BIOGRAPHICAL SKETCH

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NAME: Holzbaur, Erika

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POSITION TITLE: William Maul Measey Professor of Physiology

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE (if applicable)	Completion Date MM/YYYY	FIELD OF STUDY
College of William and Mary, Williamsburg, VA	BS	05/1982	Chemistry/History
Pennsylvania State University, University Park, PA	PhD	04/1987	Biochemistry
Pennsylvania State University, University Park, PA	Postdoctoral scientist	10/1988	Molecular Biology
Worcester Foundation for Experimental Biology, Shrewsbury, MA	Postdoctoral fellow	02/1992	Cell Biology

A. PERSONAL STATEMENT

My laboratory is focused on understanding the dynamics of organelle motility along the cellular cytoskeleton, driven by microtubule- and actin-based motors. This motility is required to drive the active transport of vesicles and organelles over distances of up to 1 meter along the axons and dendrites of neurons. We are interested in the mechanisms leading to coordinated motor activity during vesicular transport, including motor recruitment, force generation, and regulation. Our work over the years has established that opposing dynein and kinesin motors are simultaneously bound to organelles moving along the axon, and are regulated in a cargo-specific manner by effectors and scaffolding proteins. Further, our work has identified distinct regulatory zones within neurons where the initiation or termination of cargo motility is modulated by microtubule +TIPs, post-translational modifications of the microtubule, and cargo-switching at intersections between microtubules and actin filaments.

We are also interested in the causative role of defective axonal transport on neurodegeneration. We are exploring the underlying mechanisms, involving either pathogenic alterations in the motors or their regulation. These studies have led us to investigate the dynamics of autophagy and mitophagy, including cargo recognition and autophagosome biogenesis. We have identified key roles for optineurin and the regulatory kinase TBK1 in the clearance of damaged mitochondria, downstream of PINK1 kinase and the E3 ubiquitin ligase parkin. As mutations in optineurin and TBK1 cause familial ALS, and mutations in PINK1 and parkin cause familial Parkinson's disease, this discovery demonstrates that the failure to clear damaged mitochondria via mitophagy is likely to contribute to the pathogenesis observed in multiple forms of neurodegeneration, a hypothesis currently under investigation in my lab.

My lab has developed a significant record of contributions to the cellular and biophysical analysis of molecular motors, combined with expertise in the mechanistic analysis of organelle trafficking and autophagy in neurons and the pathogenesis of neurodegenerative disease. We use live cell imaging of primary neurons, in vitro reconstitution with single molecule resolution, super-resolution microscopy, and mouse models of neurodegeneration to address questions at the forefront of neuronal cell biology. Together, this expertise will allow us to continue to build on our track record of making novel contributions to the field (139 publications in PubMed; h-index=65 on Google Scholar). However, I am most proud of the outstanding accomplishments of predoctoral and postdoctoral trainees (21 and 19, respectively) from my lab, who have gone on to tenure-track positions at Yale, NIH, RPI, McGill University, Tel Aviv University, the University of Massachusetts, the University of Sheffield, the University of Pennsylvania, and Washington University in St. Louis.

B. POSITIONS AND HONORS

Positions and Employment

1982 - 1987 Graduate Student, Pennsylvania State University, University Park, PA
1988 - 1992 Postdoctoral Fellow, Worcester Foundation for Experimental Biology, Shrewsbury, MA
1992 - 1998 Assistant Professor of Biochemistry, University of Pennsylvania, Philadelphia, PA
1998 - 2001 Associate Professor of Biochemistry, University of Pennsylvania, Philadelphia, PA
2002 - 2004 Associate Professor of Physiology, University of Pennsylvania Perelman School of Medicine
2004 - present Professor of Physiology, University of Pennsylvania Perelman School of Medicine
2016 - present Appointed to the William Maul Measey Chair of Physiology

Professional Memberships

American Society for Cell Biology, Biophysical Society, Society for Neuroscience, Pennsylvania Muscle Institute

