BIOGRAPHICAL SKETCH

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NAME: Kuo, Chay Titus

eRA COMMONS USER NAME (credential, e.g., agency login): chaykuo

POSITION TITLE: Associate Professor of Cell Biology, Neurobiology, Duke University School of Medicine

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable. Add/delete rows as necessary.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
M.I.T., Cambridge, MA	BS	02/1993	Architecture
University of Chicago, Chicago, IL	PhD	12/1997	Genetics
University of Chicago, Chicago, IL	MD	08/2002	Medicine
HHMI / University of California, San Francisco	Postdoctoral	10/2002-09/2007	Neurobiology
NIH / NIEHS Laboratory of Neurobiology	Visiting Scientist	03/2013-08/2013	Neurophysiology

A. Personal Statement

The focus of my laboratory is to better understand how regenerative capacity in the brain contributes to normal physiology and tissue remodeling after injury. To achieve this, we use the rodent lateral ventricular postnatal neurogenesis as a model system. We are particularly interested in molecular and neuronal activity-dependent mechanisms regulation postnatal neural stem cell (NSC) homeostasis, and how injuries and disease can modify these pathways. My scientific background has prepared me well to direct a group of scientists working towards a common goal. As an MD/PhD student at the University of Chicago, I studied molecular control of T lymphocyte selection and activation. From this solid base in mouse genetics and biochemistry, I was able to branch out into neurobiology as a postdoctoral fellow at UCSF, in the laboratories of Drs. Yuh-Nung and Lily Jan, working on NSCs and dendritic remodeling/integration using both mice and *Drosophila* as model systems. To further enrich my scientific training, since becoming PI in 2007 I have 1) taken the summer Ion Channel Physiology Course at Cold Spring Harbor Laboratory in 2010 to incorporate new electrophysiology techniques into our research, and 2) completed 6 months of training as Visiting Scientist in Dr. Jerry Yakel's laboratory at the NIH focusing on cholinergic neurophysiology. This diverse training experience has impressed upon me the need for effective collaborations between colleagues with different expertise in biomedical sciences. We are making substantial progress pioneering new live-imaging platforms and genetic strategies that have enabled us to modulate and uncover novel principles behind postnatal NSC interactions with their native niche, as well as with local neural circuits. They have led to several concept changing discoveries. In summary, I have a demonstrated record of providing collegial environment for study, mentorship, and sound scientific research toward discoveries that will enrich our field.

a. Kuo, CT, Mirzadeh, Z, Soriano, M, Rasin, M, Wang, D, Shen, J, Sestan, N, Garcia-Verdugo, J, Alvarez-Buylla, A, Jan, LY, and Jan, YN. (2006). Postnatal deletion of Numb/Numblike reveals repair and remodeling capacity in the subventricular neurogenic niche. **Cell** 127:1253-64. (*Research highlights in Nature 444: 975*) PMC1876765
b. Paez-Gonzalez, P, Abdi, K, Luciano, D, Liu, Y, Soriano-Navarro, M, Rawlins, E, Bennett, V, Garcia-Verdugo, J, and Kuo, CT. (2011) Ank3-dependent SVZ niche assembly is required for the continued production of new neurons. **Neuron** 71:61-75. (*Cover story*) PMC3134799
c. Benner, EJ, Luciano, D, Jo, R, Abdi, K, Paez-Gonzalez, P, Sheng, H, Warner, DS, Liu, C, Eroglu, C, and Kuo, CT. (2013) Protective astrogenesis from the SVZ niche after injury is controlled by Notch modulator Thbs4. **Nature** 497:369-73. (*Top story, Neural Cell News 7.17*) PMC3667629
d. Paez-Gonzalez, P, Asrican, B, Rodriguez, E, and Kuo, CT. (2014) Identification of distinct $ChAT^+$ neurons and activity-dependent control of postnatal SVZ neurogenesis. **Nat. Neurosci.**, 17: 934-42. (*Cover story; News & Views in Nat. Neurosci. 17: 897-8*) PMC4122286

