BIOGRAPHICAL SKETCH

NAME: ESPENSHADE, PETER JOHN

eRA COMMONS USER NAME: pespens2

POSITION TITLE: PROFESSOR OF CELL BIOLOGY

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE	YEAR(s)	FIELD OF STUDY
Princeton University	B.A.	1986-1990	Molecular Biology
Massachusetts Institute of Technology	Ph.D.	1991-1998	Biology
UT-Southwestern Medical Center, Dallas, TX	Postdoc	1998-2002	Molecular Cell Biology

A. Personal Statement

I have 25 years experience in studies of yeast and mammalian molecular cell biology. As a doctoral student with Chris Kaiser at MIT, I studied protein trafficking in the secretory pathway, specifically mechanisms of COPII vesicle formation at the ER (Publications 1-4). Since 1998, I have focused on the regulation of sterol regulatory element-binding protein (SREBP) by lipid and oxygen supply. SREBP is a membrane-bound transcription factor that is the central regulator of cellular fatty acid and cholesterol synthesis. As a postdoctoral fellow with Dr. Michael Brown and Dr. Joseph Goldstein at UT-Southwestern, I described the sterol-regulated mechanism of SREBP proteolytic activation in mammals (Publications 5-8).

As an independent investigator, my lab has made several high impact discoveries including that fungal SREBP is a conserved hypoxic transcription factor that utilizes a novel cleavage mechanism for activation (Publications 9-12). We discovered a novel 2-oxoglutarate-Fe(II)-dioxygenase Ofd1 that functions as an oxygen sensor to regulate the degradation and DNA binding activity of yeast SREBP (Publications 13-16). Our studies of Ofd1 described a new paradigm for oxygen sensing and control of hypoxic gene expression.

Our lab has extensive experience with model organism research, including yeast, tissue culture, and most recently mice. In 2007, we discovered that PGRMC1 broadly regulates cytochromes P450 activity and is required for cholesterol synthesis in mammalian cells (Publication 17). For the past 3 years, we have studied the function of PGRMC1 using a conditional knockout mouse model that we generated.

Recently, we shifted our research interests from yeast back to that of mammalian SREBP, as many important questions remain unanswered. Our yeast work demonstrated that SREBP is required for adaptation to low oxygen, conditions under which lipid synthesis becomes limiting. Solid tumors, for example pancreatic ductal adenocarcinoma (PDAC), are hypoxic and nutrient-deprived. We are examining how human PDAC cells and tumors maintain lipid supply required for growth whether SREBP is required for tumor growth.

B. Positions and Honors

Positions and Employment

1990-1991 Research Assistant, Princeton University, Department of Molecular Biology
1991-1997 Graduate Student, Lab of Chris Kaiser, Massachusetts Institute of Technology
1997-2002 Postdoctoral Fellow, Lab of Brown and Goldstein, UT-Southwestern Medical Center at Dallas
2002-2008 Assistant Professor, Department of Cell Biology, Johns Hopkins University School of Medicine
2008-2013 Associate Professor, Department of Cell Biology, Johns Hopkins University School of Medicine
2013-date Professor, Department of Cell Biology, Johns Hopkins University School of Medicine
2014-date Associate Dean for Graduate Biomedical Education

Other Experience

2008	INMP NIH Study Section, Ad hoc member for two meetings
2009 – 2012	INMP NIH Study Section, Permanent Member
2014	American Heart Association, Microbiology BSc 2 Study Section
2014	NIH ZRG1 CB-C Study Section
2016	NIH ZRG1 EMNR F Study Section, November 2016

Honors

1990	Summa cum laude, Department of Molecular Biology, Princeton University
1990	Phi Beta Kappa Honor Society, Princeton University
1992	National Science Foundation Pre-Doctoral Fellowship
1998	National Research Service Award Postdoctoral Fellowship, NIH
2001	Career Award in the Biomedical Sciences, Burroughs Wellcome Fund
2006	Investigator in Pathogenesis of Infectious Disease Award, Burroughs Wellcome Fund
2007	Dean's Fund Award, Johns Hopkins University School of Medicine
2008	Established Investigator, American Heart Association
2012	ASBMB Avanti Young Investigator Award in Lipid Research
2014	Fellow of the American Association for the Advancement of Science

C. Contributions to Science (from 58 peer-reviewed publications)

Defined SEC16 as a peripheral ER membrane protein that scaffolds COPII coat protein assembly

SEC16 was an uncharacterized gene required for ER vesicle formation in *S. cerevisiae*. As a graduate student, I performed a comprehensive characterization of *SEC16* function, demonstrating that Sec16p bound directly to COPII subunits and nucleated COPII protein assembly.