Honors

1978 Thomas J. Watson National Merit Scholarship
1981 Phi Beta Kappa, College of William and Mary
1982 BS with High Honors, College of William and Mary
1983 Frank Malette Award for Graduate Distinction in Biochemistry, Pennsylvania State University
1984 Pennsylvania State University Graduate School Fellowship, Pennsylvania State University
1989 NIH Postdoctoral Fellowship, NINDS
1995 Pfizer Award for Research Excellence, University of Pennsylvania
1996 Established Investigator Award, American Heart Association
1999 Editorial Board Member, Journal of Biological Chemistry
2000 Keith R. Porter Fellow, Porter Foundation
2002 Chair, CDF-4 Study Section, NIH, 2002-2004
2003 Meeting Co-Organizer, Integration of the Cytoskeleton, Columbia University
2005 Associate Editor, Molecular Biology of the Cell
2006 Meeting Co-Organizer, Axon Dynamics, Banbury Conference
2006 Editorial Board, Journal of Cell Biology
2009 Board Member, Porter Foundation
2011 Editorial Board, Cell
2012 Meeting Co-Organizer, Axon Degeneration, Janelia Farm Conferences
2013 Chair, Gordon Research Conference on Motile and Contractile Systems
2014 Chair, Porter Foundation Board
2014 Jacob Javits Neuroscience Investigator Award, NINDS
2015 Meeting Co-Organizer, Janelia Farm Conference on Neuronal Trafficking
2015 F.E. Bennett Memorial Lecture, American Neurological Association
2015 Stanley N. Cohen Biomedical Research Award, University of Pennsylvania Perelman School of Medicine
2016 Jane M. Glick Graduate Student Teaching Award
2017 Elected to ASCB Council
2017 Meeting Co-Organizer, Banbury Conference on the Cell Biology of ALS
2017 Inducted as an inaugural ASCB Fellow for lifetime contributions to cell biology
2018 Elected Co-Chair, Gordon Research Conference on the Cell Biology of the Neuron

C. CONTRIBUTIONS TO SCIENCE

1. ***Molecular motors:*** As both a graduate student and an independent investigator, I have made consistent contributions to our understanding of the biochemistry and biophysics of molecular motors. As a graduate student, I identified the rate-limiting step in the kinetic pathway of the dynein ATPase (Holzbaur and Johnson, 1989a), and demonstrated that this step was specifically activated by microtubules (Holzbaur and Johnson, 1989b). In my own lab I have continued to investigate molecular

motors using biophysical approaches such as single molecule studies, optical trapping and optogenetics, to examine motor function both in vitro and in live cells.

Holzbaur EL, Johnson KA. Microtubules accelerate ADP release by dynein. Biochemistry. 1989 Aug 22;28(17):7010-6. PubMed PMID: 2531005.

Hendricks, A.G., E. Perlson, J.L. Ross, H.W. Schroeder, 3rd, M. Tokito, and E.L. Holzbaur. Motor coordination via a tug-of-war mechanism drives bidirectional vesicle transport. Curr Biol. 2010 Apr 27;20(8):697-702. PubMed PMID: 20399099; PubMed Central PMCID: PMC2908734.

Hendricks AG, Lazarus JE, Perlson E, Gardner MK, Odde DJ, Goldman, YE, and Holzbaur, EL. Dynein tethers and stabilizes dynamic microtubule plus ends. Curr Biol. 2012 Apr 10;22(7):632-7. PubMed PMID: 22445300; PubMed Central PMCID: PMC3347920.

Ghiretti AE, Thies E, Tokito MK, Lin T, Ostap EM, Kneussel M, Holzbaur EL. Activity-dependent regulation of distinct transport and cytoskeletal remodeling functions of the dendritic kinesin KIF21B. Neuron. 2016 Nov 23;92(4):857-872. PubMed PMID: 27817978; PubMed Central PMCID: PMC5283298.

2. ***Cytoplasmic dynein and dynactin:*** As a postdoctoral scientist and as an independent investigator, I have focused on understanding the interaction of cytoplasmic dynein and dynactin. I was the first to clone the full length p150^{Glued} subunit of dynactin (Holzbaur et al., 1991), the first to show the direct binding of p150^{Glued} to microtubules and to dynein (Waterman-Storer et al., 1995; Karki and Holzbaur, 1995), the first to use TIRF microscopy to image the motility of the dynein-dynactin complex along microtubules (Ross et al., 2006), and to show that dynactin recruits dynein to the microtubule (Ayloo et al., 2014). We are now focusing on organelle-specific activators of dynein function using optogenetics and single molecule approaches (Olenick et al., 2016).

Holzbaur EL, Hammarback JA, Paschal BM, Kravit NG, Pfister KK, et al. Homology of a 150K cytoplasmic dynein-associated polypeptide with the Drosophila gene Glued. Nature. 1991 Jun 13;351(6327):579-83. PubMed PMID: 1828535.

Waterman-Storer CM, Karki S, Holzbaur EL. The p150^{Glued} component of the dynactin complex binds to both microtubules and the actin-related protein centractin (Arp-1). Proc Natl Acad Sci U S A. 1995 Feb 28;92(5):1634-8. PubMed PMID: 7878030; PubMed Central PMCID: PMC42574.

Ross JL, Wallace K, Shuman H, Goldman YE, Holzbaur EL. Processive bidirectional motion of dynein-dynactin complexes in vitro. Nat Cell Biol. 2006 Jun;8(6):562-70. PubMed PMID: 16715075.

