BIOGRAPHICAL SKETCH

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NAME: Nathaniel J Traaseth

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

POSITION TITLE: Associate Professor of Chemistry

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 Minnesota-Duluth, Duluth, MN	B.S.	05/2003	Chemistry, Biochemistry, Political Science
University of Minnesota, Minneapolis, MN	Ph.D.	10/2007	Chemistry
University of Minnesota, Minneapolis, MN	Postdoc	04/2011	Biophysical Chemistry

A. Personal Statement

My research program is aimed at understanding how membrane proteins function within the confines of hydrophobic environments. My background in membrane proteins began at the University of Minnesota where I received extensive training in biochemistry, structural biology, and biophysics. During this time, I studied the molecular basis for understanding how small membrane proteins modulated the activity of Ca^{2+}-ATPase which is involved in cardiac and skeletal muscle contractility. In 2011, I started my independent position and have established an actively funded group with research interests in membrane protein efflux pumps that confer bacterial multidrug resistance (MDR) and receptor tyrosine kinases involved in human growth and development. My lab also actively develops NMR methods aimed at overcoming the hurdles of studying membrane protein structure and dynamics. These have included two recent novel methods for probing conformational exchange using solution NMR and oriented sample solid-state NMR. Overall, my research group is known for taking an interdisciplinary approach to study complex membrane protein systems. We are one of the few groups in the world that has the expertise to develop and apply solution NMR, magic-angle-spinning solid-state NMR, and oriented sample solid-state NMR in the study of membrane protein structure and dynamics. In addition, we recently ventured into cryoelectron microscopy and have established an active collaboration with Da-Neng Wang at the New York University School of Medicine. Taken together, our published findings and ongoing work provide strong support for our expertise to complete the goal of solving a high-resolution structure of NorA.

1. Gayen A, Leninger L, **Traaseth NJ** (2016) Protonation of a Glutamate Residue Modulates the Dynamics of the Drug Transporter EmrE. *Nat Chem Biol,* 12, 141-145. *PMCID: PMC4755857*
2. Cho MK, Gayen A, Banigan JR, Leninger M, **Traaseth NJ** (2014) Intrinsic conformational plasticity of native EmrE provides a pathway for multidrug resistance. *J Am Chem Soc*, 136, 8072-80. *PMCID: PMC4063181*
3. Chen H, Marsiglia WM, Cho M-K, Huang Z, Deng J, Blais SP, Gai W, Bhattacharya S, Neubert TA, **Traaseth NJ***, Mohammadi M* (2017) Elucidation of a four-site allosteric network in fibroblast growth factor receptor tyrosine kinases. eLife, 6, pii: e21137. doi: 10.7554/eLife.21137. *PMCID: PMC5293489*

4. Banigan JR, **Traaseth NJ** (2012). Using Afterglow Magnetization from Cross-Polarization Magic-Angle-Spinning Solid-State NMR Spectroscopy to Obtain Simultaneous Heteronuclear Multidimensional Spectra. J Phys Chem B, 116, 7138-44. *PMCID: PMC3418334*

B. Positions and Honors

Positions and Employment

2001-2003	Undergraduate Researcher, University of Minnesota-Duluth (C. Giulivi)
2003-2007	Graduate Student, Department of Chemistry, University of Minnesota (G. Veglia)
2007-2011	Post-Doctoral Associate, Department of Biochemistry, Molecular Biology, and Biophysics, University of Minnesota (G. Veglia)
2011-2017	Assistant Professor, Department of Chemistry, New York University
2017-present	Associate Professor, Department of Chemistry, New York University
2018-present	Director of Graduate Studies, Department of Chemistry, New York University

Honors

2003	American Institute of Chemist's Award
2003	Graduated with Honors from the Departments of Chemistry and Political Science
2003	Graduated *Magna Cum Laude*
2003-2004	Izaak Koltoff Fellowship
2004-2005	NIH Neuro-Physical-Computational Sciences Grant
2007	Overend Award in recognition of outstanding graduate student researcher physical chemistry
2005-2007	American Heart Association Pre-Doctoral Fellowship
2008	Paul Boyer Award for significant research contributions in biochemistry as a postdoc.
2010	NIH NRSA Postdoctoral Fellowship (*declined*)
2011-2013	NIH (NIAID) National Research Scholar Development K22 Award

