ESSAY: EPPENDORF & *SCIENCE* PRIZE

Making a Bigger Brain by Regulating Cell Cycle Exit

Anjen Chenn

Eppendorf and *Science* are pleased to present the prize-winning essay by Anjen Chenn, the 2002 grand prize winner of the Eppendorf and *Science* Prize for neurobiology.

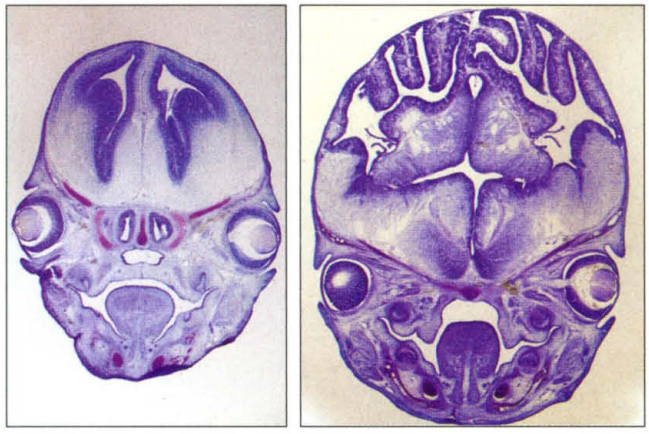
eppendorf & Science PRIZE FOR NEUROBIOLOGY

Humans possess an impressive repertoire of mental skills that include the abilities to read, write, and solve intricate problems. These skills are made possible by the cerebral cortex, the thin, layered sheet of neurons on the surface of the brain that underlies our most complex cognitive abilities. Although all mammals have cerebral cortices, the cerebral cortex in primates, especially humans, has undergone a vast expansion in size during evolution. The increase in the size of the cerebral cortex is thought to underlie the growth of intellectual capacity.

The increased size of the cerebral cortex during evolution results primarily from a disproportionate expansion of its surface area (*1*–*7*), with the appearance of folds of the cortical surface (with hills corresponding to structures known as gyri and intervening valleys called sulci) providing a means to increase the total cortical area in a given skull volume. This expansion of the length and breadth of cerebral cortex is not accompanied by a comparable increase in cortical thickness; in fact, the 1000-fold increase in cortical surface area between human and mouse is only accompanied by an approximate twofold increase in cortical thickness (*8*).

Relatively little is understood about how neural progenitors generate neurons. We performed time-lapse studies that suggested that by regulating the spatial orientation of its cell division, a precursor could divide symmetrically to generate more precursors, or asymmetrically to generate a differentiated neuron and a sister that remained in the precursor zone (*9*). These studies led us to speculate that molecules that were localized asymmetrically in precursors might be selectively inherited following an asymmetric division, and thus differentially influence the fates of the resultant daughters.

One such protein that might regulate the production of neural precursors is β-catenin, an important component of adherens junctions (*10*) that interacts with proteins of the TCF/LEF family to transduce Wnt signals (*11*). Wnts (*12*) and TCF/LEF family members (*13*, *14*) are expressed in the developing mammalian brain, and numerous studies support the role of Wnt signaling in cell fate regulation during brain development (*15*–*19*). β-Catenin has been implicated in many human cancers (*11*), including some such as medulloblastoma, where the cancer cells bear striking resemblance to neural precursor cells (*20*). We found that β-catenin is highly expressed in mammalian neural precursors, and β-catenin protein is enriched at adherens junctions at the lumen of the ventricle. These findings raised the possibility that β-catenin functions to influence cell number or cell fate decisions in the developing nervous system.



Enlarged brain and head of β-catenin transgenic animal compared with wild-type littermate control. The brains from transgenic animals (right) are enlarged overall and have a greatly increased surface area without a corresponding increase in thickness. [Adapted from figure 3 in (*22*)]

To address whether activating β-catenin signaling could regulate mammalian brain development, we generated transgenic mice expressing an engineered form of β-catenin designed to be resistant to normal cellular degradation. The gene was attached behind a promoter that directs the expression of a well-characterized neural development gene (*21*) so that it accumulated only in neuronal progenitor cells.

Transgenic embryos developed grossly enlarged brains with a considerable increase in the surface area of the cerebral cortex but without a corresponding increase in cortical thickness. The horizontal growth in length and breadth of the cortical tissue was so extensive that the normally smooth cerebral cortex of the mouse formed convoluted folds resembling the gyri and sulci of higher mammals.

