BIOGRAPHICAL SKETCH

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NAME: Perkins, Guy

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

POSITION TITLE: Director of Tomography, Project Scientist

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
University of Utah, Salt Lake City, UT	B.A. Honors	06/1986	Physics
University of Utah, Salt Lake City, UT	B.S.	06/1986	Mathematics
University of California, Berkeley, CA	Ph.D.	05/1992	Biophysics
Lawrence Berkeley Laboratory, Berkeley, CA	Postdoctoral	11/1992	Electron Microscopy
Rijksuniversiteit Groningen, The Netherlands	Postdoctoral	11/1994	Electron Microscopy
Brandeis University, Waltham, MA	Postdoctoral	07/1995	Electron Microscopy
San Diego State University and University of California-San Diego, San Diego, CA	Postdoctoral	06/1996	Electron Microscopy

A. Personal Statement

I am an NIH principal investigator and have provided leadership at the National Center for Microscopy and Imaging Research (NCMIR) in the capacity of Director of Tomography since 2003. In this role, I have applied advanced, new instrumentation and computer algorithms to studies of biological structures *in situ*. After joining NCMIR in 1995, NCMIR Director Mark Ellisman and I developed electron microscope tomography (EMT) methods to visualize the 3D architecture of mitochondria in cells and tissues. Applying my mathematics, physics, and biophysics training, I have been a driving force in the quantitative analysis of mitochondrial architecture and nanoscale structural landmarks. As a result, I am the world's most published researcher on the structure of mitochondria. The structure I named the *crista junction* in a 1997 paper has been characterized by my colleagues and I since then and has been featured in well-respected scientific journals, including journal covers, in news articles, in molecular cell biology textbooks, and was recognized by the *Royal Society of Chemistry* with its own webpage. Of my 145 peer-reviewed publications, 90 have focused on mitochondrial structure and function in healthy and disease states. I work independently with a number of collaborators around the U.S.A., and my publications demonstrate a successful record of assisting investigators from other disciplines in structural studies and in contributing to the research activities of diverse programs, including program project grants, that rely on my knowledge of cell-biology driven 3D electron microscopy. One of my mitochondrial movies was shown in the U.S. Congress (Mootha, V., Perkins, G., Ellisman, M.H. Mitochondria 101 - How Something So Small is So Important to Human Life. *Congressional Mitochondrial Disease Caucus*, Washington D.C., Sept. 20, 2012). The BBC series *Life* used another of my movies highlighting the 3D aspects of mitochondria; my electron microscopy of human mitochondria was in the 'Why Are We Here?' episode of *Human Universe* (BBC2 and the Discovery Channel). I am the director for the 3DEM website for investigators in molecular and cellular 3D structure (>4500 subscribers worldwide, highlighted by Richard Henderson in his 2017 Nobel Lecture. I am a co-inventor on a patent with Dr. Ryuji Yamaguchi, Japan, for a promising cancer therapy involving combinational treatment with compounds that show surprising synergy to force mitochondrial-induced apoptosis, even in tumor cell types resistant to apoptosis. I serve as a Review Editor for the journal "Frontiers in Neuroscience".

Ongoing and recently completed projects that I would like to highlight include:

R01 EY031697
Ju, Perkins (MPI)
09/01/20 – 06/30/24
Mitochondrial Protection In Glaucomatous Optic Neuropathy

R01 DK116624
Strack (PI)
06/01/18 – 05/31/22
Targeting Mitochondrial Fission for Neuroprotection in Diabetic Neuropathy

R01 EY030104
Bossy-Wetzel (PI)
03/01/20 – 01/31/24
Lysine Acetylation As Switch For Optic Atrophy 1 Inactivation

R01 DC012938
von Gersdorff (PI)
08/01/20 – 07/31/25
Modulation Of Exocytosis And Excitability In Mature Auditory Brainstem Neurons