C. Contribution to Science

1. Transport mechanisms of multidrug efflux pumps. A long-term goal of my lab is to reveal novel mechanisms used by multidrug transporters and to harness this knowledge to predict and control function, including in design of conformationally specific efflux pump inhibitors. Our approach is to study transporters within lipid bilayers, which provides the best mimic of the native environment and offers conditions in which dynamics can be quantitatively measured. Our studies have primarily focused on EmrE, which serves as a model transporter from the small multidrug resistance family to understand the ability of multidrug pumps to promiscuously bind substrates and to use anionic residues to couple with the pH gradient. We discovered that the drug-free form of the transporter exhibits significant conformational plasticity that we hypothesize enables it to bind and efflux a wide variety of substrates varying in size, shape, and charge. Our latest findings were used to derive a correlation between the acid dissociation constant of a conserved glutamic acid residue and the phenotype conferred to *E. coli*. We found that an elevated acid dissociation constant was needed to transport and provide resistance to toxins. We also discovered that the protonation state plays a role in modulating allostery and in the global conformational exchange required for transfer of drugs across the membrane. These findings were used to derive a model whereby we hypothesize that the inward-open conformation of the transporter is favored in the presence of a pH gradient. Namely, when the pH values on the cytoplasm and periplasm are centered around those of the acid dissociation constant, EmrE is positioned toward the cytoplasm and poised to bind to toxic compounds. This leads to a preferred resting conformation of the transporter, which is a novel finding in the transport field.

(a) Leninger M, Sae Her A, **Traaseth NJ** (2019) Inducing Conformational Preference of the Membrane Protein Transporter EmrE Through Conservative Mutations. *eLife*, 8:e48909. *PMCID: PMC6805155*

(b) Gayen A, Leninger L, **Traaseth NJ** (2016) Protonation of a Glutamate Residue Modulates the Dynamics of the Drug Transporter EmrE. *Nat Chem Biol,* 12, 141-145. *PMCID: PMC4755857*

(c) Cho MK, Gayen A, Banigan JR, Leninger M, **Traaseth NJ** (2014) Intrinsic conformational plasticity of native EmrE provides a pathway for multidrug resistance. *J Am Chem Soc*, 136, 8072-80. *PMCID: PMC4063181*

(d) Gayen A, Banigan JR, **Traaseth NJ** (2013). Ligand-Induced Conformational Changes of the Multidrug Resistance Transporter EmrE Probed by Oriented Solid-State NMR Spectroscopy. *Angew Chemie Int Ed*, 52, 10321-4. *PMCID: PMC3876743*

2. NMR methods for probing structure and dynamics of membrane proteins. My group has actively developed solution and solid-state NMR methods to be used to acquire high-resolution structural information for membrane proteins. While the solid-state NMR approach is a preferred way to study membrane protein structure and dynamics due to the ability to characterize the proteins in lipid bilayers, the two problems associated with widespread usage are the reduced spectral resolution and sensitivity. We developed new approaches to address both limitations. The first is the *Afterglow* method that makes use of residual magnetization from the cross-polarization sequence that is widely used in solid-state NMR. This magnetization is stored and used for acquisition of a second dataset, thus improving the sensitivity by a significant amount (i.e., *two for the price of one*). The second method we developed is a set of isotopic labeling schemes to be used in conjunction with the *Afterglow* technology. In this approach, we create a defined set of pairwise connectivity through the careful choice of label incorporation and apply spectroscopic filters to reduce the spectral complexity. These developments have been instrumental in increasing the speed of obtaining structural information for large, polytopic membrane transport proteins. In addition, we have developed a novel technique that can reduce the time needed to acquire chemical exchange saturation transfer (CEST) experiments that are used to probe motion on the msec to sec timescale. These approaches are time consuming because they require a series of 2D datasets (i.e., pseudo 3D acquisition). Recently, we introduced a multifrequency approach for CEST that can reduce the data acquisition by an integer factor of 3-4-fold, which will enable solution NMR users a way to *speed-up* data acquisition.

