

JULY 15, 2005

Brain Size May Depend Upon How Neural Cells Are Cleaved

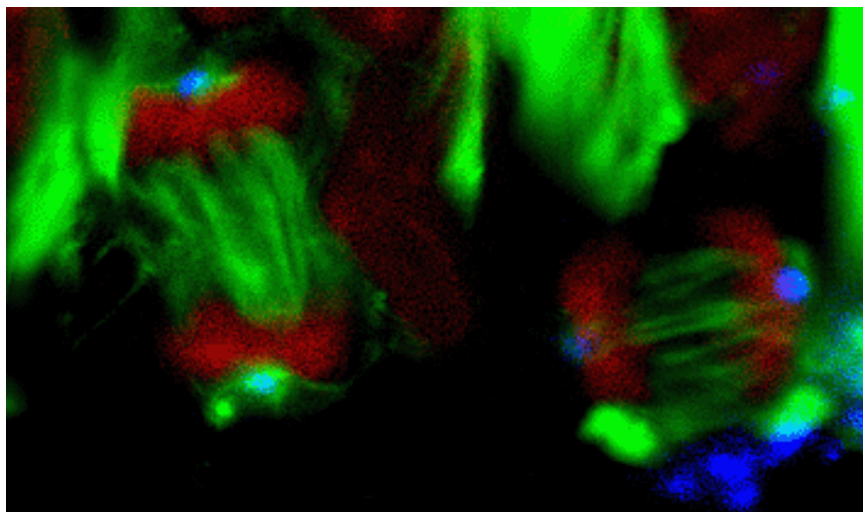


Image Title: The image shows neural progenitor cells in the proliferative ventricular zone of the developing mouse neocortex. The progenitor cells divide with the mitotic spindle (alpha-tubulin, green; pericentrin, blue; nuclei, red) perpendicular or parallel to the ventricular (apical) surface (the ventricular surface is pointing downward). The G-beta-gamma subunits of heterotrimeric G-proteins and their activator AGS3 are required for proper apical-basal division of neural progenitors and asymmetric cell fate decisions of their progeny. - Courtesy of Li-Huei Tsai, HHMI at Harvard Medical School.

Howard Hughes Medical Institute researchers have discovered a novel way in which the brain size of developing mammals may be regulated. They have identified a signaling pathway that controls the orientation in which dividing neural progenitor cells are cleaved during development.

The way these cells are sliced during development is critical because at later stages of neurogenesis, vertical cleavage gives rise to two mature neurons that are incapable of further division, while horizontal cleavage yields one neuron and one progenitor cell that can continue to support brain growth.

"This is a very important question, because how these cells divide ultimately determines the final number of cells that will be generated during brain development."

— Li-Huei Tsai

The researchers speculate that this type of regulatory decision point may play a powerful role in determining the ultimate size of the mammalian brain. Inherited disorders that cause the brain to develop too small or too large may also influence this developmental pathway.

Howard Hughes Medical Institute investigator Li-Huei Tsai and postdoctoral fellow Kamon Sanada, both at Harvard Medical School, published their findings in the July 15, 2005, issue of the journal *Cell*.

The researchers drew on studies by other researchers that showed that the orientation of cleavage planes in dividing neural progenitor cells in the neocortex determines the fate of the resulting daughter cells. However, nothing was known about the molecular signaling mechanism that regulates the decision to cleave one way or another, said Tsai.

Studies in fruit flies and roundworms had hinted that major regulatory molecules called heterotrimeric G proteins play a role in orienting the mitotic spindles that govern the orientation of cell cleavage during cell division, or mitosis.

"Based on that knowledge, we knew that heterotrimeric G proteins were very good candidates as regulators of the plane of cell division in neural precursors," said Tsai.

"And this is a very important question, because how these cells divide ultimately determines the final number of cells that will be generated during brain development," she said.

Using the embryonic mouse brain as a model, Sanada and Tsai sought to determine whether G proteins play a role in the developing mammalian brain. In their initial experiments, they impaired the function of the G $\beta\gamma$ subunits of heterotrimeric G proteins, in the developing mouse brain. They saw a dramatic interference with orientation of cell cleavage to overproduce "postmitotic" neurons—cells that could no longer divide—at the expense of progenitor cells, which could still divide. "This observation led us to want to further test whether impairment of G $\beta\gamma$ has any consequences for cell fate in division, because it has been speculated that the different division planes dictate the ultimate cell fate adoption of daughter cells," said Tsai.

To determine definitively whether impairment of G $\beta\gamma$ had a direct effect on cell fate, the researchers impaired G $\beta\gamma$ signaling in the mouse brains in utero, then isolated the progenitor cells to study the effects in vitro. Those studies

revealed that impairing $G\beta\gamma$ did result in overproduction of neurons as a result of both daughter cells adopting the neuronal fate. Thus, the researchers concluded that $G\beta\gamma$ does control the orientation of cleavage and the identity of the daughter cells—whether they will become neurons or progenitor cells.

Sanada and Tsai also sought to determine how $G\beta\gamma$ is regulated. Studies on asymmetric cell division in fruit flies and roundworms had implicated a particular class of proteins—known as AGS3 and mPins in mammals—as upstream regulators of G proteins.

The researchers' analyses showed that AGS3 is, indeed, expressed in progenitor cells. And when they “silenced” AGS3 expression in embryonic mouse brains, the resulting abnormalities mimicked those produced when $G\beta\gamma$ signaling was disrupted.

Now that the role of $G\beta\gamma$ in neural cell proliferation has been discovered, said Tsai, further studies will try to pinpoint how its signaling determines the orientation of spindles during mitosis, and thus the orientation of cell cleavage.

“While this is basic research, we do know that this mechanism is very important in determining the ultimate size of the brain,” said Tsai. “And, there are humans born with too few neurons, called microcephaly, or too many neurons, called macrocephaly. I would speculate that many of these cases are the outcome of some sort of impairment in the regulation of cell division, perhaps in the plane of division,” said Tsai.

“And with the possibility that neural stem cells may find therapeutic use, the role of G protein signaling in the differentiation of such progenitor cells is going to be a very exciting area to explore,” she said.