B. Positions, Scientific Appointments, and Honors

Positions and Scientific Appointments

2012 - present	Project Scientist, Univ. of CA, San Diego
2007 - present	Member, Scientific Advisory Board, NCMIR, Univ. of CA, San Diego
2003 - present	Director of Tomography, National Center for Microscopy and Imaging Research
2006 - 2012	Associate Project Scientist, Univ. of CA, San Diego
1999 - 2006	Assistant Project Scientist, Univ of CA, San Diego
1996 - 2012	Adjunct Faculty, San Diego State Univ
1989 - 1992	Graduate Student Research Assistant, Lawrence Berkeley Laboratory, Berkeley, CA
1986 - 1989	NIH Research Trainee, Univ. of CA, Berkley

Other Experience and Professional Memberships

2007 - 2008	School of Medicine Recruitment and Admissions Committee, Univ. of CA, San Diego
2006 - 2008	Faculty Mentor, Univ. of CA, San Diego
2001 - present	Member, Mitochondria Interest Group

Honors

2015	Speaker: Crick-Jacobs Workshop: Function and Failure of Calcium at Synapses
2011	Speaker: The Expanding Roles of Mitochondria Conference, HHMI
1993	Plenary Speaker: Dutch Electron Microscopy Society, Papendaal, Netherlands
1989	National Research Service Award from the N.I.H
1980	Honors at Entrance Scholarship

C. Contributions to Science

1. Since the late 1990s, my focus has been on the study of mitochondria using the then new technique of electron tomography. My work resulted in a new paradigm of mitochondrial structure, featured in well-respected journals, including journal covers, in news articles, in molecular cell biology textbooks, and by mainstream media. My 3D mitochondrial structures are on display in 2 science museums, in Maryland and Amsterdam, the Netherlands. Example publications are:
    a. Perkins, G.A., Guo-Sun, M., and Frey, T.G. (2009) Correlated Light and Electron Microscopy/Electron Tomography of Mitochondria *in situ, Methods Enzym*., 456:27-50. PMCID: PMC2730195

b. Perkins, G.A., Tjong, J., Brown, J.M., Poquiz, P.H., Scott, R.T., Kolson, D.R., Ellisman M.H. and Spirou, G.A. (2010) The micro-architecture of mitochondria at active zones: Electron tomography reveals novel anchoring scaffolds and cristae structured for high rate metabolism. *J. Neurosci.,* 30:1015-1026. PMCID: 2829299
c. Perkins, G.A. and Ellisman, M.H. (2011) Mitochondrial Configurations in Peripheral Nerve Suggest Differential ATP Production. *J. Structural Biology*, 173:117-127. PMCID: PMC3078762.
d. Weissert, V., Rieger, B., Morris, S., Arroum, T., Psathaki, O.E., Perkins, G., Zobel, T. and Busch, K.B. (2020) Inhibition of the mitochondrial ATPase function by IF1 changes the spatiotemporal organization of ATP synthase. *BBA Bioenergetics*, Oct 13;148322.

2. I then broadened my focus to investigate mitochondrial structure/function relationships in disease states working with collaborators. A fissioning mitochondrion in a Huntington's Disease model was awarded a prize by the French journal *La Recherche* as "one of the most impressive images of 2011." A few example publications are:
   a. Lee, J.H., Budanov, A.V., Park, E.J., Birse, R., Kim, T.E., Perkins, G.A., Ellisman, M.H., Bodmer, R., Bier, E., Karin, M. (2010) Sestrin is a feedback inhibitor of TOR that prevents age-related pathologies. Science, 327: 1223-1228. Cover Figure for the journal issue. PMCIDL PMC2866632
   b. Zhang, J., Guan, Z., Murphy, A.N., Wiley, S.E., Perkins, G.A., Worby, C.A., Engel, J.L., Heacock, P., Nguyen, O.K., Wang, J.H., Raetz, C.R.H., Dowhan, W., and Dixon, J.E. (2011) Mitochondrial phosphatase PTPMT1 is essential for cardiolipin biosynthesis. Cell Metabolism, 13:690-700. PMCID: PMC3119201
   c. Lee, J.H., Budanov, A.V., Talukdar, S., Park, E.J., Park, H., Park, H.-W., Bandyopadhyay, G., Li, N., Aghajan, M., Jang, I., Wolfe, A.M., Perkins, G.A., Ellisman, M.H., Bier, E., Scadeng, M., Viollet, B., Olefsky, J., Karin, M. (2012) Maintenance of metabolic homeostasis by Sestrin 2 and 3., Cell Metabolism, 16:311-321. PMCID: PMC3687365
   d. Lee, Y.S., Morinaga, H., Kim, J.J., Lagakos, W., Taylor, S., Keshwani, M., Perkins, G., Dong, H., Kayali, A.G., Sweet, I.R. and Olefsky, J. (2013). The Fractalkine and CX3CR1 System Regulates Beta Cell Function: A Novel Pathway for Regulation of Insulin Secretion. Cell, 153:413-25. PMID: 23582329, PMCID: PMC3717389