(a) Leninger M, Marsiglia WM, Jerschow A, **Traaseth NJ** (2018) Multiple frequency saturation pulses reduce CEST acquisition time for quantifying conformational exchange in biomolecules. *J Biomolec NMR*, 71, 19-30. *PMCID: PMC5989009*

(b) Banigan JR, Gayen A, **Traaseth NJ** (2015) Correlating Lipid Bilayer Fluidity with Sensitivity and Resolution of Polytopic Membrane Protein Spectra by Solid-State NMR Spectroscopy. *Biochim Biophys Acta*, 1848, 334-41. *PMCID: PMC4438312*

(c) Banigan JR, Gayen A, **Traaseth NJ** (2013). Combination of ^{15}N Reverse Labeling and Afterglow Spectroscopy for Assigning Membrane Protein Spectra by Magic-Angle-Spinning Solid-State NMR: Application to the Multidrug Resistance Protein EmrE. *J Biomolec NMR*, 55, 391-9. *PMCID: PMC3747971*

(d) Banigan JR, **Traaseth NJ** (2012). Using Afterglow Magnetization from Cross-Polarization Magic-Angle-Spinning Solid-State NMR Spectroscopy to Obtain Simultaneous Heteronuclear Multidimensional Spectra. J Phys Chem B, 116, 7138-44. *PMCID: PMC3418334*

3. Role of intrinsic kinase dynamics in biological activity. A second biological project in the lab is aimed at elucidating allosteric control mechanisms underpinning receptor tyrosine kinase (RTK) regulation by using the fibroblast growth factor receptor (FGFR) kinase subfamily as the model system. RTKs represent remarkable examples of systems that facilitate long-range communication in cell biology, taking place between an extracellular ectodomain and an intracellular kinase via a single-pass transmembrane (TM) segment. Toward our goal, we have established a strong collaboration with Moosa Mohammadi, to discover novel allosteric control mechanisms within the FGFR system, which is translatable to other RTKs. In our initial work at NYU, we discovered that pathogenic mutations in the kinase activation loop (A-loop) altered the conformational dynamics, which shifted the equilibrium toward the active form of the enzyme by introducing intramolecular contacts that stabilize the active state. This article received an F1000 recommendation (http://f1000.com/prime/718043498). Recently, we proposed a detailed allosteric network involving four key hotspots that are involved in regulating the transition between autoinhibited and activated states. Furthermore, we have shown how pathogenic mutations at the kinase hinge bypass the autoinhibitory network at the molecular brake to lead to activation. Localized conformational perturbations triggered by mutations propagate to the active state through an

intermediary isoleucine that influences the position of the phenylalanine from the conserved DFG motif. This propagation results in the destabilization of the autoinhibited state and a corresponding shift in the equilibrium toward the activated form. Comparative structural analyses to other tyrosine kinases provided evidence that this allosteric mechanism constitutes a shared mechanism across the tyrosine kinase family.

(a) Marsiglia WM, Katigbak J, Zheng S, Mohammadi M, Zhang Y, Traaseth NJ (2019). A Conserved Allosteric Pathway in Tyrosine Kinase Regulation. Structure, *in-press*. doi: 10.1016/j.str.2019.05.002. *PMID: 31204250*

(b) Chen H, Marsiglia WM, Cho M-K, Huang Z, Deng J, Blais SP, Gai W, Bhattacharya S, Neubert TA, **Traaseth NJ***, Mohammadi M* (2017) Elucidation of a four-site allosteric network in fibroblast growth factor receptor tyrosine kinases. eLife, 6, pii: e21137. doi: 10.7554/eLife.21137. *PMCID: PMC5293489*

(c) Huang Z, Marsiglia WM, BasuRoy U, Rahimi N, Ilghari D, Wang H, Chen H, Gai W, Blais S, Neubert TA, Mansukhani A, **Traaseth NJ**, Li X, Mohammadi M (2016). Two FGF Receptor kinases act in concert to recruit and transphosphorylate PLCγ. *Molecular Cell*, 61, 98-110. *PMCID: PMC4838190*

(d) Chen H, Huang Z, Dutta K, Blais S, Neubert TA, Li X, Cowburn D, **Traaseth NJ**, Mohammadi M (2013) Cracking the Molecular Origin of Intrinsic Tyrosine Kinase Activity through Pathogenic Gain-of-Function Mutations. *Cell Reports*, 4(2), 376-84. *PMCID: PMC3752781*