B. Positions and Honors

Positions and Employment

1993-2002	MD / PhD Student, Pritzker School of Medicine / Committee on Genetics, University of Chicago. Advisor: Jeffrey M. Leiden, M.D., Ph.D.
2002-2007	Postdoctoral Researcher, Howard Hughes Medical Inst., Department of Physiology, University of California, San Francisco. Advisors: Yuh-Nung and Lily Jan, Ph.D.
2007-2015	Assistant Professor (primary), Department of Cell Biology, Duke Univ. School of Medicine
2008-2015	Assistant Professor (2ndary), Deptartment of Neurobiology, Duke Univ. School of Medicine
2008-	Member, Preston Robert Tisch Brain Tumor Center, Duke University Medical Center
2009-	Member, Duke Institute for Brain Sciences, Duke University
2013	Visiting Scientist, Laboratory of Neurobiology, NIH / NIEHS. Host: Jerrel L. Yakel, PhD
2015-	Associate Professor (tenured), Department of Cell Biology, Duke Univ. School of Medicine
2015-	Associate Professor (2ndary), Department of Neurobiology, Duke Univ. School of Medicine

Other Experience and Professional Memberships

2007-	Member, Society for Neuroscience
2009-	Member, American Society for Cell Biology
2009-	Faculty of 1000 Member, Neurobiology of Disease & Regeneration Section
2016-	Editorial board member, Open Biology
2014-16	Ad hoc grant reviewer, NIH Neurogenesis and Cell Fate Study Section
2016-22	Standing member, NIH Neurogenesis and Cell Fate Study Section
2010-	Ad hoc grant reviewer, Medical Research Council, United Kingdom
2015-	Ad hoc grant reviewer, DFG Research Foundation, Germany
2008-	Admissions Committee, Medical Scientist Training Program (MSTP), Duke University
2013-	External Advisory Board, NICHD MD/PhD Training Program, University of Chicago
2014-16	Member, Duke University Faculty Academic Council

Ad Hoc Journal Referee

Nature; Nature Neuroscience; Neuron; Cell Stem Cell; Current Biology; Cell Reports; Journal of Neuroscience; Journal of Clinical Investigation; Journal of Experimental Medicine; Development; Glia; Molecular Biology of the Cell; Nature Methods; JoVE; PLoS One

Honors and Awards

1996	Francis L. Lederer MD/PhD Scholar, University of Chicago
1998	Harold Lamport Award - Best Dissertation in Biological Sciences, University of Chicago
2000	NIH Growth & Development Medical Scientist Training Fellowship, University of Chicago
2002	HHMI Postdoctoral Fellowship, UCSF
2006	California Institute of Regenerative Medicine Postdoctoral Scholar, UCSF
2008	Sontag Foundation Distinguished Scientist Award
2008	NIH Director's New Innovator Award
2008	David & Lucile Packard Fellowship
2009	Basil O'Connor Starter Scholar Award
2010	Alfred P. Sloan Research Fellowship
2010	George W. Brumley Jr. Assistant Professor, Duke University
2010	Kavli Frontiers Fellow, National Academy of Sciences
2015	Ruth & Morris Williams Faculty Research Prize, Duke University

C. Contribution to Science

1. Developing a key genetic tool for the postnatal neurogenesis field

To understand how newborn cells in the brain contribute to remodeling after injury, I focused on the subventricular zone (SVZ) niche of the lateral ventricles in the postnatal mouse brain, an area that contains a self-renewing population of neural stem cells (NSCs) generating new interneurons. I wanted to develop a genetically tractable model of injury response in the mammalian brain that is highly reproducible. Back in 2002, there were many controversies about adult neurogenesis in the mammalian brain, starting with the exact identity of NSCs. I wondered if there was a way to visualize adult NSCs and to identify their progeny unambiguously in the postnatal rodent brain. To solve this problem, I generated what is now

widely believed the first tamoxifen-inducible *nestin-CreER* mouse line, which specifically and efficiently deletes floxed target genes in postnatal NSCs after tamoxifen injection. Upon validating the specificity of this genetic tool, I deleted Numb and Numblike (related molecules that are critical regulators of embryonic neurogenesis) from the postnatal SVZ niche. To our knowledge this was the first study to successfully disrupt mammalian postnatal neurogenesis using a genetic strategy in vivo. These experiments not only revealed previously unknown roles for Numb/Numblike in maintaining SVZ niche homeostasis, they also showed for this first time that this stem cell microenvironment has considerable plasticity and can repair local damage to the brain. Since our proof-of-principle publication in 2006 (below), the *nestin-CreER* genetic strategy has become a widely-used standard in the postnatal/adult neurogenesis field.

