Fates and functions of NG2 cells in the postnatal CNS

William D Richardson PhD

Wolfson Institute for Biomedical Research, University College London

Abstract:

Oligodendrocyte precursors (OLPs, also known as NG2 cells) are generated in the ventricular zones (VZ) of the embryonic central nervous system (CNS), from the same set of neuroepithelial stem cells that generated neurons at earlier developmental times. NG2 cells subsequently proliferate and migrate widely throughout the CNS before associating with and ensheathing axons. Many NG2 cells persist in the adult CNS, where they comprise ~5% of all neural cells, raising questions about their function during adulthood. Since they can be induced to generate neurons as well as oligodendrocytes and astrocytes in culture, the idea developed that NG2 cells might behave as multipotent stem cells during normal adulthood or following CNS disease or injury. Many labs have now examined the fates of NG2 cells in the normal mouse brain or following disease or injury, using Cre-lox recombination in transgenic mice. The conclusion so far seems to be that NG2 cells overwhelmingly generate new myelinating oligodendrocytes during adulthood. Early reports that NG2 cells might generate astrocytes or neurons have not been upheld. This draws attention to the role of new myelinating cells in the healthy adult CNS. Do these myelinate previously unmyelinated axons, or do they remodel existing myelin? Are new oligodendrocytes and myelin involved in neural plasticity – for example, motor skills learning and memory? These ideas and experiments to test them will be discussed.

References:

Richardson, W.D., Young, K.M., Tripathi, R.B., and McKenzie, I. (2011). NG2-glia as multipotent neural stem cells: fact or fantasy? *Neuron* 70, 661-673.

Tripathi, R.B., Rivers, L.E., Jamen, F. Young, K.M. and Richardson, W.D. (2010). NG2 glia generate new oligodendrocytes but few astrocytes in a murine experimental autoimmune encephalomyelitis model of demyelinating disease. *J. Neurosci.* 30, 16383-16390.

Rivers, L.E., Young, K.M., Rizzi, M., Jamen, F., Psachoulia, K., Wade, A., Kessaris, N. and Richardson, W.D. (2008). PDGFRA/NG2-positive glia generate myelinating oligodendrocytes and piriform projection neurons in adult mice. *Nat. Neurosci.* 11, 1392-1401.

Curriculum Vitae

William D Richardson PhD
Wolfson Institute for Biomedical Research,
University College London

Prof William (Bill) Richardson obtained his BSc (Physics) from Manchester University in 1973 and PhD (Biophysics; 1978) from King's College London, where he examined chromatin structure in the electron microscope. His first postdoctoral position was at the National Institutes of Health USA (Child Health and Human Development), investigating adenovirus gene structure and expression, followed by a second postdoc at the National Institute for Medical Research, London, where he was involved in ground-breaking research into the mechanism of nuclear targeting of proteins (1982-1985). He continued this work for a time after moving (in 1985) to the Dept of Zoology at UCL as a new Lecturer. Soon after, he switched to work in the emerging field of neural development, a path he has followed most of his professional life. Recently, he has become intrigued by the continuing production of neurons and glia (notably myelinating oligodendrocytes) during adulthood and the potential role of this "adult development" in learning and memory. He is now Professor of Biology in the Wolfson Institute for Biomedical Research at UCL. He has published more than 100 original papers.

Molecular Mechanisms of Barrel Cortex Development

Tomomi Shimogori
RIKEN BSI

In the mouse primary somatosensory cortex (barrel cortex), thalamocortical axons (TCAs) from individual thalamic barrelloids are almost entirely confined to single barrel clusters, followed by arrangement of cortical layer IV neurons into barrel hollows and septa during early postnatal stage. Furthermore, spiny stellate neurons in barrel hollows form unidirectional dendrite toward barrel TCAs during first postnatal week for efficient synapse formation. To elucidate the molecular mechanism of unique dendrite development, we searched for genes expressed in the barrel cortex using Allen Brain Atlas. As a consequence, we identified Btbd3, BTB/POZ domain containing 3, is expressed exclusively in barrel hollow. BTB/POZ domain mediates homomeric/heteromeric dimerisation and its family member, Abrupt, controls dendrite formation in *Drosophila* (ref 1 and 2). Therefore, we tested Btbd3 function in spiny stellate dendrite formation and revealed that suppression of Btbd3 is efficient to generate more numbers of primary dendrite. We also revealed that initial expression of Btbd3 is induced by TCA innervation that suggests correct synapse formation control gene expression in postsynaptic neuron. We further tested whether this induction of Btbd3 expression is controlled by neuronal activity. However, no difference of Btbd3 expression was observed in neuronal activity suppressed somatosensory cortex. We next performed microarray analysis from neuronal activity suppressed barrel cortex and isolated molecule, which has BTB/POZ domain in its internal sequence. These results suggest that dendrite formation of spiny stellate cells is controlled by dimerization of both molecules induced by neuronal activity independent Btbd3 expression and neuronal activity dependent gene expression. Taken together, our results provide molecular framework of activity dependent/independent circuit development.

