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: Victoria Robinson

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

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
Trinity College, Hartford CT	BS	05/1988	Biochemistry
Villanova University, Villanova PA	MS	05/1992	Chemistry
University of Iowa, Iowa City IA	PHD	12/1997	Biochemistry/Structural Biology
HHMI/UMDNJ-RWJMS, Piscataway NJ	Post-Doc	08/2004	Structural Biology/ Microbiology

A. Personal Statement

The goal of the research in my laboratory is to understand the relationship between events triggered from the onset of stress and cellular survival. We are particularly focused on how protein biogenesis, an inherently taxing process, is regulated to conserve resources and to support adaption to adverse environmental factors ensuring homeostasis. We have many years of experience deciphering the structural and biochemical properties of proteins. Although, considered a protein structure laboratory (protein crystallography and cryoEM) laboratory, there is a wide range of experience in my group utilizing biophysical techniques, such as fluorescence spectroscopy, ITC, MST, small angle scattering (SAXS), CD, as well as numerous biochemical assays to examine enzymatic properties of proteins and their interactions with various cellular partners. Current projects are centered around studying the interaction between the ribosome and two unusual GTPases. The first is BipA, a GTPase required for regulating physiologic responses in bacteria upon the onset of adverse growth conditions. The second is a circularly permuted GTPase, nucleostemin, which shuttles from the nucleolus to the cytoplasm to support the maturation of the 60S ribosomal subunit. We are utilizing many techniques to understand how biochemical properties of these proteins regulate their cellular function focusing on how they help organisms adapt to the onset of stress.

For several years**,** my career was disrupted due to extreme elder care responsibilities followed COVID. My research program is again well established and over the past two years, has grown to include four Ph.D. students.

Ongoing Research Support

Department of Defense Robinson (co-I) 1/22/20 to 12/31/2023
Cytoplasmic Suppression of Inflammatory Signaling
Define the intrinsic disorder of tNIP1 protein and its role in regulating A20 partnering, ubiquitin-binding and inflammatory pathways in cultured cells.

UConn Research Excellence Award Heaslip, Robinson (Co-Is) 06/15/23 to 06/15/24
UConn Office of Vice President for Research (OVPR)

Development of new anti-parasitic drugs targeting *Toxoplasma gondii*
We are validating BipA, an essential protein in *Toxoplasma gondii*, as a target for anti-parasitic therapeutics.

Atomwise AIMS and subsequent AIMS Prime Award Robinson (PI) 12/1/18 - present
Virtual Screening for Inhibitors of the Bacterial Translation Factor BipA
Working with scientist at Atomwise to utilize virtual screening and docking with their proprietary AtomNet software to identify small molecules which modulate the biochemical activities of BipA.

Completed Research Support

START Preliminary Proof-of-Concept Fund Robinson (PI) 12/31/19 to 12/31/22
UConn Office of Vice President for Research (OVPR)
Structure Based Virtual Screening to Uncover Inhibitors of BipA
These funds are to continue our work characterizing small molecule inhibitors of BipA as potential antimicrobials.

START Preliminary Proof-of-Concept Fund Heaslip, Robinson (Co-PIs) 5/31/21 to 06/30/22
UConn Office of Vice President for Research (OVPR)
Development of new anti-parasitic drugs targeting *Toxoplasma gondii*
We are validating BipA, an essential protein in Toxoplasma gondii, as a target for anti-parasitic therapeutics.

UConn Research Excellence Program Award Robinson, Benson (co-PIs) 7/1/18 to 6/30/22
Harmonizing Physiology with Structural Biology Approaches to Define the Roles of the BipA in Translation
The goal of this research is to define the biochemical features of BipA that support its role in stress adaptation.

START Preliminary Proof-of-Concept Fund Robinson (PI) 4/01/19 to 3/31/21
UConn Office of Vice President for Research (OVPR)
Investigating Host-Pathogen Interactions Regulated by BipA for Antimicrobial Development
Using an enterohemorrhagic *Escherichia coli* (EHEC) model system we are determining how BipA is regulating actin pedestal formation associated with host-pathogen interactions and colonization .