a. Kuo, CT, Mirzadeh, Z, Soriano, M, Rasin, M, Wang, D, Shen, J, Sestan, N, Garcia-Verdugo, J, Alvarez-Buylla, A, Jan, LY, and Jan, YN. (2006). Postnatal deletion of Numb/Numblike reveals repair and remodeling capacity in the subventricular neurogenic niche. **Cell** 127:1253-64. (*Research highlights in Nature 444: 975*) PMC1876765
b. Pan, YW, Chan, G, Kuo, CT, Storm, DR, and Xia, Z. (2012) Inhibition of adult neurogenesis by inducible and targeted deletion of ERK5 mitogen-activated protein kinase specifically in adult neurogenic regions impairs contextual fear extinction and remote fear memory. **J. Neurosci.** 32: 6444-55. PMC3363363
c. Wang, W, Pan, YW, Wietecha, T, Zou, J, Abel, GM, Kuo, CT, and Xia, Z. (2013) Extracellular signal-regulated kinase 5 (ERK5) mediates prolactin-stimulated adult neurogenesis in the subventricular zone and olfactory bulb. **J. Biol. Chem.** 288: 2623-31. PMC3554929
d. Dieni, CV, Panichi, R, Aimone, JB, and Kuo, CT, Wadiche, JI, and Overstreet-Wadiche, L. (2016) Low excitatory innervation balances high intrinsic excitability of immature dentate neurons. **Nat. Comm.** doi: 10.1038/ncomms11313. PMC4843000

2. Identifying niche and neural-circuit control of postnatal NSC fate choices before and after injury

Neurogenesis during development represents a transient state. However, during adult neurogenesis in the rodent brain, production of new neurons is continuous. How is adult neurogenesis sustained? Adult NSCs isolated and cultured in a dish do not continue to make new neurons: what is so special about the brain that adult NSCs maintain their neurogenic capacity? If we can understand how neurogenesis is sustained, we may reveal its functional importance to the brain in health and after injury. To make these questions tractable, we generated the first inducible genetic model to specifically block ependymal niche formation postnatally, and concurrently developed the first in vitro self-assembly assay for ependymal niche progenitors. We discovered that: **i**) the ependymal niche controls neurogenesis vs. astrogenesis fate choice. When the mature ependymal niche is disrupted in vivo, we found that instead of new neurons, astrocyte clusters were made. This was the first study to reveal not only how the SVZ niche is constructed postnatally, it also demonstrated that ependymal organizational cues are required to sustain new neuron production in the adult rodent brain.

ii) We discovered that SVZ niche-generated astrocytes express high levels of Thrombospondin-4 ($Thbs4^{hi}$), a secreted homopentameric glycoprotein, in contrast to cortical astrocytes which are $Thbs4^{low}$. We found that cortical injury initiates a marked increase in $Thbs4^{hi}$ astrocyte production from the SVZ niche that then homed in on the injured cortex. $Thbs4^{KO/KO}$ animals showed severe defects in cortical injury-induced SVZ astrogenesis, instead producing DCX^{+} neuroblasts that migrated to the injured areas. Consequently, this alteration in cellular response resulted in abnormal glial scar formation after injury, and significantly increased microvascular hemorrhage into the brain parenchyma of $Thbs4^{KO/KO}$ animals. It is to our knowledge the first time that astrogenesis from the postnatal SVZ niche has been described.

iii) Over the last 4 years we have completed a difficult chemical screen using intact SVZ niche brain slices, and found unexpectedly that cholinergic modulators have robust effects on SVZ neurogenesis. In search of potential sources for neurotransmitter acetylcholine (ACh) in the niche, we uncovered direct cholinergic innervation from novel subependymal $ChAT^{+}$ (subep-$ChAT^{+}$) neurons. This previously undescribed subpopulation of cholinergic neurons released ACh into the niche in activity-dependent fashion, resulting in rapid NSC electrical responses necessary and sufficient to control SVZ neurogenic proliferation. Contrary to the widely-held view that adult neurogenesis is primarily directed by stem-cell intrinsic and local signals, we have discovered a previously undescribed gateway showing neural circuit activity patterns can directly control adult neurogenesis.