Olenick MA, Tokito M, Boczkowska M, Dominguez R, Holzbaur EL. Hook adaptors induce unidirectional processive motility by enhancing the dynein-dynactin interaction. J Biol Chem. 2016 Aug 26;291(35):18239-51. PMID: 27365401; PubMed Central PMCID: PMC5000072.

3. ***Mechanistic analysis of axonal transport:*** We were the first, and still lead the field, in using a sophisticated combination of in vitro single molecule and reconstitution approaches coupled to live cell imaging in primary neurons to probe the underlying mechanisms regulating opposing motors during the axonal transport of vesicles and organelles. For example, we used both single molecule approaches and live cell imaging to define the molecular mechanisms regulating motors to produce the highly processive motility of APP and autophagosomes in neurons (Fu and Holzbaur, 2013; Fu and Holzbaur, 2014), and we were the first to identify the molecular mechanisms underlying retrograde transport initiation from the axon terminal (Moughamian and Holzbaur, 2012; Moughamian et al., 2013; Twelvetrees et al., 2016), which we have now reconstituted in vitro (Nirschl et al., 2016), allowing the detailed analysis of regulatory mechanisms.

Dixit R, Ross JL, Goldman YE, Holzbaur EL. Differential regulation of dynein and kinesin motor proteins by tau. Science. 2008 Feb 22;319(5866):1086-9. PubMed PMID: 18202255; PubMed Central PMCID: PMC2866193.

Moughamian AJ, Holzbaur EL. Dynactin is required for transport initiation from the distal axon. Neuron. 2012 Apr 26;74(2):331-43. PubMed PMID: 22542186; PubMed Central PMCID: PMC3347924.

Fu MM, Nirschl JJ, Holzbaur EL. LC3 binding to the scaffolding protein JIP1 regulates processive dynein-drive transport of autophagosomes. Dev Cell 2014 Jun 9;29(5):577-90. PubMed PMID: 24914561; PubMed Central PMCID: PMC3734084. PubMed Central PMCID: PMC4109720.

Nirschl JJ, Magiera MM, Lazarus, JE, Janke C, and Holzbaur EL. α-Tubulin Tyrosination and CLIP-170 Phosphorylation Regulate the Initiation of Dynein-Driven Transport in Neurons. Cell Rep. 2016 Mar 22;14(11):2637-52. PubMed PMID: 26972003; PubMed Central PMCID: PMC4819336.

4. ***Molecular motors and neurodegeneration:*** Based on our observations that the interaction of dynein and dynactin is essential for normal vesicular transport (Waterman-Storer et al., 1995), we generated a transgenic mouse with a targeted disruption of the dynein-dynactin interaction in motor neurons (LaMonte et al., 2002). This mouse model developed late-onset, slowly progressive motor neuron degeneration, which led to an exciting collaboration with Dr. Kenneth Fischbeck identifying a G59S point mutation in the CAP-Gly domain of $p150^{Glued}$ as causative for motor neuron degeneration (Puls et al., 2003). We used both cellular approaches and mouse models (Levy et al., 2006; Chevalier-Larsen et al., 2008) to show that the G59S mutation disrupts dynactin function by perturbing microtubule-binding and inducing protein aggregation. The identification of additional mutations within the same domain of dynactin as causative for Perry syndrome (Farrer et al., 2009) led us to discover that these mutations cause neurodegeneration by a distinct mechanism (Moughamian and Holzbaur, 2012). Finally, we have used live cell and single molecule approaches to investigate the molecular basis for axonal transport deficits in neurodegenerative diseases including ALS (Perlson et al., 2009; Klinman and Holzbaur, 2016; Gopal et al., 2017) and Huntington's disease (Wong and Holzbaur, 2014).
   LaMonte BH, Wallace KE, Holloway BA, Shelly SS, Ascaño J, et al. Disruption of dynein/dynactin inhibits axonal transport in motor neurons causing late-onset progressive degeneration. Neuron 2002 May 30;34(5):715-27. PubMed PMID: 12062019.
   Wong YC, Holzbaur EL. The regulation of autophagosome dynamics by huntingtin and HAP1 is disrupted by expression of mutant huntingtin, leading to defective cargo degradation. J Neurosci. 2014 Jan 22;34(4):1293-305. PubMed PMID: 24453320; PubMed Central PMCID: PMC3898289.
   Klinman E, Holzbaur EL. Stress-induced CDK5 activation disrupts axonal transport via Lis1/Ndel1/Dynein. Cell Report 2016 Jul 21;12(3):462-73. PubMed PMID: 26166569; PubMed Central PMCID: PMC4532378.
   Gopal PP, Nirschl JJ, Klinman E, Holzbaur EL. Amyotrophic lateral sclerosis-linked mutations increase the viscosity of liquid-like TDP-43 RNP granules in neurons. Proc Natl Acad Sci U S A. 2017 Mar 21;114(12):E2466-E2475. PubMed PMID: 28265061; PubMed Central PMCID: PMC5373408.

