BIOGRAPHICAL SKETCH

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NAME: Hughson, Frederick M.

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

POSITION TITLE: Professor

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
Yale University	B.S.	05/1984	Molecular Biophysics & Biochemistry
Stanford University School of Medicine	Ph.D.	08/1990	Biochemistry
Harvard University	Postdoctoral	10/1994	Biochemistry

A. Personal Statement

My group uses diverse methods, especially X-ray crystallography and cryo-EM, to study fundamental problems in cell biology. Our primary focus has been on intracellular trafficking in eukaryotes. Several of our current projects involve structural and mechanistic studies of large multi-subunit protein complexes (MTCs) that orchestrate the docking and fusion of transport vesicles. These complexes guide cargo-laden vesicles to their destinations and coordinate the activities of other components of the trafficking machinery, including the SNARE proteins that catalyze membrane fusion itself. We are also studying the roles of Sec1/Munc18 (SM) proteins in the assembly of fusogenic SNARE complexes. Our structural and functional studies of MTCs and SM–SNARE complexes have led to new mechanistic models for these important proteins.

I am well-qualified for these studies based on my graduate work on protein folding, my postdoctoral work determining the crystal structure of influenza hemagglutinin in its active, low-pH induced conformation, and my 25 years as a structural biologist at Princeton, during which we have determined over 40 X-ray crystal structures. I have extensive experience administering a productive laboratory as demonstrated by our funding and publication records. I have trained over 30 Ph.D. students and postdoctoral fellows, many of whom have gone on to tenure-track faculty positions (and four of whom are now tenured). I maintain a highly interactive environment where lab members are encouraged to share ideas, expertise, and reagents. Moreover, I continue to perform laboratory research, ensuring that I remain connected and accessible to my trainees.

B. Positions and Honors

Positions and Employment

1984 - 1990 Graduate Fellow, Stanford Department of Biochemistry
Robert L. Baldwin, advisor. Research: Protein folding
1990 - 1994 Postdoctoral Fellow, Harvard Dept. of Biochemistry & Molecular Biology
Don C. Wiley, advisor. Research: X-ray crystal structure of low-pH hemagglutinin
1994 - 2002 Assistant Professor of Molecular Biology, Princeton University
2002 - 2009 Associate Professor of Molecular Biology, Princeton University
2006 - 2015 Director, Princeton/HHMI Undergraduate Science Education Program
2009 - Professor of Molecular Biology, Princeton University

Other Experiences and Professional Memberships

1999 -	Member of more than ten NIH Special Emphasis Panels
2003	Ad hoc reviewer, NIH CDF-4 Study Section
2003 -	Editorial Board, PLoS Biology
2008	Ad hoc reviewer, NIH MSFB Study Section
2010 - 2017	Member, AHA Peer Review Committee (PC1: Basic Cell Biology)
2011 - 2017	Member, ASBMB Education and Professional Development Committee
2015	Chair, Gordon Research Conference on Molecular Membrane Biology
2015 - 2020	Board of Reviewing Editors, Science

Honors

1984 - 1987	NSF Pre-doctoral Fellowship
1990 - 1994	Helen Hay Whitney Postdoctoral Fellowship
1995 - 1998	Searle Scholar
1995 - 1997	Beckman Young Investigator
2015	President's Award for Distinguished Teaching, Princeton University

C. Contributions to Science

1. **Conformational control of SNARE assembly.** The mechanisms by which cells distribute materials among intracellular organelles, and execute exo- and endocytosis, are of fundamental importance in cell biology. SNARE proteins, discovered in 1993 by Jim Rothman's group, play central roles in these processes, yet when I started my lab in 1994 little was known about the structures and conformational dynamics of this protein family. Our early work, focused on exocytic SNAREs in yeast and neurons, helped to fill this gap by (1) characterizing the pathway of SNARE assembly in vitro and its relationship to protein folding, (2) showing that SNARE assembly is kinetically controlled by the requirement for a conformational change in a key SNARE protein, (3) investigating this conformational change in energetic and structural terms, and (4) characterizing SNARE assembly in vivo.

    a. Nicholson, K.L., Munson, M., Miller, R.B., Filip, T.J., Fairman, R., and Hughson, F.M. (1998) Regulation of SNARE complex assembly by an N-terminal domain of the t-SNARE Sso1p. Nature Struct Biol 5, 793-802.
    b. Lerman, J.C., Robblee, J., Fairman, R., and Hughson, F.M. (2000) Structural analysis of the neuronal SNARE protein syntaxin-1A. Biochemistry 39, 8470-8479.
    c. Munson, M., Chen, X., Cocina, A.E., Schultz, S.M., and Hughson, F.M. (2000) Interactions within the yeast t-SNARE Sso1p that control SNARE assembly. Nature Struct Biol 7, 894-902.
    d. Munson, M. and Hughson, F.M. (2002) Conformational regulation of SNARE assembly and disassembly in vivo. J Biol Chem 277, 9375-9381.