a. Paez-Gonzalez, P, Abdi, K, Luciano, D, Liu, Y, Soriano-Navarro, M, Rawlins, E, Bennett, V, Garcia-Verdugo, J, and Kuo, CT. (2011) Ank3-dependent SVZ niche assembly is required for the continued

production of new neurons. **Neuron** 71:61-75. (*Cover story; Top story, Neural Cell News 5.27*) PMC3134799

b. Benner, EJ, Luciano, D, Jo, R, Abdi, K, Paez-Gonzalez, P, Sheng, H, Warner, DS, Liu, C, Eroglu, C, and Kuo, CT. (2013) Protective astrogenesis from the SVZ niche after injury is controlled by Notch modulator Thbs4. **Nature** 497:369-73. (*Top story, Neural Cell News 7.17*) PMC3667629

c. Paez-Gonzalez, P, Asrican, B, Rodriguez, E, and Kuo, CT. (2014) Identification of distinct $ChAT^+$ neurons and activity-dependent control of postnatal SVZ neurogenesis. **Nat. Neurosci.**, 17: 943-42. (*Cover story; News & Views in Nat. Neurosci. 17: 897-8; Top story, Neural Cell News 8.21*) PMC4122286

d. Asrican, B, Paez-Gonzalez, P, Erb, J, and Kuo, CT. (2016) Cholinergic circuit control of postnatal neurogenesis. **Neurogenesis** doi: 10.1080/23262133.2015.1127310. (*Peer reviewed*) *PMC# in process*.

e. Adlaf, E, Mitchell-Dick, A, and Kuo, CT. (2016) Discerning neurogenic vs. non-neurogenic postnatal lateral ventricular astrocytes via activity-dependent input. **Front. Neurosci.** doi: 10.3389/ fnins.2016.00111. (*Peer reviewed*) PMC4805585

3. Developing a new system to study neuronal dendrite innervation of changing sensory fields

Back in 2002 it was largely unknown whether neuronal dendrites can regenerate after damage, and re-innervate a changed sensory field. To make this problem tractable, I looked into *Drosophila* metamorphosis, where a fully formed nervous system in the larvae undergoes structural changes to innervate the adult fly. There I made the unexpected discovery that a class of sensory neurons can sever all of their dendrites during metamorphosis while maintaining their axons, and subsequently re-grow their dendritic arbors. I completed a screen for degradation mechanisms and further uncovered that using a balance between the ubiquitin-proteasome system and the apoptotic machinery, these neurons can selectively activate caspases just in their dendrites during pruning while preserving their soma and axons. This was the first identification of caspase function during dendrite remodeling (later confirmed to be conserved in mammals). After pruning, these sensory neurons regrow dendrites during metamorphosis that innervate the adult sensory field, but the molecular mechanisms remained unknown. Performing a GFP-expression screen, we identified *Cysteine proteinase-1* (*Cp1*) as a hormonally-responsive gene that is required for dendrite regrowth after pruning, but surprisingly not during embryonic development. We found that *Cp1* controls context-dependent production of a truncated Cut homeodomain transcription factor with altered nuclear localization and function. This process revealed a molecular pathway whereby hormonal signals, acting through a protease to modify a key transcription factor function, allow a single neuron to pattern dendritic arbors to innervate two distinct sensory fields. These studies opened up a new experimental area to examine molecular mechanisms controlling dendrite pruning and regeneration.

a. Kuo, CT, Jan, LY, and Jan, YN. (2005) Dendrite-specific remodeling of *Drosophila* sensory neurons requires matrix metalloproteases, ubiquitin-proteasome, and ecdysone signaling. **Proc. Natl. Acad. Sci. USA** 102:15230-5. PMC1242853

b. Kuo, CT, Zhu, SJ, Younger, S, Jan, LY, and Jan, YN. (2006) Identification of E2/E3 ubiquitinating enzymes and caspase activity regulating *Drosophila* sensory neuron dendrite pruning. **Neuron** 51: 283-90. (*Research highlights in Nature Rev. Neurosci. 7: 685*.)

c. Lyons, GR, Andersen, RO, Abdi, K, Song, WS, and Kuo, CT. (2014) *Cysteine proteinase-1* and Cut protein isoform control dendritic innervation of two distinct sensory fields by a single neuron. **Cell Reports**, 6:783-91. (*Cover story*) PMC4237277