3. Part of my current research is the investigation of mitochondria in healthy neurons, neuropathies, and neurodegeneration. A few example publications are:
   a. Perkins, G.A., Jackson, D.R., Spirou, G.A. (2015) Resolving Presynaptic Structure by Electron Tomography. Synapse, 69:268-282. Cover figure for issue. PMCID4955585
   b. Yin, X., Kidd, G.J., Ohno, N., Perkins, G.A., Ellisman, M.H., Bastian, C., Brunet, S., Baltan, S. and Trapp, B.D. (2016) Myelin modulates axonal mitochondria viability via metabolic coupling. J. Cell Biology, 215(4):531-542. Featured in a JCB Spotlight by Beirowski, Babetto and Wrabetz.
   c. Perkins, G., Lee, J.H., Park, S., Kang, M. Ju, S., Phillips, G., Lysakowski, A., Gratton, M.A., and Yamoah, E.N. (2020) Altered outer hair cell mitochondrial and subsurface cisternae connectomics are emergent mechanisms for hearing-loss in mice. J. Neurosci. 40(44):8556–8572. PMC7605424
   d. Choi, S.H., Kim, K.Y., Perkins, G.A., Phan, S., Edwards, G., Xia, Y., Kim, J., Skowronska-Krawczyk, D., Weinreb, R.N., Ellisman, M.H., Miller, Y.I., Ju, W.K. (2020) AIBP protects retinal ganglion cells against neuroinflammation and mitochondrial dysfunction in glaucomatous neurodegeneration. Redox Biol. 37:101703.

4. In 2011 - 2013, four of my publications garnered press releases that mainstream online news outlets covered because of their breakthroughs of general interest. One figure from one of these papers was chosen by the French journal *La Recherche* as "one of the most impressive images of 2011" and was published in its January 2012 issue.
   a. Song, W., Chen, J., Petrilli, A., Liot, G., Klinglmayr, E., Zhou, Y., Poquiz, P., Tjong, J., Pouladi, M.A., Hayden, M., Masliah, E., Ellisman, M., Rouiller, I., Schwarzenbacher, R., Bossy, B., Perkins, G., Bossy-Wetzel, E. (2011) Mutant huntingtin binds the mitochondrial fission GTPase DRP1, and increases its enzymatic activity. Nature Medicine, 17:377-382. Cover Figure for the journal issue. PMC3051025

b. Nogueira, L., Ramirez-Sanchez, I., Perkins, G., Murphy, A., Taub, P.R., Ceballos, G., Villarreal, F.J., Hogan, M.C., and Malek, M.H. (2011) (-)-Epicatechin enhances fatigue resistance and oxidative capacity in mouse muscle. J. Physiol., 589.18: 4615-4631. PMC3208228
c. Yamaguchi, R. and Perkins, G. (2012) Finding a Panacea Among Combination Cancer Therapies, Cancer Research, 72:18-23. PMC3282559
d. Lee, Y.S., Morinaga, H., Kim, J.J., Lagakos, W., Taylor, S., Keshwani, M., Perkins, G., Dong, H., Kayali, A.G., Sweet, I.R. and Olefsky, J. (2013) The Fractalkine and CX3CR1 System Regulates Beta Cell Function: A Novel Pathway for Regulation of Insulin Secretion. Cell, 153:413-25. PMC3717389