By examining the expression of markers that label neural precursors and differentiating neurons, we determined that the precursor population was markedly expanded in these transgenic animals (*22*). The expression of different markers of cortical neuron populations suggested that despite the massive expansion of cortical surface area, transgenic precursors appeared to differentiate into young neurons in an approximately normal spatial pattern. Taken together, the expression studies suggested that overactivating β-catenin did not disrupt the normal developmental sequence of neuronal differentiation, and the horizontal expansion of the cortical plate was a result of an increased number of proliferative precursor cells.

What caused the expansion in the progenitor pool? Enlargement of the precursor pool in transgenic brains can result from increased mitotic rates, decreased cell death, or changes in cell fate choice (whether to differentiate or to proliferate). After examining rates of cell division and cell death, we found that the expansion of the progenitor cell population was not caused by a simple effect of β-catenin on cell division, or by decreased apoptotic cell death. Instead, we found an approximately twofold increase in the proportion of transgenic precursors that reenter the cell cycle after dividing, compared with wild-type neural precursors. Together, these studies suggest that β-catenin activation functions in neural precursors by influencing cell fate choice so that precursors reenter the cell cycle instead of differentiating.

The author is in the Department of Pathology, Northwestern University, The Feinberg School of Medicine, Chicago, IL 60611–3008, USA.

2002 Grand Prize Winner

Anjen Chenn was born in Taipei, Taiwan, and grew up in Marion, Ohio. Dr. Chenn received his bachelor's degree in biochemical sciences from Harvard University in 1990. He went on to graduate studies in the Medical Scientist Training Program at Stanford University where he joined Dr. Susan McConnell's laboratory and studied mammalian cerebral cortical development. His research on asymmetric divisions in mammalian neurogenesis was published in a first-author paper in *Cell* and was featured on the cover. He received his Ph.D. in neurosciences from Stanford University in 1996 and his M.D. in 1997. From there Dr. Chenn moved to residency training in clinical pathology at the Brigham and Women's Hospital in Boston and became board-certified in clinical pathology in 2000. During his residency training, Dr. Chenn was awarded a Howard Hughes Physician Postdoctoral Fellowship and pursued postdoctoral research in Dr. Christopher A. Walsh's laboratory at the Beth Israel Deaconess Medical Center. His postdoctoral research on genetic regulation of cerebral cortical size resulted in a first-author research article and cover figure in *Science*. Dr. Chenn is now an assistant professor in pathology at the Feinberg School of Medicine at Northwestern University in Chicago where his laboratory continues to pursue research in mammalian neural development.

Finalists

Liqun Luo, for his essay, "From Single Neuron to Neural Circuits," reporting research carried out in the Department of Biological Sciences at Stanford University. Dr Luo grew up in Shanghai, China, and earned his bachelor's degree in molecular biology from the University of Science and Technology of China. After obtaining his Ph.D. from Brandeis University and pursuing postdoctoral studies at the University of California, San Francisco, Dr. Luo started his own laboratory in December 1996. Work in his laboratory focuses on using genetic tools to understand the logic of brain wiring. Dr. Luo also teaches a course in molecular and cellular neurobiology to Stanford University undergraduate and graduate students.

Lisa Stowers, for her essay, "How Mice Detect and Respond to Pheromones," based on work done in Dr. Catherine Dulac's laboratory at Harvard University. Dr. Stowers was born in Petaluma, California, and received a B.A. in bacteriology from the University of California at Davis. In 1997 she was awarded a Ph.D. in molecular and cellular biology from Harvard University for work in Dr. John Chant's laboratory characterizing signal transduction components of mammalian cell polarity. As a postdoctoral fellow in the laboratory of Dr. Dulac, she used a molecular genetic approach to study the neurobiology of mouse behavior. This work identified the sensory neurons that respond to pheromones and illuminated the influence of the chemical environment on both the social behavior and the neuroendocrine response of the mouse. She is currently continuing this work as an assistant professor of cell biology at The Scripps Research Institute.

Thomas Thannickal, for his essay, "Human Narcolepsy as a Neurodegenerative Puzzle," based on postdoctoral research done in Dr. J. M. Siegel's laboratory at the Sepulveda VA Medical Center and University of California, Los Angeles. Dr. Thannickal was a Ph.D. student in the laboratory of Dr. V. C. Thomas at the Mahatma Gandhi University, Kerala, India, and received his Ph.D. in 1995. In 1996 he joined Dr. V. Mohan Kumar's laboratory at the All India Institute of Medical Sciences, New Delhi, where he studied the basic mechanism of sleep. Dr. Thannickal moved to the United States in 1999 and, working with Dr. Siegel, investigated the neurophysiological basis of human narcolepsy. Their work indicated that narcolepsy is associated with a neurodegenerative process that causes significant loss of hypocretin neurons.