4. Molecular mechanism of cardiac contractility regulation by phospholamban. Prior to my independent career at NYU, my research focused on elucidating the regulatory mechanism on the Ca^{2+}-ATPase (SERCA) from phospholamban (PLN). PLN is a single-pass membrane protein that inhibits SERCA, an ATP-driven pump that translocates calcium ions into the lumen of the sarcoplasmic reticulum, initiating muscle relaxation. PLN binds SERCA through intramembrane interactions, impeding calcium translocation. While phosphorylation of PLN at Ser-16 and/or Thr-17 reestablishes calcium flux, the molecular details of inhibition and relief of inhibition remained elusive prior to our studies. While SERCA had been crystallized in several different states along the enzymatic reaction coordinates, the lack of high-resolution crystals in the presence of PLN and SLN limited the understanding of the regulatory mechanism. Our use of NMR and EPR experiments showed that PLN regulates SERCA through a series of conformational rearrangements involving membrane interactions and the folding and unfolding of domains within the juxtamembrane and cytoplasmic regions of PLN. These results have improved our understanding of the calcium translocation process and are the basis for designing novel therapeutic approaches to ameliorate muscle malfunctions.

(a) **Traaseth NJ**, Verardi R, Torgersen KD, Karim CB, Thomas DD, Veglia G (2007). Spectroscopic Validation of the Pentameric Structure of Phospholamban. *Proc Natl Acad Sci,* 104, 14676-81. *PMCID: PMC1976191*

(b) Gustavsson M, Verardi R, Mullen DG, Mote KR, **Traaseth NJ**, Gopinath T, Veglia G. (2013) Allosteric Regulation of SERCA by Phosphorylation-Mediated Conformational Shift of Phospholamban. *Proc Natl Acad Sci*, 110, 17338-43. *PMCID: PMC3808617*

(c) Verardi R, Shi L, **Traaseth NJ,** Walsh N, Veglia G (2011) Structural Topology of Phospholamban Pentamer in Lipid Bilayers by a Hybrid Solution and Solid-State NMR Method. *Proc Natl Acad Sci,* 108, 9101-6. *PMCID: PMC3107283*

(d) Traaseth NJ, Thomas DD, Veglia G (2006) Effects of Ser16 phosphorylation on the allosteric transitions of phospholamban/Ca(2+)-ATPase complex. *J Mol Biol*, 358, 1041-50. *PMID: 16564056*

5. Structure Determination Protocol for Membrane Proteins. To fully describe the fold space and ultimately the biological function of membrane proteins, it is necessary to determine the specific interactions of the protein with the membrane. As a post-doctoral associate, I spearheaded work to develop a structure determination protocol that simultaneously defined the structure, orientation, and depth of insertion of membrane proteins in the lipid bilayer. This hybrid objective function was incorporated into XPLOR-NIH and was the first to combine solution and solid-state NMR restraints in a quantitative fashion to calculate membrane protein structures with respect to the lipid bilayer. This approach was originally demonstrated with the cardiac membrane protein phospholamban and has since been applied to other systems.

(a) **Traaseth NJ**, Shi L, Verardi R, Mullen D, Barany G, Veglia G (2009) Determination of Membrane Protein Structure and Topology Using a Hybrid Solution and Solid-State NMR approach. *Proc Natl Acad Sci* 106, 10165-70. *PMCID: PMC2700893*

(b) Shi L, **Traaseth NJ**, Verardi R, Gustavsson M, Gao J, Veglia G (2011) Paramagnetic-based NMR restraints lift residual dipolar coupling degeneracy in multidomain detergent-solubilized membrane proteins. *J Am Chem Soc,* 133, 2232-41. *PMCID: PMC3328396*

(c) Shi L, **Traaseth NJ**, Verardi R, Cembran A, Gao J, Veglia G (2009) A refinement protocol to determine structure, topology, and depth of insertion of membrane proteins using hybrid solution and solid-state NMR restraints. *J Biomol NMR,* 44, 195-205. *PMCID: PMC2824793*

(d) **Traaseth NJ**, Verardi R, Veglia G (2008) Asymmetric methyl group labeling as a probe of membrane protein homo-oligomers by NMR spectroscopy. *J Am Chem Soc,* 130, 2400-1. *PMCID: PMC2699765*

Complete List of Published Work in My Bibliography (a total of 60 peer-reviewed publications):

http://www.ncbi.nlm.nih.gov/sites/myncbi/nathaniel.traaseth.1/bibliography/47380351/public/?sort=date&direction=descending

D. Research Support

ONGOING RESEARCH SUPPORT

1. R01 AI108889 07/15/14 – 06/30/20 (NCE) Traaseth (PI)
NIH-NIAID
Mechanisms of Allostery and Molecular Recognition in the Small Multidrug Resistance Family
The goal of this study is to elucidate the allosteric basis for multidrug resistance in bacteria conferred by the small multidrug resistant family of transporters.