1. **Espenshade P**, Gimeno RE, Holzmacher E, Teung P, Kaiser CA. 1995. Yeast *SEC16* gene encodes a multidomain vesicle coat protein that interacts with Sec23p. ***J. Cell Biol.*** 131:311-324. PMCID: PMC2199983

2. Gimeno RE, **Espenshade P**, Kaiser CA. 1995. *SED4* encodes a yeast endoplasmic reticulum protein that binds Sec16p and participates in vesicle formation. ***J. Cell Biol.*** 131:325-338. PMCID: PMC2199979

3. *Gimeno RE, ***Espenshade P**, Kaiser CA. 1996. COPII coat subunit interactions: Sec24p and Sec23p bind to adjacent regions of Sec16p. ***Mol. Biol. Cell*** 7:1815-1823. *Co-first author. PMCID: PMC276028

4. Shaywitz DA, **Espenshade P**, Gimeno RE, Kaiser CA. 1997. COPII subunit interactions in the assembly of the vesicle coat. ***J. Biol. Chem.*** 272:25413-25416.

Described the mechanism for sterol-regulated proteolytic activation of SREBP transcription factors

The ER membrane-bound transcription factor sterol regulatory element-binding proteins (SREBP) are central transcriptional regulators of cellular lipid homeostasis. As a postdoc with Brown and Goldstein at UT-Southwestern, I determined that the Site-1 protease, which first cleaves SREBP, is localized in the Golgi and that SREBP cleavage requires ER-to-Golgi transport. I demonstrated that cholesterol exerts negative feedback regulation on SREBP by controlling its incorporation into COPII, ER transport vesicles. In addition, I showed that the SREBP cleavage activating protein bound an unknown factor in the presence of sterols, leading to the identification of INSIG proteins, key negative regulators of the SREBP pathway.

5. **Espenshade PJ**, Cheng D, Goldstein JL, Brown MS. Autocatalytic processing of Site-1 protease removes propeptide and permits cleavage of sterol regulatory element-binding proteins. 1999. ***J. Biol. Chem.*** 274:22795-227804.

6. DeBose-Boyd RA, Brown MS, Li WP, Nohturfft A, Goldstein JL, **Espenshade PJ**. 1999. Transport-dependent proteolysis of SREBP: relocation of Site-1 protease from Golgi to ER obviates the need for SREBP transport to Golgi. ***Cell*** 99:703-712.
7. Nohturfft A, Yabe D, Goldstein JL, Brown MS, **Espenshade PJ**. 2000. Regulated step in cholesterol feedback localized to budding of SCAP from ER membranes. ***Cell*** 102:315-323.

8. Yang T, **Espenshade PJ**, Wright ME, Yabe D, Gong Y, Aebersold R, Goldstein JL, Brown MS. Crucial step in cholesterol homeostasis: sterols promote binding of SCAP to INSIG-1, a membrane protein that facilitates retention of SREBPs in the ER. 2002. ***Cell*** 110:489-500.

<u>Discovered that fungal SREBP functions as an oxygen responsive transcription factor</u>

Prior to this work, the SREBP transcription factors were thought to function only as regulators of systemic lipid homeostasis. As an independent investigator, I asked the question: What then is this transcription factor doing in a unicellular organism? We discovered that SREBP is an oxygen-responsive transcription factor required for growth in hypoxic environments. These findings created the field of fungal SREBP research and led to our discovery that SREBP is required for virulence in the fungal pathogen *Cryptococcus neoformans*. Subsequently, others showed this to be true for *Aspergillus fumigatus* and other pathogens. Our studies of yeast SREBP activation revealed a novel mechanism for SREBP cleavage activation that requires the multi-subunit Golgi Dsc E3 ligase and the rhomboid serine protease Rbd2. In addition, these studies prompted the search for additional SREBP functions in mammals, which now include roles in the inflammatory response and autophagy.

9. Hughes AL, Todd BL, **Espenshade PJ**. 2005. SREBP pathway responds to sterols and functions as an oxygen sensor in fission yeast. ***Cell*** 120:831-842.

10. Chang YC, Bien CM, Lee H, **Espenshade PJ*,** Kwon-Chung KJ*. 2007. Sre1p, a regulator of oxygen sensing and sterol homeostasis, is required for virulence in *Cryptococcus neoformans*. ***Mol. Microbiol.*** 64:614-629. *Corresponding authors.