The full text of essays by the finalists and information about applying for next year's awards can be viewed on *Science* Online at www.sciencemag.org/feature/data/prizes/eppendorf/eppenprize.shtml

Our findings support evidence suggesting that epithelial architecture and proteins at adherens junctions regulate growth control and cell proliferation (*23*). Disruptions of adherens junctions may cause misregulation and accumulation of cytoplasmic β-catenin. Our findings that β-catenin signaling can regulate the decisions of neural precursors to reenter or exit the cell cycle lend support to the possibility that β-catenin signaling may mediate the loss of growth control when adherens junctions are disrupted.

It has been proposed that subtle regulation of the decision of neural precursors to divide or differentiate can underlie the expansion of the precursor population without changing the thickness of the cortex (*5*, *24*). We found that β-catenin activation can regulate the size of the neural precursor pool by influencing the decision to divide or differentiate without increasing cell cycle rate, decreasing cell death, or grossly altering neuronal differentiation. Although larger brains can be generated in different ways as well (*25*–*29*), our findings suggest that subtle changes in the decisions of neural precursors to reenter or exit the cell cycle result in horizontal expansion of the surface area of the developing cerebral cortex without increased cortical thickness. Many questions remain about how β-catenin might regulate neural precursor decisions, and whether selective β-catenin inheritance plays a role in cell fate determination. Given its function in a wide variety of tissues, the role of β-catenin in cortical progenitors is likely to be complex. Further understanding of how the decision to divide or differentiate is regulated by β-catenin and other molecules during evolution can provide insight into the mechanisms that underlie the growth of the cerebral cortex in higher mammals.

References

1. B. L. Finlay, R. B. Darlington, *Science* **268**, 1578 (1995).
2. R. A. Barton, P. H. Harvey, *Nature* **405**, 1055 (2000).
3. D. A. Clark, P. P. Mitra, S. S. Wang, *Nature* **411**, 189 (2001).
4. V. S. Caviness Jr., T. Takahashi, R. S. Nowakowski, *Trends Neurosci.* **18**, 379 (1995).
5. P. Rakic, *Trends Neurosci.* **18**, 383 (1995).
6. H. Elias, D. Schwartz, *Science* **166**, 111 (1969).
7. H. Haug, *Am. J. Anat.* **180**, 126 (1987).
8. P. Rakic, *Science* **241**, 170 (1988).
9. A. Chenn, S. K. McConnell, *Cell* **82**, 631 (1995).
10. R. T. Cox, C. Kirkpatrick, M. Peifer, *J. Cell Biol.* **134**, 133 (1996).
11. M. Peifer, P. Polakis, *Science* **287**, 1606 (2000).
12. B. A. Parr, M. J. Shea, G. Vassileva, A. P. McMahon, *Development* **119**, 247 (1993).
13. M. Oosterwegel *et al.*, *Development* **118**, 439 (1993).
14. E. A. Cho, G. R. Dressler, *Mech. Dev.* **77**, 9 (1998).
15. R. T. Moon, J. D. Brown, M. Torres, *Trends Genet.* **13**, 157 (1997).
16. A. P. McMahon, A. Bradley, *Cell* **62**, 1073 (1990).
17. S. M. Lee, S. Tole, E. Grove, A. P. McMahon, *Development* **127**, 457 (2000).
18. J. Galceran *et al.*, *Development* **127**, 469 (2000).
19. V. Brault *et al.*, *Development* **128**, 1253 (2001).
20. R. H. Zurawel, S. A. Chiappa, C. Allen, C. Raffel, *Cancer Res.* **58**(5), 896 (1998).
21. P. J. Yaworsky, C. Kappen, *Dev. Biol.* **205**, 309 (1999).
22. A. Chenn, C. A. Walsh, *Science* **297**, 365 (2002)
23. D. Bilder, M. Li, N. Perrimon, *Science* **289**, 113 (2000).
24. V. J. Caviness, T. Takahashi, R. S. Nowakowski, *Trends Neurosci.* **18**, 379 (1995).
25. K. Kuida *et al.*, *Cell* **94**, 325 (1998).
26. R. Hakem *et al.*, *Cell* **94**, 339 (1998).
27. M. L. Fero *et al.*, *Cell* **85**, 733 (1996).
28. H. Kiyokawa *et al.*, *Cell* **85**, 721 (1996).
29. K. Nakayama *et al.*, *Cell* **85**, 707 (1996)