mitotic spindle. In PEX11b depleted epidermal cells NuMA was selectively lost from the apical cortex providing a molecular explanation for the loss of perpendicular cell divisions. Intriguingly, given that NuMA and peroxisomes both localized to the spindle pole in wild type cells, the lost cortical localization of NuMA in the absence of PEX11b may indicate a mutual dependency for localization by NuMA and peroxisomes, possibly mediated by a direct interaction between PEX11b and NuMA.

In addition to the altered angle of cell division, the time between nuclear envelope breakdown and anaphase onset in PEX11b depleted cells was significantly lengthened. This could be related to the mis-localization of NuMA, since impaired mitotic spindle formation and positioning can compromise chromosome alignment and trigger the spindle assembly checkpoint, similar to observations made for the Golgi (*4*). Alternatively, Pex11b depletion may lead to an altered peroxisome distribution that is directly sensed by the cell, by some form of organelle segregation checkpoint or position sensing machinery. To begin to resolve these questions, Asare et al. employed optogenetics to experimentally re-direct peroxisomes to different regions of the mitotic spindle upon blue light triggered association with different microtubule binding proteins. Remarkably, a mitotic delay was observed only when peroxisomes were directed to microtubule plus-ends of interpolar spindle microtubules at the spindle midzone, suggesting that indeed altered peroxisome localization is monitored by the cell. By contrast, targeting peroxisomes to microtubule minus-ends clustered at the spindle poles supported normal mitotic progression. These results provide good support for the existence of an organelle position sensing machinery linked to peroxisomes by Pex11b.

This study provides further support for the idea that organelle inheritance, mitotic spindle organization and cell differentiation are mechanistically coupled. While many details need to be elucidated, common themes relating to the different membrane

organelles are beginning to emerge from this and previous work. One of these points to the oncogenic protein kinase Aurora A mutated in a number of cancers. During spindle orientation NuMa is a target of Aurora A (*5*), which is also linked to Golgi inheritance sensing (*6-8*). Furthermore, because Aurora A activity is spatially restricted to the poles of the mitotic spindle (*9*), this could provide a simply way to determine where an organelle is relative to different parts of the mitotic spindle (Figure). Further analysis of the mechanisms of organelle inheritance in multicellular organisms will fill in the details of the basic cellular pathways needed for cell growth and division but may also deliver surprising insights into pathways altered in cancer.

1. A. Musacchio, The Molecular Biology of Spindle Assembly Checkpoint Signaling Dynamics. *Curr Biol* **25**, R1002-1018 (2015).
2. X. Li *et al.*, PEX11 beta deficiency is lethal and impairs neuronal migration but does not abrogate peroxisome function. *Mol Cell Biol* **22**, 4358-4365 (2002).
3. F. di Pietro, A. Echard, X. Morin, Regulation of mitotic spindle orientation: an integrated view. *EMBO Rep* **17**, 1106-1130 (2016).
4. G. Guizzunti, J. Seemann, Mitotic Golgi disassembly is required for bipolar spindle formation and mitotic progression. *Proc Natl Acad Sci U S A* **113**, E6590-E6599 (2016).
5. S. Gallini *et al.*, NuMA Phosphorylation by Aurora-A Orchestrates Spindle Orientation. *Curr Biol* **26**, 458-469 (2016).
6. C. Sutterlin, P. Hsu, A. Mallabiabarrena, V. Malhotra, Fragmentation and dispersal of the pericentriolar Golgi complex is required for entry into mitosis in mammalian cells. *Cell* **109**, 359-369 (2002).
7. J. H. Wei, Z. C. Zhang, R. M. Wynn, J. Seemann, GM130 Regulates Golgi-Derived Spindle Assembly by Activating TPX2 and Capturing Microtubules. *Cell* **162**, 287-299 (2015).
8. A. Persico, R. I. Cervigni, M. L. Barretta, D. Corda, A. Colanzi, Golgi partitioning controls mitotic entry through Aurora-A kinase. *Mol Biol Cell* **21**, 3708-3721 (2010).
9. A. R. Barr, F. Gergely, Aurora-A: the maker and breaker of spindle poles. *J Cell Sci* **120**, 2987-2996 (2007).

Figure legend

A depiction of the mitotic spindle with microtubules (green) and aligned chromosomes (blue). Aurora A (shaded yellow area) is localized to the poles of the mitotic spindle where its activity regulates factors, including NuMa (purple) required for spindle formation and positioning. In this model, peroxisomes (grey) within the Aurora A gradient would create a state permissive for cell cycle progression and asymmetric division. Failure to correctly localize peroxisomes, and possibly other organelles such as Golgi, results in cell cycle delays and a non-permissive state for asymmetric divisions, and ultimately skewed daughter cell fates.