11. Stewart EV, Nwosu CC, Tong Z, Roguev A, Cummins TD, Kim DU, Hayles J, Park HO, Hoe KL, Powell DW, Krogan NJ, **Espenshade PJ**. 2011. Yeast SREBP cleavage activation requires the Golgi Dsc E3 ligase complex. ***Mol. Cell*** 42:160-171. PMCID: PMC3083633

12. Hwang J, Ribbens D, Raychaudhuri S, Cairns L, Gu H, Frost A, Urban S, **Espenshade PJ**. 2016. A Golgi rhomboid protease Rbd2 recruits Cdc48 to cleave yeast SREBP. ***EMBO J.*** 35:2332-2349. PMCID: PMC5090219

<u>Discovered the 2-oxoglutarate-Fe(II)-dioxygenase Ofd1 that functions as an oxygen sensor to regulate SREBP activity in fungi</u>

Prior to this work, known eukaryotic mechanisms for control of oxygen-regulated transcription factors included expression (Rox1 in *S. cerevisiae),* coactivator binding (hypoxia-inducible factor - HIF), degradation (HIF), and proteolytic activation (yeast SREBP). In this series of studies, we demonstrated that the prolyl hydroxylase Ofd1 regulates both DNA binding and degradation of activated yeast SREBP Sre1. Mathematical modeling studies showed that both mechanisms are critical for the cellular response to hypoxia. This work defined a new mechanism for hypoxic gene regulation.

13. Hughes BT and **Espenshade PJ**. 2008. Oxygen-regulated degradation of fission yeast SREBP by Ofd1, a prolyl hydroxylase family member. ***EMBO J.*** 27:1491-1501. PMCID: PMC2396400

14. Lee CY, Stewart EV, Hughes BT and **Espenshade PJ**. 2009. Oxygen-dependent binding of Nro1 to the prolyl hydroxylase Ofd1 regulates SREBP degradation in yeast. ***EMBO J****.* 28:135-143. PMCID: PMC2634736

15. Lee CSY, Yeh TL, Hughes BT, **Espenshade PJ**. 2011. Regulation of the Sre1 hypoxic transcription factor by oxygen-dependent control of DNA binding. ***Mol. Cell*** 44:225-234. PMCID: PMC3208185

16. Porter JR, Lee CSY, **Espenshade PJ***, Iglesias PA*. 2012. Regulation of SREBP during hypoxia requires Ofd1-mediated control of both DNA binding and degradation. ***Mol. Biol. Cell*** 23:3764-3774. PMCID: PMC3442422

Defined PGRMC1 as a new regulator of cytochrome P450 enzymes in yeast and mammals

The human genome codes for 57 cytochromes P450 that perform important chemical reactions ranging from Phase I drug metabolism to steroid synthesis. Prior to our work, cytochrome P450 activity was thought to only require electron donation through transient interactions with cytochrome P450 reductase. Through a series of genetic and biochemical experiments, we demonstrated that Dap1/PGRMC1 is an ER membrane hemoprotein and a positive regulator of cytochrome P450 enzymes functioning in sterol synthesis. This study demonstrated that Dap1/PGRMC1 is a direct regulator of cytochrome P450 enzymes and a new protein required for sterol synthesis in yeast and mammals. The fact that PGRMC1 binds to all enzymes tested raises the possibility that PGRMC1 functions in a multitude of biochemical pathways.

17. Hughes AL, Powell DW, Bard M, Eckstein J, Barbuch R, Link AJ, **Espenshade PJ**. 2007. Dap1/PGRMC1 binds and regulates cytochrome P450 enzymes. ***Cell Metabolism*** 5:143-149.

Complete List of Published Work (NCBI MyBibliography):
http://www.ncbi.nlm.nih.gov/sites/myncbi/peter.espenshade.1/bibliography/40609512/public/?sort=date&direction=ascending

D. Research Support

Ongoing Research Support

R01 DK107643 National Institute of Diabetes, Digestive and Kidney Diseases – NIH 12/16/15 – 11/30/20
Espenshade - PI
Title: Mechanism of SREBP Cleavage Activating Protein (SCAP) Golgi-to-ER Recycling
The goals of this proposal are to identify sequences and machinery required for SCAP recycling and to test whether SCAP recycling is required for regulation of lipid homeostasis.

R01 HL77588 National Heart, Lung, Blood Institute – NIH 07/01/04 – 03/31/18
Espenshade - PI
Title: Regulation of Cellular Cholesterol Homeostasis
The primary objective of this project is to use fission yeast as a genetic model for mammalian sterol homeostasis. Specifically, we will 1) identify genes required for yeast SREBP (Sre1) cleavage, 2) define the machinery for Sre1 cleavage, and 3) define the mechanism of sterol-regulated Sre1 cleavage.