2. R01 GM117118 01/01/17 – 12/31/20 Traaseth (PI), Mohammadi (PI)
NIH-NIGMS
FGF Receptor Structure, Dynamics and Function
The goal of the proposed project is to reveal intracellular allosteric control mechanisms present in receptor tyrosine kinase cellular signaling by using the FGF receptor subfamily as the system. This is a multi-PI grant with Dr. Moosa Mohammadi (School of Medicine, NYU). Dr. Traaseth is the contact PI on the grant.
Role: contact PI

3. R01GM117118-03S1 01/01/17 – 12/31/20 Traaseth (PI), Mohammadi (PI)
NIH-NIGMS
FGF Receptor Structure, Dynamics and Function
This is an equipment supplement for addition of a helium recovery system. This system will work in a shared instrumentation facility to collect helium gas boil-off from several NMR magnets.
Role: contact PI

4. NSF MCB1506420 08/01/19 – 07/31/22 Traaseth (PI)
Directorate for Biological Sciences - MCB
Tools for Probing Conformational Dynamics of Membrane Proteins
The goal of this award is to develop NMR tools to study membrane proteins dynamics.

5. R01 DE013686 07/01/16 – 03/31/21 Mohammadi (PI)
NIH-NIDCR
Mechanisms of FGF receptor regulation and signaling

The overall goal of this study is to elucidate the structural mechanisms that regulate FGF receptor activation and signaling. Specifically, my lab's role in this project is to carry out NMR experiments to investigate the kinase dimer that mediates transphosphorylation within the activation loop.
Role: Co-Investigator

6. NSF CHE1827902 08/01/18 – 07/31/21 Diao (PI), Traaseth (co-PI)
Division of Chemistry, Chemical Instrumentation
MRI: Acquisition of an Electron Paramagnetic Resonance Spectrometer
The goal of this award is to acquire an EPR spectrometer to be housed in a shared instrumentation facility for research and education.

7. I-1155 02/01/20-01/31/22 Arora (PI)
CHDI Foundation
Rational Design and Screening Strategies to Identify Proteomimetic Ligands that Modulate Huntington Protein Synthesis and Aggregation
The goal of this work is to design small molecule modulators of aggregation prone proteins involved in Huntington's disease. The Traaseth lab will characterize complexes involving the small molecules with the aggregated proteins.
Role: Co-Investigator

BIOGRAPHICAL SKETCH

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NAME: Brawley, Douglas

eRA COMMONS USER NAME (credential, e.g., agency login): dnb303

POSITION TITLE: Graduate Student Research Assistant

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)*	Start Date MM/YYYY	Completion Date MM/YYYY	FIELD OF STUDY
UNC Asheville	BS	08/2008	05/2013	Biology
NYU School of Medicine	PhD	08/2014	Present	Molecular Biophysics

A. Personal Statement

Since childhood I have always been trying to understand how things work. Today that child-like curiosity is directed at the elegant, robust, and efficient machines within our cells known as proteins. My interest in protein structure and function developed in college and motivated me to engage in undergraduate research in the laboratory of Dr. Ted Meigs. There I investigated protein-mediated signaling cascades implicated in metastatic transformation. To identify key residues mediating signal transduction, I engineered numerous point mutations in the alpha subunit of the heterotrimeric guanine nucleotide binding protein, Galpha12, and tested these mutants for interaction with downstream effector proteins by co-precipitation experiments and *in-vivo* reporter assays. This work culminated in a publication in the *Journal of Molecular Signaling* where we elucidated a novel mechanism for Galpha12 stimulation of the RhoGEF-Rho signaling pathway. While this discovery was exciting, I knew that I would not be able to fully understand how proteins interact, or more broadly, the molecular mechanisms governing other important biological processes, without actually being able to “see” them. This realization drove me to pursue a PhD in Molecular Biophysics so I could receive training in the biophysical techniques commonly employed to determine high-resolution structures of proteins.