Reference

1. Li W, Wang F, Menut L, Gao FB. BTB/POZ-zinc finger protein abrupt suppresses dendritic branching in a neuronal subtype-specific and dosage-dependent manner. Neuron. 2004 43:823-34.

2. Sugimura K, Satoh D, Estes P, Crews S, Uemura T. Development of morphological diversity of dendrites in Drosophila by the BTB-zinc finger protein abrupt. Neuron. 2004 43:809-22.

CURRICULUM VITAE

Tomomi Shimogori

PERSONAL INFORMATION

Affiliation : RIKEN BSI Team Leader of Lab for Molecular Mechanisms of Thalamus Development

Address: 2-1 Hirosawa, Wako, Saitama, Japan

Phone: 81-48-467-9779

Fax: 81-48-467-9763

E-mail: tshimogori@brain.riken.jp

EDUCATION:

B.A. Hoshi College of Pharmacy, Tokyo, Japan 1993

Ph.D., Pharmaceutical Sciences, Graduate School, Chiba University, Chiba, Japan 1998

ACADEMIC APPOINTMENTS

1998 to 2004	Dept. Neurobiology, Pharmacology and Physiology, University of Chicago, USA Laboratory of Dr. Elizabeth A. Grove
2004 to 2010	RIKEN BSI Unit Leader for Shimogori Research Unit
2010 to present	RIKEN BSI Team Leader of Lab for Molecular Mechanisms of Thalamus Development

SERVICE TO PROFESSIONAL PUBLICATIONS:

2008-now Review Editor, Frontiers in Neural Circuits

2008-now Review Editor, Frontiers in Neuroanatomy Journal reviewer for J Neuroscience

PUBLICATIONS

Latest Peer Reviewed Journal Articles – selected

1. **Shimogori T***, Lee DA, Miranda-Angulo A, Yang Y, Jiang L, Yoshida AC, Kataoka A, Mashiko H, Avetisyan M, Qi L, Qian J, and Blackshaw S*. (2010) A genomic atlas of mouse hypothalamic development. *Nat Neurosci.* 13:767-75. *corresponding authors.
2. Suzuki-Hirano A, Ogawa M, Kataoka A, Yoshida AC, Itoh D, Ueno M, Blackshaw S, **Shimogori T**. (2011) Dynamic spatiotemporal gene expression in embryonic mouse thalamus. J Comp Neurol. 519; 528-43.
3. Yuge K, Kataoka A, Yoshida AC, Itoh D, Aggarwal M, Mori S, Blackshaw S, **Shimogori T**. (2011) Region-specific expression in early postnatal mouse thalamus. J Comp Neurol. 519; 544-61.
4. Matsui A, Yoshida AC, Kubota M, Ogawa M and **Shimogori T**. (2011) Mouse in utero electroporation: Controlled spatio-temporal gene transefection. J Vis Exp. 54 pii: 3024. doi: 10.3791/3024.
5. Hama H, Kurokawa H, Kawano H, Ando R, **Shimogori T**, Noda H, Fukami K, Sakaue-Sawano A, Miyawaki A. (2011) Scale: a chemical approach for fluorescence imaging and reconstruction of transparent mouse brain. doi: 10.1038/nn.2928.

