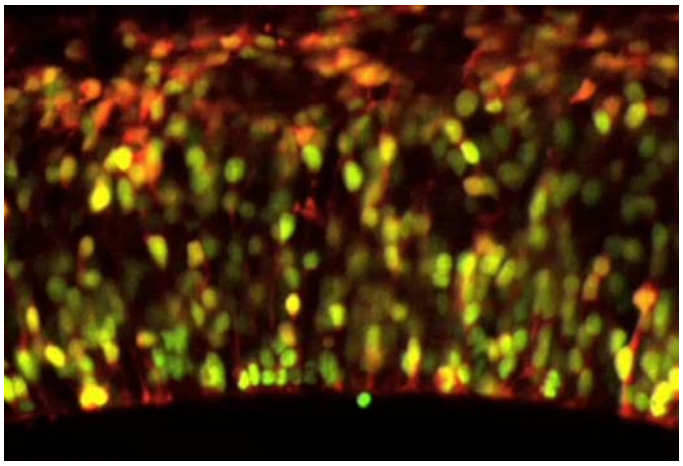
RIKEN Center for Developmental Biology (CDB)
2-2-3 Minatojima minamimachi, Chuo-ku, Kobe 650-0047, Japan

Neurorientation: Finding direction in mammalian neuroepithelial cell divisions

January 8, 2008 – When a cell divides, mitotic spindles form to segregate the replicated chromosomes and allocate the contents of the parent cell into the two daughters. The orientation of these spindles is therefore important, as it impacts directly on the cellular components that each daughter will inherit, and can influence or determine whether the division will be symmetric (producing daughters of identical cell fate) or asymmetric. Asymmetric cell division is particularly important in stem cell biology, as many types of stem and progenitor cell self-renew by generating one copy of itself along with a daughter of another fate. This is true of the cells of the neuroepithelium in mammals, which serve as progenitors that both self-renew and give rise to neurons, suggesting that the orientation of the spindle apparatus may play a significant role in ensuring asymmetric mitosis, but its contribution, if any, remains unclear.



Transverse section of neuroepithelial cells from cortical slice, showing interkinetic nuclear movement and planar division of neuroepithelial cells, which serve as neural stem cells during development. Chromosomes labeled with H2B-EGFP.
(Click image to play movie in separate window)

Daijiro Konno, Go Shioi and colleagues in the Laboratory for Cell Asymmetry (Fumio Matsuzaki; Group Director) have now shed light on the control of spindle orientation in neuroepithelial cells, and its function in maintaining the neuroprogenitor population. This work, published in *Nature Cell Biology*, identifies the gene *LGN* as a critical component for orienting the mitotic spindle so as to allow neuroepithelial cell division to proceed asymmetrically.

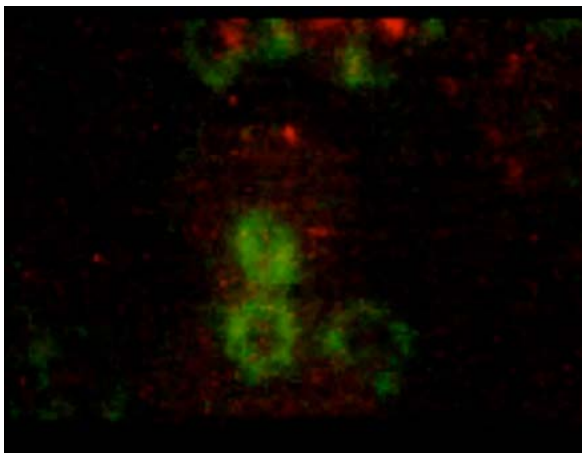
Mitotic spindle orientation has been studied extensively in the fruit fly *Drosophila melanogaster*, revealing an elaborate network of genes that steers the spindles and links them to the cell cortex. Recent work from a number of labs has suggested that similar genetics may underlie this process in mouse neuroepithelium as well. Shioi and Konno began by creating a mutant mouse lacking the gene *LGN*, a murine counterpart to the *Drosophila* gene, *Pins* (*Partner of Inscuteable*), which participates in the cortical complex responsible for spindle orientation in the fly. Looking at dividing cells in the brain preparations from the *LGN* knockouts, they found that their orientations had become randomized, rather than occurring parallel to the epithelial surface as they do in wild type animals. A similar spindle misorientation phenotype could also be produced by overexpression of *mInsc*, the mouse ortholog of *Inscuteable*, a *Drosophila* gene that functions to rotate the mitotic spindle perpendicular to the epithelial surface.

To determine whether such misorientation had an effect on daughter cell fate, the Matsuzaki group next examined the distribution of *LGN* mutant daughter cells

Contact: Douglas Sipp sipp@cdb.riken.jp TEL: +81-78-306-3043
RIKEN CDB, Office for Science Communications and International Affairs

expressing the gene *Pax6*, which normally marks apical progenitors. They found that, rather than remaining in the ventricular zone (the region nearest the apical aspect of the neuroepithelium, where cell division generally occurs), Pax6 cells were scattered more basally. This was also the case for cells overexpressing *mInsc*, suggesting linkage between the failure of the spindles to orient properly and the basal scattering of progenitors.

Apical progenitor cells are normally polarized, extending processes called “feet” from both their apical and basal surfaces. In wild type, most progenitors divide in such as way that both daughters inherit part of the mother cell’s apical foot. But a third member of the group, Atsunori Shitamukai, found that in the orientation mutants, the apical foot was segregated to only one of the daughters at a much higher frequency. In both cultured brain slice preparations and in vivo, the progenitors produced fewer apical and more non-surface progenitors on division, and as it is the non-surface progenitors that primarily generate neurons, the rate of neurogenesis was unaffected despite this change in composition.



Apical view of neural progenitor cell division. Both daughter receive apical components in the majority of divisions. Boundary between apical cells shown by ZO1:EGFP fusion protein (green). Centrosomes labeled with PACT domain-KO1 (red).
(Click image to play movie in separate window)

The larger picture that emerged from these findings was intriguing for its subtlety, as it suggests that the inheritance of the basal or apical compartment alone is not sufficient to determine cell fate of the apical progenitors; the failure to acquire an apical foot also seems to play a role in the generation of non-surface progenitors. The group’s observations also call into question some widely held views on what constitutes the normal orientation of neurogenic cell divisions, suggesting that such divisions take place within, rather than perpendicular or oblique to, the epithelial plane.

“It is predicted that randomizing the orientation of cell divisions should affect epithelial morphogenesis and other phenomena, “ notes Matsuzaki. “So the finding that LGN knockout mice, in which the orientation is indeed random, are viable came as something of a surprise to us, and highlights the robustness in the face of perturbations of mammalian development.”

Contact: Douglas Sipp sipp@cdb.riken.jp TEL: +81-78-306-3043
RIKEN CDB, Office for Science Communications and International Affairs