Presently I am a 6th year doctoral student in the Molecular Biophysics Graduate Program at NYU School of Medicine. In the Traaseth Lab, I am using both biophysical and biochemical techniques to investigate the molecular basis of antibiotic resistance through multi-drug efflux pumps in pathogenic bacteria. Specifically, I am interested in elucidating the structure and function of a multidrug efflux transporter, termed NorA, that is overexpressed in the membrane of methicillin-resistant *S. aureus* (MRSA). For structure determination work, I have been collaborating with two additional structural biology labs with expertise in cryo-electron microscopy (Dr. Da-Neng Wang’s group) and antibody engineering (Dr. Shohei Koide’s group). The near-term goal of the project is to determine a high-resolution structure of NorA by cryoEM (~ 3.0-3.5Å) which will complement existing functional data and provide new insight into the transport mechanism.

B. Positions and Honors

Positions and Employment

2011 - 2013 Undergraduate Student Researcher, University of North Carolina at Asheville

2014 – Graduate Student Research Assistant, NYU School of Medicine

Other Experience and Professional Memberships

2016 - 2019	Co-president, Molecular Biophysics Seminar Committee
2016 -	Class liaison, Sackler Student Council at NYU School of Medicine
2017 -	Member, Students Advocating for Science, Education and Medicine (SASEM)
2017 -	Leadership, NYU Biotech Association and GRO-Biotech

Honors

2008- 2013	Dean's List, UNC Asheville
2011- 2012	Undergraduate Biotechnology Research Fellowship, NC Biotech Center
2013	Bernhardt-Perry Award for Excellence in Undergrad Research in Biology, UNC Asheville
2013	Distinction as an University Research Scholar, UNC Asheville
2013	University-wide honors, B.S. *Cum Laude*, UNC Asheville
2016- 2018	NIH T32 Training Grant Fellowship, NYU School of Medicine
2017	Special McCraken Award for Leadership and Scholastic Achievement, NYU School of Medicine
2017	Best Presentation Award at the Molecular Biophysics Graduate Student Symposium, NYU School of Medicine

C. Contributions to Science

Undergraduate Research: I engaged in two separate projects while in Dr. Ted Meigs' Lab at UNC Asheville. The long-standing goal of the Meigs Lab is to better understand the role of G-protein alpha subunits, particularly Galpha12 and Galpha13, in cancer-implicated signal transduction pathways. More specifically, the Lab aims to elucidate the structural determinants on Galpha12 and Galpha13 that mediate interaction with upstream activating proteins and downstream effector proteins in different cell signaling cascades. In my first project, I investigated the molecular basis for Galpha12 activation by a cytosolic guanine nucleotide exchange factor (GEF), termed Ric-8. Using a combination of scanning mutagenesis and GST pulldown assays, I identified a nine amino acid stretch on the C-terminus of Galpha12 necessary, but not sufficient, for interaction with Ric-8. These studies have since spurred further investigation by Dr. Steve Rogers' Lab at UNC Chapel Hill to test whether the homologous deletion introduced in the *Drosophila* Galpha12 homolog, Concertina (Cta), affects its ability to mediate a well-characterized signaling pathway stimulating cell contractility. In my second project, I engineered a new epitope tagging system permitting side-by-side comparison of the binding affinities of Galpha12 and Galpha13 for different effector proteins. These studies revealed key differences in the specificities of Galpha12 and Galpha13 for downstream signaling partners suggesting that these proteins stimulate both distinct and overlapping cellular signal transduction pathways.

a) UNC-Asheville Annual Spring Symposium on Undergraduate Research and Creativity, April 2012. *Structural Characterization of Gα12 Interaction with the Non-receptor Activator Ric-8* (Oral presentation)
b) UNC-Asheville Annual Spring Symposium on Undergraduate Research and Creativity, April 2013. *Fluorescent protein and epitope tagging within the helical domain of G12 subfamily proteins* (Oral presentation)
c) Experimental Biology 2013. Boston, MA, April 2013. *Fluorescent protein and epitope tagging within the helical domain of G12 subfamily proteins* (Poster presentation)
d) Martin, J.W.*, Cavagnini, K.S.*, **Brawley, D.N.**, Berkley, C.Y., Smolski, W.C., Garcia, R.D., Towne, A.L., Sims, J.R., Meigs, T.E, A Gα12-specific Binding Domain in AKAP-Lbc and p114RhoGEF. Journal of Molecular Signaling, 2016. **11** (3): p. 1–17