Neural stem cells and early-born neurons collaborate three-dimensionally for neocortical histogenesis

Takaki Miyata, MD., Ph.D.
Nagoya University

We are asking how neural stem cells' morphology is regulated three-dimensionally and how this regulation contributes to continuous cell production and the overall brain formation. Stem cells in the mammalian brain primordia originally take a neuroepithelial structure in which their nuclei diffusely occupy the entire wall (about ten nuclei thick) of the neural tube or brain vesicle. This diffuse nuclear distribution is due to the cell cycle-dependent, to-and-fro nuclear movement (called interkinetic nuclear migration) exhibited by each neuroepithelial cell (80 μm long), which spans from the apical (inner) surface to the basal (outer) surface of the wall. When the first neuronal group comes out as a result of divisions within the initial neuroepithelium, neurons accumulate in an outer zone (1-2 cell thick) just beneath the basal lamina and stem cells become longer (90-100 μm). The elongated stem cells keep their apicobasal attachment as well as nuclear migration trajectory in a range of 80 μm (ten nuclei thick) with a basal process (~20 μm) extended. Stem cells' elongation coupled with maintenance/renewal of basal processes further continues as the wall thickens. How this elongation occurs is unknown and it is important to understand whether and if so how this phenomenon might affect stem cells' cytogenetic behavior. Through in vivo RNAi experiments and live imaging in slice culture, we found that the earliest cohort of neurons in the developing mouse neocortex may play an important role in extrinsically shaping the neural stem cells.

Miyata, T., and Ogawa, M.: Twisting of neocortical progenitor cells underlies a spring-like mechanism for daughter-cell migration. *Curr. Biol.* 17, 146-151 (2007)

Miyata, T., Kawaguchi, A., Saito, K., Kawano, M., Muto, T., and Ogawa, M.: Asymmetric production of surface-dividing and non-surface-dividing cortical progenitor cells. *Development* 131, 3133-3145 (2004)

Miyata, T., Kawaguchi, A., Okano, H., and Ogawa, M.: Asymmetric inheritance of radial glial fibers by cortical neurons. *Neuron* 31, 724-741 (2001)

Curriculum Vitae

Takaki Miyata, MD., Ph.D.

2004/1- present:

Professor, Department of Anatomy and Cell Biology, Nagoya University Graduate School of Medicine, Japan

1999/11-2003/12:

Research Scientist, Laboratory for Cell Culture Development (Masaharu Ogawa's Lab), Brain Science Institute, RIKEN, Japan

1998/10-1999/10:

Research Associate, Department of Neuroanatomy (Hideyuki Okano's Lab), Osaka University Graduate School of Medicine, Japan

1997/4-1998/9:

Postdoctoral Fellow, Department of Molecular, Cellular, and Developmental Biology (Jacqueline Lee's Lab), University of Colorado at Boulder, Colorado, USA

1994/4-1997/3:

Postdoctoral Fellow, Department of Molecular Neurobiology (Katsuhiko Mikoshiba's Lab), Institute for Medical Science, University of Tokyo, & Tsukuba Life Science Center, RIKEN Japan

1990/4-1994/3:

Graduate Student, Department of Physiology, Kochi Medical School, Japan (supervised by Masaharu Ogawa)

1988/5-1990/3:

Department of Otolaryngology and Head and Neck Surgery, Kochi Medical School Hospital

Developmental and Evolutionary Roles of a Noncoding DNA Element in Perisylvian Cerebral Cortex

Byoung-Il Bae

Children's Hospital Boston, Harvard Medical School

The Sylvian fissure (SF), dividing frontal and parietal lobes from the temporal lobe, is remarkably enlarged in humans, but is indistinct in non-primates. Despite the signal importance of perisylvian cortex, which mediates crucial human cognitive functions including language, it is completely unknown how development and evolution of these regions is regulated. Here, we identify a conserved noncoding DNA element (CNE) critical for perisylvian cortex development. Mutation of the CNE selectively and symmetrically disrupts perisylvian cortex, including "Broca's area," the primary language area. The CNE is present only in placental mammals, with its associated exon showing the signature of an ancient L4 retrotransposon insertion after divergence of placental and non-placental mammals. The CNE represents one of ~17 promoters for the *GPR56* gene, and the CNE mutation disrupts interaction with RFX transcription factors, ablating the promoter activity. GPR56, a G protein-coupled receptor essential for cortical development, mediates adhesion between cortical progenitors and the basal lamina. When fused to the beta-galactosidase gene (β-gal), the human CNE drives gene expression in mice in a sharply delineated lateral cerebral cortical zone, representing a potential orthologue of human perisylvian cortex, whereas the corresponding mouse CNE drives expression diffusely in cerebral cortex. Evolutionary differences between the mouse and human CNE also shift gene expression from apical progenitors, common to all vertebrates, to $SOX2^{+}TBR2^{-}$ outer subventricular zone (OSVZ) progenitors that are absent from non-placental vertebrates, rare in mice, and most abundant in humans. Our data show that perisylvian development requires an evolutionarily dynamic CNE, and suggest that modulation of *GPR56* expression in OSVZ progenitors by this CNE may play important roles in development and evolution of perisylvian cortex. Our study also represents a potential starting point to identify transcriptional programs regulating development of OSVZ progenitors in distinct cortical regions.