4. Identifying novel Kruppel-like (KLF2) transcription factor and active control of T cell quiescence

After the first year of medical school at the University of Chicago (I entered in 1993 as a straight MD student), I joined its MD/PhD program. My main graduate thesis project was to identify potentially new Kruppel-like Factor (KLF) family members expressed in T lymphocytes, based on the first identified EKLF transcription factor in red blood cells (now renamed KLF1). I cloned LKLF (renamed KLF2) and found that its expression is highly induced during the terminal selection process of T lymphocytes in the thymus. I mapped the *LKLF* genomic locus by identifying overlapping phage clones in a library, designed and built the gene targeting construct, grew and selected mouse embryonic stem (ES) cell clones, generated chimera mice and breed them to germ-line. My subsequent analyses showed that LKLF actively maintained the quiescent phenotype of circulating peripheral T cells. The widely held view at the time was

that quiescent T cells represent the default/basal state, but our results demonstrated that this quiescent state needs to be actively programmed and maintained, a paradigm changing finding.

a. Kuo, CT, Veselits, ML, and Leiden, JM. (1997) LKLF: A transcriptional regulator of single positive T cell quiescence and survival. **Science** 277:1986-1990. (*Perspectives in Science 277: 1950*.)
b. Kuo, CT, Veselits, ML, Barton, KP, Lu, MM, Clendenin, C, and Leiden, JM. (1997) The LKLF transcription factor is required for normal tunica media formation and blood vessel stabilization during murine embryogenesis. **Genes & Dev.** 11: 2996-3006. (*Cover story*) PMC316695.
c. Kuo, CT and Leiden, JM. (1999) Transcriptional regulation of T lymphocyte development and function. **Annu. Rev. Immunol.** 17: 149-187. (*Peer reviewed*)
d. Buckley, AF, Kuo, CT, and Leiden, JM. (2001) Transcription factor LKLF is sufficient to program T cell quiescence via a c-Myc-dependent pathway. **Nature Immunol.** 2:698-704. (*News and Views in Nature Immunol. 2: 667-8*.)

Complete List of Published Work in MyBibliography:
http://www.ncbi.nlm.nih.gov/sites/myncbi/10SieZnwRzrkB/bibliography/43857592/public/?sort=date&direction=ascending

D. Research Support

Ongoing Research Support

NIH R01 NS078192 Kuo (PI) 03/01/12 – 02/28/17
Ependymal control of new neuron production in the adult brain.
The goal of this project is to determine the functional importance of ependymal cell organization and homeostasis in controlling/sustaining new neuron production in the adult rodent brain.

NIH R01 MH105416 Kuo (PI) 11/19/14 – 10/31/19
Control of postnatal SVZ neurogenesis via cholinergic circuit activity.
The goal of this project is to anatomically and functionally map a novel cholinergic neuronal circuit from the lateral ventricular neurogenic niche in the postnatal rodent brain.

The March of Dimes Kuo (PI) 06/01/13 – 05/31/19
Investigating the mechanisms regulating brain ventricular wall maturation.
The goal of this project is to understand if defects in brain ventricular wall maturation are major causes of congenital hydrocephalus/ventriculomegaly.

David & Lucile Packard Foundation Kuo (PI) 11/01/08 – 10/31/17 (no cost extension)
Using live imaging to discover new pathways regulating postnatal neurogenesis.
The goal of this proposal is to develop a new high-throughput assay to understand cell-cell interactions within a tissue stem cell niche.

Completed Research Support

The March of Dimes Kuo (PI) 02/01/09 – 05/31/16
Investigating the mechanisms regulating brain ventricular wall maturation and hydrocephalus.

1DP2 OD004453 Kuo (PI) 09/01/08 – 08/30/13
NIH Director's New Innovator Program
Discovering pathways regulating neurogenesis and brain remodeling after injury.

The Sontag Foundation Kuo (PI) 10/01/08 – 09/30/13
Do postnatal neural stem cells cause brain tumors? Targeting the pathways regulating stem cell homeostasis.

Alfred P. Sloan Foundation Kuo (PI) 09/01/10 – 08/30/13
Understanding mechanisms regulating dendritic integration.