5. As part of NCMIR, I have developed new tools for electron tomography.
   a. Mumcuoglu, E.U., Hassanpour, R., Tasel, S.F., Perkins, G., Martone, M. and Gurcan, M.N. (2012) Computerized Detection and Segmentation of Mitochondria on Electron Microscope Images, J. Microscopy, 246:248-65. PMID: 22506967
   b. Perkins, G. (2014) The use of miniSOG in the localization of mitochondrial proteins, In Anne N. Murphy, David C. Chan, editors: Mitochondrial Function, Vol 547, Methods in Enzymology, UK: Academic Press, pp. 165-179.
   c. Tasel, S.F., Mumcuoglu, E.U., Reza Z. Hassanpour, R.Z. and Perkins, G. (2016) A validated active contour method driven by parabolic arc model for detection and segmentation of mitochondria. J. Struct. Biol., 194:253-271. PMID: 26956730
   d. Sastri, M., Darshi, M., Mackey, M., Ramachandra, R., Ju, S., Phan, S., Adams, S., Stein, K., R. Douglas, C.R., Kim, J.J., Ellisman, M.H., Taylor, S.S., Perkins, G.A. (2017) Sub-Mitochondrial Localization of Genetic-Tagged MIB Interacting Partners: Mic19, Mic60 and Sam50. J. Cell Science, 30:3248-3260. Featured in “In This Issue.”

Complete List of Published Work in MyBibliography:
https://www.ncbi.nlm.nih.gov/myncbi/1fYyCv4kViD5t/bibliography/public/

BIOGRAPHICAL SKETCH

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NAME: Carsten Mim

eRA COMMONS USER NAME (credential, e.g., agency login): Carsten2020

POSITION TITLE: Associate 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
Johan Wolfgang Goethe Universität	Diploma	05/2002	Biochemistry
	PhD	11/2006	Biophysics

A. Personal Statement

I have a longstanding interest in processes at the membrane interface. I started my research as an electrophysiologist, where I became familiar with the molecular mechanism of solute transport through their respective proteins. Later, I transitioned into electron microscopy of membrane complexes to gain insight into structures at this important biological interface. In the laboratory of PostDoctoral supervisor, I learned about the isolation and reconstitution of transmembrane proteins. Since I started my own group, I have been working on structures of ion channels and their scaffolding proteins.

In total, I have 13 years of experience in electron microscopy (EM) and my laboratory works on projects that require different methods of electron microscopy; namely single particle EM, helical reconstruction EM and cryo-electron tomography. The topics in my group revolve around lipid-ion channel interactions and membrane associated proteins. I do not treat EM only as a means, but develop tools, like a ‘lab on a grid’ to move the field forward.

I am fortunate enough to be part of a network of structural biologists and computational biologists in Stockholm. However, my interest in working with different researchers resulted in funded research collaborations with scientists in different countries on different continents.

B. Positions and Honors

2007-2010	PostDoctoral Fellow	(Yale university)
20010-2014	PostDoctoral Fellow	(Northwestern University)
2015-2018	Assistant Professor	(Kungliga Tekniska Högskolan)
2018-present	Associate Professor	(Kungliga Tekniska Högskolan)

C. Contributions to Science

1. Function of plasma membrane Glutamate transporters

The only mechanism to terminate glutamatergic transmission is the removal of glutamate from the synaptic cleft. In humans there are four different (plasma membrane) glutamate transporters to achieve this task. All transporters are dependent on the Na^+/K^+ Gradient and have an anion conductance. Glutamate transporters fall into two categories either low affinity-high turnover or high affinity-low turnover. This lead to two questions

in the field: 1. Why are there so many transporters for one task? 2. What is the significance of the two categories of glutamate transporters?