Curriculum Vitae

BYOUNG-IL BAE, Ph.D.

Harvard Medical School, Children's Hospital Boston,
Department of Genetics

EDUCATION AND TRAINING

Harvard Medical School/Children's Hospital Boston/HHMI Boston, MA
- Postdoctoral fellow in Neurology 09/2006 – Present
- Advisor: Dr. Christopher A. Walsh

Johns Hopkins University School of Medicine Baltimore, MD
- Postdoctoral fellow in Neuroscience 09/2005 –09/2006
- Advisor: Dr. Solomon H. Snyder

Johns Hopkins University School of Medicine Baltimore, MD
- Ph.D. in Neuroscience 09/1999 –09/2005
- Advisor: Dr. Solomon H. Snyder

Seoul National University Seoul, Republic of Korea
- B.S. in Molecular Biology 03/1994 –02/1998
- *Magna Cum Laude* (2nd out of 19)

HONORS AND FELLOWSHIPS

- Leonard and Isabelle Goldenson Research Fellowship, Department of Neurobiology, Harvard Medical School, 2011-2013.
- Harvard Stem Cell Institute/NIH Training Grant (T32 HL087735-01A1), 2009-2011.
- Harvard Medical School Genetics/NIH Training Grant (T32), 2009 (declined).
- Children's Hospital Boston/NIH Developmental Neurology Training Grant (T32 NS007473), 2008-2009.
- Korea Foundation for Advanced Studies Predoctoral Fellowship, 1999-2004.
- The Glenn/American Federation for Aging Research Scholarships, 2004 (honorable mention).
- Howard Hughes Medical Institute Predoctoral Fellowship, 2000 (honorable mention).
- Korea Foundation for Advanced Studies Undergraduate Fellowship, 1996-1998.
- Seoul National University Scholarship, 1994-1995.

SELECTED PUBLICATIONS

1. **Bae, B.-I.**, Tietjen, I., Evrony, G.D., Chang, B.S., Barkovich, A.J., Murayama, A., Shimada, H., Asare, E., Atabay, K.D., Im, K., Grant, P.E., Crosier, M., Lisgo, S.N., Lindsay, S., Johnson, M.B., Šestan, N., Topçu, M., Politsky, J., Okano, H., Piao, X., and Christopher A. Walsh. Developmental and Evolutionary Roles of a Noncoding DNA Element in Perisylvian Cerebral Cortex. (in preparation)
2. Sen, N., Hara, M.R., Kornberg, M.D., Cascio, M.B., **Bae, B.-I.**, Shahani, N., Thomas, B., Dawson, T.M., Dawson, V.L., Snyder, S.H., and Sawa, A. (2008) Nitric oxide-induced nuclear GAPDH activates p300/CBP and mediates apoptosis. *Nat. Cell Biol.* 10: 866-873.
3. **Bae, B.-I.**, Hara, M.R., Cascio, M.B., Wellington, C.L., Hayden, M.R., Ross, C.A., Ha, H.C., Li, X.-J., Snyder, S.H., and Sawa, A. (2006) Mutant Huntingtin: Nuclear Translocation and Cytotoxicity Mediated by GAPDH. *Proc. Natl. Acad. Sci. U S A* **103**(9):3405-3409.
4. **Bae, B.-I.**, Xu, H., Igarashi, S., Fujimuro, M., Agrawal, N., Taya, Y., Hayward, S.D., Moran, T.H., Ross, C.A., Montell, C., Snyder, S.H., and Sawa, A. (2005) p53 Mediates Cellular Dysfunction and Behavioral Abnormalities in Huntington's Disease.
   Neuron **47**(1):29-41. This work was previewed in *Neuron* (2005) **47**(1):1-3, and highlighted in *Nature* (2005) **436**(7048):154-155, *Chemical & Engineering News* (2005) **53**(29):9, *The Lancet Neurology* (2005) **4**(9):528-529 and *Science* (2005) **310**(5745):43-45.
5. Baranano, D.E., Wolosker, H., **Bae, B.-I.**, Barrow, R.K., Snyder, S.H., and Ferris, C.D. (2000) A Mammalian Iron ATPase Induced by Iron. *J. Biol. Chem.* **275**(20):15166-73.