Graduate Research: My current research in the Traaseth Lab at NYU seeks to more thoroughly understand the molecular basis of antibiotic resistance mediated by multi-drug efflux transporters in pathogenic bacteria. Specifically, I am interested in elucidating the structure and function of NorA, a multi-drug efflux pump overexpressed in the membrane of methicillin-resistant *S. aureus* (MRSA). NorA confers resistance to a broad spectrum of compounds including fluoroquinolones, quaternary ammonium compounds, and ethidium bromide.

While basic structural insight can be derived from available high-resolution structures of homologous transporters, an atomistic understanding of the key structural determinants governing substrate binding, energy coupling, and conformational exchange remains unavailable. To that end, I am using cryo-electron microscopy (Cryo-EM) to determine the atomic structure of NorA which will complement data from functional studies to provide new insight into the transport mechanism.

a) Molecular Biophysics Graduate Program Symposium, NYU Med. Cntr., May 2018. *Mechanism of the multidrug efflux pump NorA* (poster presentation)
b) New York City Area CryoEM Meeting, CUNY ASRC, October 2019. *Towards the structure of the multidrug efflux pump NorA* (poster presentation)
c) Skirball Institute Retreat, NYU School of Medicine, October 2019. *Structure and function of the multidrug efflux pump NorA* (poster presentation)

BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors.
Follow this format for each person. **DO NOT EXCEED FIVE PAGES.**

NAME: David B. Sauer, Ph.D.

eRA COMMONS USER NAME (credential, e.g., agency login): sauerd01

POSITION TITLE: Postdoctoral Fellow

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
Purdue University, West Lafayette, IN	B.S.	8/2005	Chemistry
UT Southwestern Medical Center, Dallas, TX	Ph.D.	6/2012	Molecular Biophysics
New York University School of Medicine, NY, NY	Postdoc	present	Structural Biology

A. Personal Statement

I have been studying cell membranes for 17 years, focusing on the structure and mechanism of membrane proteins. My most recently completed work focused on the mechanisms of ion channel folding and selectivity utilizing high-resolution X-ray crystallography in conjunction with bioinformatics and functional studies. My current work aims to address the mechanisms of prokaryotic and eukaryotic membrane transporters by X-ray crystallography and Cryo-Electron Microscopy.

B. Positions and Honors

Positions and Employment

2002 Undergraduate Research Assistant, Department of Chemistry, Univ. of Missouri St. Louis
2003 – 2005 Undergraduate Research Assistant, Department of Chemistry, Purdue University
2006 – 2012 Graduate Research Assistant, Department of Physiology, UT Southwestern
2012 – present Postdoctoral Fellow, Skirball Institute of Biomolecular Medicine, NYU School of Medicine

Honors

2004 Best Science Paper Award, Butler University and Eli Lilly
2004 Harrison M. Stine Memorial Scholarship, Purdue University Department of Chemistry
2005 Dale W. Margerum Undergraduate Research Award, Purdue University Dept. of Chemistry
2005 Outstanding Research Award, Purdue Undergraduate Research and Poster Symposium
2007 – 2010 Predoctoral Training Grant, NIH and UT Southwestern Molecular Biophysics
2013 Outstanding Poster Presentation, New York University Skirball Institute Retreat
2014 Biophysics Wiki-edit Contest Winner, Biophysical Society
2019 Poster Award, New York Structural Biology Discussion Group

Professional Activities

2019 – present NYU Medical Center Cryo-EM Discussion Group Organizer
2019 NYU Medical Center Skirball Institute Retreat Organizing Committee
2018 – present F1000Prime Associate Faculty Member

2004 Purdue Science Student Council President
2002 – 2005 Purdue Science Student Council Member

C. Contributions to Science

My interests in science have focused on the biophysical properties of the cellular membrane, with a particular bent toward using structure to describe the mechanics of transport across the lipid bilayer.