To answer this question I characterized the transport cycle of a high affinity-low turnover transporter (EAAT4), to complement previous findings from my supervisor's laboratory. We discovered a voltage sensitive step in EAAT4 that slows the turnover rate at a hyperpolarized membrane potential and increases EAAT4's apparent affinity. Taken together, we think that EAAT4 acts as a glutamate sink in the periphery of the synapse. We also showed that the binding of glutamate is enthalpy-driven. Surprisingly this is not the case for the binding of Na^+. Our temperature dependent experiments revealed two distinct, kinetic states in the translocation step, which are characterized by high activation energy. We interpreted these events as the closing of the external gate and opening of the internal gate, respectively. Taken together our data indicate that glutamate transporters are fast enough to transport glutamate out of the cleft within the time scale of synaptic transmission. This contradicted the prevalent assumption that glutamate diffusion terminates the signal.

Two conformational changes are associated with glutamate translocation by the glutamate transporter EAAC1.
Mim C, Tao Z, Grewer C.
Biochemistry. 2007 Aug 7;46(31):9007-18. doi: 10.1021/bi7005465. Epub 2007 Jul 13.
PMID: 17630698 Free PMC article.

The glutamate transporter subtypes EAAT4 and EAATs 1-3 transport glutamate with dramatically different kinetics and voltage dependence but share a common uptake mechanism.
Mim C, Balani P, Rauen T, Grewer C.
J Gen Physiol. 2005 Dec;126(6):571-89. doi: 10.1085/jgp.200509365.
PMID: 16316976

2. Protein complexes on curved membranes

My work on the Bin/Amphyphysin/Rvs domain (BAR) protein endophilin in complex with the bilayer resulted in the unexpected discovery that the stability and dynamics of endophilin scaffolds entirely depend on non-specific interactions between amphipathic helices in the bilayer. My findings also provided a first structurally motivated hypothesis how BAR-scaffolds selectively recruit downstream interaction partners through steric selection mechanisms. We also showed that membrane perforation and fusion is a function of the local concentration of N-BAR proteins. My work provided more evidence that endophilin is fusogenic. The controlled nature of this phenomenon is an alternative avenue how cells can generate membranous structures. Further, the simulations showed that endophilin can act as a diffusion barrier for lipids and sort lipids in the nascent vesicles or membrane structures.

Protein-mediated transformation of lipid vesicles into tubular networks.
Simunovic M, **Mim C**, Marlovits TC, Resch G, Unger VM, Voth GA.
Biophys J. 2013 Aug 6;105(3):711-9. doi: 10.1016/j.bpj.2013.06.039.
PMID: 23931319 PMCID: PMC3736692

Understanding the role of amphipathic helices in N-BAR domain driven membrane remodeling.
Cui H, **Mim C**, Vázquez FX, Lyman E, Unger VM, Voth GA.
Biophys J. 2013 Jan 22;104(2):404-11. doi: 10.1016/j.bpj.2012.12.006.
PMID: 23442862 PMCID: PMC3552260

Structural basis of membrane bending by the N-BAR protein endophilin.
Mim C, Cui H, Gawronski-Salerno JA, Frost A, Lyman E, Voth GA, Unger VM.
Cell. 2012 Mar 30;149(1):137-45. doi: 10.1016/j.cell.2012.01.048.
PMID: 22464326 PMCID: PMC3319357

N-BAR New insights into BAR domain-induced membrane remodeling.
Ayton GS, Lyman E, Krishna V, Swenson RD, **Mim C**, Unger VM, Voth GA.
Biophys J. 2009 Sep 16;97(6):1616-25. doi: 10.1016/j.bpj.2009.06.036.
PMID: 19751666 PMCID: PMC2749773