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Neural Development and Disorder from a viewpoint of stem cells

Hideyuki Okano
Chair of Keio University Graduate School of Medicine
Professor, Keio University School of Medicine
Team Leader, RIKEN-Keio Univ. Joint Research Laboratory

In the human brain, there are structures that have been conserved through evolution, as well as structures that are unique to only primates, acquired through the enlargement of the cerebral cortex. The clarification of these types of structure and their fundamental brain functions is required to properly understand the normal brain functioning of humans, as well as mental health, and illnesses caused by abnormal brain functioning. Existing research on the operating principles of the brain, however, has suffered from the biases and limitations of information derived from animal experiments. Many were optimistic that the complementary nature of genetic engineering techniques, which focus on rodents and fish, and cognitive neuroscience techniques, which focus on primates, would lead to progress in this area. However, results have been disappointing, with few practical or theoretical connections between these techniques having developed.

Recently, however, a connection has finally been made with the success of our team in creating the world's first transgenic primate using marmosets. This technological breakthrough promises to trigger a huge paradigm shift by enabling researchers to analyze both brain structures that are conserved through evolution as well as brain structures, acquired through the enlargement of the cerebral cortex, that are unique to non-human primates and humans.

In this talk, I will present not only marmoset research, but also patient-derived iPS cell results, discussing neural development and regeneration from a viewpoint of stem cells.

Reference

1. Sasaki E, Suemizu H, Shimada A, Hanazawa K, Oiwa R, Kamioka M, Tomioka I, Sotomaru Y, Hirakawa R, Eto T, Shiozawa S, Maeda T, Ito M, Ito R, Kito C, Yagihashi C, Kawai K, Miyoshi H, Tanioka Y, Tamaoki N, Habu S, Okano H, Nomura T. Generation of transgenic non-human primates with germline transmission. Nature. 2009;459:523-7.
2. Okano H, Temple S. Cell types to order: temporal specification of CNS stem cells. Curr Opin Neurobiol. 2009;19:112-9

CURRICULUM VITAE

Hideyuki Okano
Chair of Keio University Graduate School of Medicine
Professor, Keio University School of Medicine
Team Leader, RIKEN-Keio Univ. Joint Research Laboratory

Profile

Hideyuki Okano was born in 1959 and started his research carrier in 1983 with the molecular genetic studies of myelination and hereditary dysmyelinating disease model mice at Department of Physiology, Keio University School of Medicine, after graduating from the same school. In 1988, he received Ph.D. from Keio University with this theme. He performed pioneering work on the molecular genetic study of mammalian neural development, which led to subsequent extensive studies of neural development and regeneration as follows. From 1989 to 1991, he began the investigation of molecular neurobiology of *Drosophila* as a Post-Doctoral fellow in Department of Biological Chemistry in Johns Hopkins University School of Medicine in U.S.A. From 1992 to 1994, he worked in Institute of Medical Science, University of Tokyo, where he started molecular biology of mammalian neural development and stem cells. He was promoted to Professor at University of Tsukuba in 1994 and became Head of the laboratory of Molecular Neurobiology at Institute of Basic Medical Science. He moved to Department of Neuroanatomy, Osaka University Medical School in 1997, where he started the investigation on the regeneration of adult mammalian central nervous system. He moved to Department of Physiology, Keio University School of Medicine in 2001, where he educated many students in the field of neuroscience and regenerative medicine. He was awarded Naka-akira Tsukahara Award in 2001, Distinguished Scientific Award from University of Catania School of Pharmacy in 2004, Minister Award of Ministry of Education, Culture, Sports, Science and Technology in 2006, and Medal with Purple Ribbon in 2009 for his pioneering studies on the neural development and regeneration. 2007 ~ present, he acts as a Chair of Keio University Graduate School of Medicine. From 2011, he added the role of the Team Leader, RIKEN-Keio Univ. Joint Research Laboratory, RIKEN Brain Science Institute.