2. **Structure/function studies of multisubunit tethering complexes (MTCs): the Dsl1 complex.** In a second phase of our work on the machinery of vesicle trafficking, we turned to the multisubunit 'tethering' complexes (MTCs) that interact with SNAREs to orchestrate vesicle docking and fusion. By combining single particle negative-stain EM (carried out in collaboration with Tom Walz) and X-ray crystallography, we were able to propose a nearly complete atomic-resolution model for the hetero-trimeric, 260-kDa Dsl1 complex. Our results strongly suggested that even the simplest MTC is capable of orchestrating vesicle capture, uncoating, and fusion. In recent work, we have elucidated the structural basis for the binding of the Dsl1 complex to both the vesicle and target membranes.

    a. Ren, Y., Yip, C., Tripathi, A., Huie, D., Jeffrey, P.D., Walz, T., and Hughson, F.M. (2009) Structural basis for vesicle tethering by the Dsl1p complex. Cell 139, 1119-1129. PMCID: PMC2806190
    b. Suckling, R.J., Poon, P.P., Travis, S.M., Hughson, F.M., Evans, P.R., Duden, R., and Owen, D.J. (2015) Structural basis for the binding of tryptophan-based motifs by delta-COP. Proc Natl Acad Sci USA 112, 14242-14247. PMCID: PMC4655537

c. Travis, S.M., Kokona, B., Fairman, R., and Hughson, F.M. (2019) Roles of singleton tryptophan motifs in COPI coat stability and vesicle tethering. Proc Natl Acad Sci USA 116, 24031-24040. PMCID: PMC6883825
d. Travis, S.M., DAmico, K., Yu, I.M., McMahon, C., Hamid, S., Ramirez-Arellano, G., Jeffrey, P.D., and Hughson, F.M. (2020). Structural basis for the binding of SNAREs to the multisubunit tethering complex Dsl1. J Biol Chem 295, 10125-10135. PMCID: PMC738367

3. **Structure/function studies of multisubunit tethering complexes (MTCs): the COG complex.** Another and much larger multisubunit tethering complex (MTC), the hetero-octameric COG complex (650 kDa), was initially isolated on the basis of its ability to stimulate an in vitro Golgi transport assay. Subsequent work strongly implied that COG functions to tether vesicles carrying cargo between Golgi cisternae. It therefore plays a key role in maintaining the proper distribution of glycosylation enzymes and other materials within the Golgi. As a consequence, COG defects can lead to fatal glycosylation disorders. To provide a foundation for understanding COG function in mechanistic terms, we used biochemical and negative-stain EM-based approaches to determine the molecular architecture of the yeast and mammalian COG complexes, elucidated the structural basis for human glycosylation disorders caused by mutations in several COG subunits, and (3) used X-ray crystallography to reveal the structural basis for the subunit interactions that hold COG (and other multisubunit tethering complexes) together.

a. Richardson, B.C., Smith, R.D., Ungar, D., Nakamura, A., Jeffrey, P.D., Lupashin, V.V., and Hughson, F.M. (2009) Structural basis for a human glycosylation disorder caused by mutation of the *COG4* gene. Proc Natl Acad Sci USA 106, 13329-13334. PMCID: PMC2716380
b. Lees, J.A., Yip, C.K., Walz, T., and Hughson, F.M. (2010) Molecular organization of the COG vesicle tethering complex. Nature Struct Mol Biol 17, 1292-1297. PMCID: PMC3113405
c. Ha, J.Y., Pokrovskaya, I.D., Climer, L.K., Shimamura, G.R., Kudlyk, T., Jeffrey, P.D., Lupashin, V.V., and Hughson, F.M. (2014) Cog5-Cog7 crystal structure reveals interactions essential for the function of a multisubunit tethering complex. Proc Natl Acad of Sci USA 111, 15762-15767. PMCID: PMC4226102
d. Ha, J.Y., Chou, H.T., Ungar, D., Yip, C.K., Walz, T., and Hughson, F.M. (2016) Molecular architecture of the complete COG tethering complex. Nature Struct Mol Biol 23, 758-760. PMCID: PMC4972656