AG161195 Lynes (PI) 9/1/17 to 8/31/20
Connecticut Innovations
A Therapeutic Monoclonal Antibody for the Treatment of Inflammatory Bowel Disease
The goal of this project is to determine how an antibody raised against the stress response protein metallothionine (MT) influences the progression of the immune response.
Role: Co-Investigator

R21 AI104841 Wright (PI), Robinson (co-I) 06/15/2018 to 05/31/2020
Targeting Glycolytic Enzymes as an Antibacterial Strategy
The goal is to explore if nonbenzenoid aromatic tropolone natural products are exceptional candidates for developing cell-permeable enolase inhibitors and if differences at either the target or pathway level can be exploited for selective blockade of the bacterial glycolytic machinery.
Role: Co-Investigator

PITCH2 Award Robinson (PI) 04/1/19 to 12/31/20
Program for Innovative Therapeutics for Connecticut's Health (PITCH) Stage 2
Connecticut Innovations
High Throughput Screening to Identify Small Molecule Inhibitors of BipA
Support the physiochemical characterization of small molecule effectors of BipA and determine key structure-activity relationships.

B. Positions, Scientific Appointments, and Honors

Positions and Employment:

2022-present	Associate Dept. Head, Graduate Research and Education, MCB Dept., UConn Storrs.
2017-present	Director, UConn Professional Master's Program in Applied Biochemistry and Cell Biology
2010-present	Associate Professor, Dept. of Molecular and Cell Biology, University of Connecticut, Storrs
2004-2010	Assistant Professor, Dept. of Molecular and Cell Biology, University of Connecticut, Storrs
1999-2004	Post-doctoral Associate, HHMI/ UMDNJ-RWJMS
1998	Post-doctoral Fellow, SmithKline Beecham Pharmaceuticals, King of Prussia, PA
1992-1997	Graduate Research Assistant, University of Iowa
1990-1992	Assistant Scientist, Rhone-Poulenc Rorer Biotechnology, Inc., King of Prussia, PA
1988-1990	Research Assistant, Rhone-Poulenc Rorer Biotechnology, Inc., King of Prussia, PA

Honors and Awards:

2009	Faculty Early Career Development Award, National Science Foundation
2006	American Heart Association (AHA) Scientist Development Grant
2001	Research Achievement Award, UMDNJ-RWJMS
1994-1997	Predoctoral Fellowship, Univ. of Iowa Center for Biocatalysis and Bioprocessing
1994	American Heart Association (AHA) Predoctoral Fellowship (declined)

Other Experience and Professional Memberships:

2021-present	Associate Editor, Molecular Recognition, Frontiers in Molecular Biosciences
2019-present	NSF Panel Participant
2020-2022	Member, Medicinal Chemistry Faculty Search, UConn School of Pharmacy
2020-present	UConn MCB/STEM Workplace Navigator
2017-present	Member, Graduate School Hearing and Conflict Mediation Advisory Committees
2017-2018	Member, Biophysical Core Director Search Committee
2016-2017	Member, MCB Tenure Track Search Committee
2015-2021	Review Editor, Molecular Recognition, Frontiers in Molecular Biosciences
2015-2022	Member, Graduate Faculty Council
2012-present	Director, UConn Partnership for Excellence in Structure Biology
2011-2017	Co-head, Protein X-ray Crystallography Facility, University of Connecticut
2011-2016	Structural Biology, Biochemistry and Biophysics Graduate Program Chair
2010-2018	Member, Dept. Promotion, Tenure and Review Committee; Chaired, 2016-18, 2020-22
2010-present	Member, American Heart Assoc. Basic Cell - Protein & Crystallography Peer Review Committee
2010-2014	Ombudsperson, Department of Physics, University of Connecticut
2007-2016	NSF Peer Reviewer, ad hoc
2005-present	Member, Institute for Material Sciences, Univ. of Connecticut, Storrs, CT

C. Contributions to Science

1. The Role of Bacterial GTPases in Translation and Stress: My interest in bacterial GTPases began during my time at UMDNJ where I solved the first structure of a GTPase with multiple G-domains, a bacterial protein EngA (Der). This work continued at UConn where my group determined the ribosome binding properties of the translation factor BipA. We had the novel finding that BipA forms two distinct biologically relevant complexes with the ribosome, 70S:BipA and 30S:BipA. This is the only bacterial protein known to have two distinct ribosome binding modes. The formation of these species is dependent upon the guanine nucleotide pool in the cell, specifically GTP and ppGpp, an alarmone responsible for adaptation to altered growth conditions in bacterial cells. We have submitted an article describing the requirement of BipA for adaptation to and re-stablishing homeostasis after the onset of various stress (Bova, R.A., Benson, D.R. and Robinson, V.L. Physiological Role of BipA in *Escherichia coli* MG1655 During Stress Adaptation, under revision at *Appl. Environ. Microbiol.*). Our data suggests that BipA acts as an intermediary between the ribosome and the cellular environment and that its structural and regulatory properties influence how the ribosome responds to current cellular conditions. We have another under review utilized solution biophysics, mutagenesis and biochemistry to identify a specificity determinant for ppGpp binding to BipA. Our initial work stimulated numerous investigations into how ppGpp

might regulate the biochemical and biophysical properties of various GTPases to synchronize adaptive responses to translational events.