My first publication, through undergraduate research, examined basic biophysical properties of the lipid membrane itself. This demonstrated the effects of lipid chemistry and shape on the thermodynamic properties of the lipid bilayer.

1. M.C. Hull, D.B. Sauer, and J.S. Hovis. "The Influence of Lipid Chemistry on the Osmotic Response of Cell Membranes: Effect of Non-Bilayer Forming Lipids," J. Phys. Chem. B, 108, 15890 -15895 (2004).

Potassium channels set the resting potential of the cell and are thus critical to the basic physiology of all excitable cell types. While fundamental, potassium channel selectivity and channel gating are areas of continued debate. While much had been done addressing the physical properties of ion coordination, the uniformity of selectivity filters solved limited our understanding of site number in selectivity. I solved the structure of a number of engineered channels of varying selectivity. This work noted for the first time that the role of filter structure, stability, and binding site number in K+ selectivity. The differences in ion binding, despite identical filter structure, were later addressed in careful crystallographic titration experiments. Further, I was involved in describing the first structure of a full length, dual-RCK domain. This provided new insights into the mechanics of multi-ligand binding and channel gating. This structure has been important in understanding the similarly dual-RCK domain containing human BK channel. Collectively, this work provides new structural insights into potassium channel folding, ion selectivity, and channel gating.

2. M.G. Derebe*, D.B. Sauer*, W. Zeng, A. Alam, N. Shi and Y. Jiang. "Tuning the Ion Selectivity of Tetrameric Cation Channels by Changing the Number of Ion Binding Sites" Proc. Natl. Acad. Sci. USA, 108, 598-602 (2011). *- authors contributed equally
3. D.B. Sauer, W. Zeng, S. Raghunathan and Y. Jiang. "Protein Interactions Central to Stabilizing the K+ Channel Selectivity Filter in a 4-sited Configuration for Selective K+ Permeation" Proc. Natl. Acad. Sci. USA, 108, 16634-16639 (2011).
4. C. Kong, W. Zeng, S. Ye, L. Chen, D.B. Sauer, Y. Lam, M.G. Derebe, Y. Jiang. "Distinct Gating Mechanisms revealed by the structures of a multi-ligand gated K+ channel" eLife, 1 (2012).
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During my graduate studies I also contributed to the first structural characterization of the Sodium/Calcium Exchanger (NCX) family. Using this structure the unique means of ion exchange was described, and in doing so explain the remarkably high transport rate of the NCX family.

7. J. Liao, H. Li, W. Zeng, D.B. Sauer, R. Belemares, Y. Jiang. "Structural Insight into the Ion Exchange Mechanism of Sodium/Calcium Exchanger" Science, 335, 686-690 (2012).

Additionally, I developed multiple novel, yet general, bioinformatic methods for identifying thermostable proteins and thermostabilizing mutants. These methods have been validated on a protein otherwise intractable to structural study. These methods offer the opportunity for the quickly and efficiently improving the biochemical stability of a target protein, which has been crucial in successful structural determination of difficult membrane proteins.

8. D.B. Sauer[§], N.K. Karpowich, J. Song, D.N. Wang[§] "Rapid Bioinformatic Identification of Thermostabilizing Mutations" Biophys. J., 109, 1420-1428 (2015). [§] - Co-corresponding authors

9. D.B. Sauer[§] and D.N. Wang[§] “Predicting the Optimal Growth Temperature of Prokaryotes using only Genome Derived Features” Bioinformatics, doi:10.1093/bioinformatics/btz059 (2019).
10. D.B. Sauer[§] and D.N. Wang[§] “Using machine learning to predict quantitative phenotypes from protein and nucleic acid sequences” BioRxiv doi:10.1101/677328 (preprint)

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

Completed Research Support

Horizon Award, Department of Defense — Sauer (PI) — 2017 – 2019
Structure and Function of the Reduced Folate Carrier
The goal of this project was to structurally and functionally characterize the human Reduced Folate Carrier.
Role: PI

Postdoctoral Fellowship, American Cancer Society — Sauer (PI) — 2016 – 2017
Structure and Function of the Reduced Folate Carrier
The goal of this project was to structurally and functionally characterize the human Reduced Folate Carrier.
Role: PI