5. ***Autophagy in neurons:*** We are using live cell imaging to examine the dynamics of autophagosome biogenesis (Maday et al, 2012; Maday and Holzbaur, 2014) and transport (Wong and Holzbaur, 2014a; Fu et al., 2014) in primary neurons. We were the first to demonstrate the robust, constitutive biogenesis of autophagosomes at the axon tip, and the first to show that constitutive autophagosomes engulf aggregated proteins such as $SOD1^{G93A}$ and polyQ-huntingtin. We are also investigating the molecular mechanisms regulating mitophagy. We were the first to identify optineurin as an essential autophagy receptor required for the Parkin-dependent clearance of damaged mitochondria (Wong and Holzbaur, 2014b); this clearance requires the upstream regulatory kinase TBK1 (Moore and Holzbaur, 2016). Importantly, mutations in both optineurin and TBK1 are causative for ALS, thus implicating mitophagy as a critical pathway involved in the pathogenesis of both ALS and Parkinson's disease.
   Maday S, Wallace KE, Holzbaur EL. Autophagosomes initiate distally and mature during transport toward the cell soma in primary neurons. J Cell Biol. 2012 Feb 20;196(4):407-17. PubMed PMID: 22331844; PubMed Central PMCID: PMC3283992.
   Maday S, Holzbaur EL. Autophagosome biogenesis in primary neurons follows an ordered and spatially regulated pathway. Dev Cell. 2014 Jul 14;30(1):71-85. PubMed PMID: 25026034; PubMed Central PMCID: PMC4109719.
   Wong YC, Holzbaur EL. Optineurin is an autophagy receptor for damaged mitochondria in parkin-mediated mitophagy that is disrupted by an ALS-linked mutation. Proc Natl Acad Sci U S A. 2014 Oct 21;111(42):E4439-48. PubMed PMID: 25294927; PubMed Central PMCID: PMC4210283.
   Moore AS, Holzbaur EL. Dynamic recruitment and activation of ALS-associated TBK1 with its target optineurin are required for efficient mitophagy. Proc Natl Acad Sci U S A. 2016 Jun

14;113(24):E3349-58. doi: 10.1073/pnas.1523810113. PMID: 27247382; PubMed Central PMCID: PMC4914160.

For a complete list of publications (≥139) please see:
http://www.ncbi.nlm.nih.gov/pubmed/?term=holzbaur+el

D. RESEARCH SUPPORT

Ongoing Research Support

1993/07/01-2018/07/31
R01 GM048661-23, National Institute of General Medical Sciences (NIGMS)
Holzbaur, Erika L (PI)
The Interaction of Cytoplasmic Dynein and Dynactin
This grant focuses on the following questions: (1) How does dynactin activate dynein-driven transport? (2) What are the mechanisms regulating the engagement of dynein-driven retrograde transport? (3) What are the mechanisms coordinating bidirectional vesicle transport along the axon?

2014/08/01-2019/07/31
P01 GM087253, NIH
Ostap, E. Michael (PI); Holzbaur, Erika L. (Subproject PI)
Molecular motor dynamics in organelle transport and membrane remodeling
This grant focuses on: (1) examining the interactions of opposing microtubule-based and actin-based motors, at cytoskeletal intersections both *in vitro* and in the cell; (2) examining the role of opposing dynein and kinesin motors in modulating membrane tubulation and cargo sorting; (3) examining the role of Hook1 and BAR-domain proteins in endosome motility and tubulation.

2007/12/01-2018/01/31
R37 NS060698-07, National Institute of Neurological Disorders and Stroke (NINDS)
Holzbaur, Erika L (PI)
Mechanistic analysis of axonal transport defects in neurodegenerative disease.
This grant focuses on the following questions: (1) How is axonal transport altered in mouse models of ALS by the aberrant activation of upstream kinases? (2) What are the pathways for autophagosome biogenesis and cargo-loading and how are these affected by cellular stress? and (3) How do defects in autophagy lead to neurodegeneration?

2016/08/01-2017/04/30
R01 NS093383, National Institute of Neurological Disorders and Stroke (NINDS)
Smith, Wanli (PI); Holzbaur, Erika L (Subproject PI)
LRRK2, KIF1A and Parkinson disease
This subaward focuses on investigating the interaction of LRRK2 with kinesin-1 using in vitro and live cell imaging assays.