3 Control of membrane bending by BAR domains

Despite their versatility, BAR domains are barely found on permanent membranous structures. The transversal tubular system in muscle cells is the exception. We could show that BIN1 alone generates t-tubule-like membrane topologies in vitro. It has been shown that myopathy causing mutations that eliminate electrostatic charges impair membrane bending, irrespective if a positive or negative charge is eliminated. We expanded on this finding and show that BIN1 bending of low curvature membranes is controlled by electrostatics. We show that a reduction of the membrane's surface charge reduces membrane bending. Further, electrostatics regulate localization of BIN1's interaction partners, eg Dynamin 2. These findings hold true in vitro as well as in cells. We speculate that cells control membrane bending and protein trafficking by manipulation of the membrane surface charge.

Cells Control BIN1-Mediated Membrane Tubulation by Altering the Membrane Charge.
Gowrisankaran S, Wang Z, Morgan DG, Milosevic I, **Mim C**.
J Mol Biol. 2020 Feb 14;432(4):1235-1250. doi: 10.1016/j.jmb.2019.12.001. Epub 2019 Dec 17.
PMID: 31857086

4. Structural biology of ex vivo clathrin coated vesicles

Previous studies of high resolution clathrin coats have been conducted on samples that have been assembled in vitro. For this report, we solved structures of clathrin coated vesicles isolated from animal tissues. Our reconstruction allows us to propose a steric-hindrance model how the vesicular ATPase is inhibited by the clathrin coat. This prevents the acidification (and maturation) of the synaptic vesicles until the clathrin coat is shed.

Clathrin coat controls synaptic vesicle acidification by blocking vacuolar ATPase activity.
Farsi Z, Gowrisankaran S, Krunic M, Rammner B, Woehler A, Lafer EM, **Mim C**, Jahn R, Milosevic I.
Elife. 2018 Apr 13;7:e32569. doi: 10.7554/eLife.32569.
PMID: 29652249 PMCID: PMC5935483

5. Activation of Wallerian Axon degeneration

SARM1 (sterile α and HEAT/armadillo motif–containing protein) is a protein that directly executes neuronal degeneration through its NADase activity. SARM1 knock-out mice show prolonged resistance for a form of neuronal degeneration (known as 'Wallerian Degeneration') resulting from insults, including viral infections, oxygen and glucose deprivation, and mechanical damage. Here we show that the protein forms a symmetrical assembly reminiscent of the assembly of apoptosomes and inflammamosomes. Impairment of the assembly interferes with the protein's function. Interestingly, NAD inhibits SARM1 and only after energetic stress and depletion of NAD SARM1 is activated.

Structural Evidence for an Octameric Ring Arrangement of SARM1.
Sporny M, Guez-Haddad J, Lebendiker M, Ulisse V, Volf A, **Mim C**, Isupov MN, Opatowsky Y.
J Mol Biol. 2019 Sep 6;431(19):3591-3605. doi: 10.1016/j.jmb.2019.06.030. Epub 2019 Jul 3.
PMID: 31278906

Structural basis for SARM1 inhibition and activation under energetic stress.
Sporny M, Guez-Haddad J, Khazma T, Yaron A, Dessau M, Shkolnisky Y, **Mim C**, Isupov MN, Zalk R, Hons M, Opatowsky Y.
Elife. 2020 Nov 13;9:e62021. doi: 10.7554/eLife.62021.
PMID: 33185189 PMCID: PMC7688312

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

Swedish Research Council (2016-03810)
Hebert (PI), Jegerschöld (Co-PI), **Mim (Co-PI)**, Köck (Co-PI)
01/2017-12/2020
'Structure analysis of biological macromolecules using electron microscopy'

Swedish Research Council (2018-06865)
Mim (PI)
01/2019-01/2020
Structural Biology of membrane proteins

Human Frontier Science Program; Young Investigator Grant (RGY0074/2016)
King (PI), **Mim (Co-PI)**, Yameen (Co-PI)
11/2016-10/2019
‘Building from scratch: How nanomaterials can help resolve membrane scaffold geometry and function’