4. **Sec1/Munc18 (SM) family proteins as templates for SNARE complex assembly.** The study of SM proteins has become a major focus of my lab. Our structural studies of the SM protein Vps33 bound to SNARE proteins led to a new model of SM protein function—specifically, that SM proteins act as templates to catalyze SNARE complex assembly. Single-molecule optical tweezers experiments validated this model and generalized it to other SM proteins including Munc18-1, which catalyzes neuronal SNARE assembly.

a. Baker, R.W., Jeffrey, P.D., and Hughson, F.M. (2013) Crystal structures of the Sec1/Munc18 (SM) protein Vps33, alone and bound to the homotypic fusion and vacuolar protein sorting (HOPS) subunit Vps16. PLoS One 8, e67409. PMCID: PMC3693963
b. Baker, R.W., Jeffrey, P.D., Zick, M., Phillips, B.P., Wickner, W.T., and Hughson, F.M. (2015) A direct role for the Sec1/Munc18-family protein Vps33 as a template for SNARE assembly. Science 349, 1111-1114. PMCID: PMC4727825
c. Jiao, J., He, M., Port, S.A., Baker, R.W., Xu, Y., Qu, H., Xiong, Y., Wang, Y., Jin, H., Eisemann, T.J., Hughson, F.M.*, and Zhang, Y.* (2018) Munc18-1 catalyzes neuronal SNARE assembly by templating SNARE association. eLife, e41771. PMCID: PMC6320071 (*Corresponding authors)
d. Eisemann, T., Allen, F., Lau, K., Shimamura, G.R., Jeffrey, P.D., and Hughson, F.M. (2020). The Sec1/Munc18 protein Vps45 holds the Qa-SNARE Tlg2 in an open conformation. eLife 17, e60724. PMCID: PMC7470827

5. **Structure/function studies of bacterial quorum sensing.** In 2002, we initiated a collaboration with my Princeton colleague, Bonnie Bassler, to study bacterial cell-cell communication, or quorum sensing. Our work has elucidated (1) the structures of signal molecules used to mediate quorum sensing, (2) structures of the bacterial receptors for these signal molecules, and (3) mechanisms whereby quorum sensing can be modulated by synthetic molecules. The last of these represents a new approach to the discovery and optimization of broad spectrum anti-bacterial compounds.

a. Chen, X., Schauder, S., Potier, N., Van Dorsselaer, A., Pelczer, I., Bassler, B.L., and Hughson, F.M. (2002). Structural identification of a bacterial quorum-sensing signal containing boron. Nature 415, 545-549.
b. Neiditch, M.B., Federle, M.J., Pompeani, A.J., Kelly, R.C., Swem, D.L., Jeffrey, P.D., Bassler, B.L., and Hughson, F.M. (2006) Ligand-induced asymmetry in histidine sensor kinase complex regulates quorum sensing. Cell 126, 1095-1108.
c. Chen, G., Swem, L.R., Swem, D.L., Stauff, D.L., O'Loughlin, C.T., Jeffrey, P.D., Bassler, B.L., and Hughson, F.M. (2011). A strategy for antagonizing quorum sensing. Mol Cell 42, 199-209. PMCID: PMC3982643
d. Boyaci, H., Shah, T., Hurley, A., Kokona, B., Li, Z., Ventocilla, C., Jeffrey, P.D., Semmelhack, M.F., Fairman, R., Bassler, B.L., and Hughson, F.M. (2016) Structure, regulation, and inhibition of the quorum-sensing signal integrator LuxO. PLoS Biol 14, e1002464. PMCID: PMC4878744

Complete List of Published Work in MyBibliography:
http://www.ncbi.nlm.nih.gov/sites/myncbi/1ZWDwyXl9p/bibliography/40445178/public/?sort=date&direction=descending

D. Additional Information: Research Support and/or Scholastic Performance

Ongoing Research Support

R01 GM071574 Hughson (PI) 03/01/2005–02/28/2025
Structural Analysis of Membrane Tethering and Fusion Proteins
This study has three aims: (1) structural and single-molecule studies of SM protein-catalyzed SNARE complex assembly; (2) structural and proteomic studies of Dsl1-mediated vesicle capture and SNARE assembly; and (3) structural studies of the HOPS complex and its function in SNARE assembly.
Role: PI