a. Robinson, V.L., Hwang, J., Fox., E., Inouye., M. and Stock, A.M. (2002) Domain arrangement of Der, A Switch Protein Containing Two GTPase Domains. *Structure* **10**:1649-58.
b. deLivron, M.A. and Robinson, V.L. (2008) *Salmonella enterica* Typhimurium BipA Exhibits Two Distinct Ribosome Binding Modes. *J. Bacteriol.* **190**:5944-52.
c. deLivron, M.A., Makanji, H.S., Lane, M.C. and Robinson, V.L. (2009) A Novel Domain In Translational GTPase BipA Mediates Interaction with the 70S Ribosome and Influences GTP Hydrolysis. *Biochemistry* **48**:10533-41.

2. Drug Design for DHFR and other antibiotics inhibit Ribosomal Processes. We have had a large role supporting structure-based design strategies with several collaborators at UConn as well as the development of antimicrobials drug development projects within my own group. We have been working with Dr. Dennis Wright's group to design new antifolate chemotype that is able to maintain potency against trimethoprim (TMP)-resistant dihydrofolate reductase (DHFR) enzymes. In addition, we have supported ongoing work from Dr. Mark Peczuh group (Chemistry, UConn) to develop novel classes of glycosylated small molecules to target the 70S ribosome in gram negative bacteria. And finally, we have been validating and screening small molecule libraries for their influence on the biochemical and cellular properties of BipA. Its involvement in rapidly shifting the cellular physiology of bacteria to promote an adaptive response by facilitating the formation of new translational complexes may make it a good antimicrobial target.

a. Krucinska, J., Lombardo, M.N., Erlandsen, H., Hazeen, A., Duay, S.S., Pattis, J.G., Robinson, V.L., May, E.R. and Wright, D.L. (2019) "Functional and structural basis of *E. coli* enolase inhibition by SF2312: a mimic of the carbanion intermediate." *Sci Rep.* 9(1):17106. PMCID: PMC6863902.
b. Mayo, C.B., Erlandsen, H., Mouser, D.J., Feinstein, A.G., Robinson, V.L., May, E.R. and Cole. J.L. (2019) "Structural Basis of Protein Kinase R Autophosphorylation." *Biochemistry*. 58(27):2967-77. PMCID: PMC6615999
c. Krucinska, J., Falcone. E, Erlandsen, H., Hazeen, A., Lombardo, M.N., Estrada, A., Robinson, V.L., Anderson, A.C. and Wright, D.L. (2019) Structural and Functional Studies of Bacterial Enolase, a Potential Target against Gram-Negative Pathogens. *Biochem.* **58**:1188-97. PMCID: PMC6511404.
d. Cannone, Z., Shaqra, A.M., Lorenc, C., Henowitz, L., Keshipeddy, S., Robinson, V.L., Zweifach, A., Wright, D. and Peczuh, M.W. (2019) Post-Glycosylation Diversification (PGD): An approach for Assembling Collections of Glycosylated Small Molecules. *ACS Comb. Sci.* 21:192-7. PMCID: 30607941

3. Understanding Nucleo-cytoplasmic Trafficking and IDPs: We have been addressing questions regarding the molecular mechanism of action of nucleostemin, a circularly permuted GTPase that plays an important role in maintaining nucleolar integrity. NS shuttles from the nucleolus to the nucleoplasm in response to cell cycle changes and stress conditions. This trafficking depends on interactions with multiple partners that regulate critical processes such as cellular proliferation, ribosome biogenesis and DNA maintenance thereby ensuring homeostasis in eukaryotic cells. All of these activities suggest that NS acts as a GTP dependent regulatory link between developmental controls and cell cycle machinery. We have applied bioinformatics tools together with numerous biophysical techniques to begin to understand the solution properties of *Drosophila melanogaster* nucleostemin 1 (NS). Our studies indicate that NS has large regions of disorder at the termini while the core cpGTPase domain is structured (Daman, T.H. *et al.* Intrinsic Structural Disorder Revealed in the Circularly Permuted GTPase Nucleostemin, under review*)*. Combining SAXS data and molecular dynamics flexible fitting, we provide the first structural model of this family of proteins. Future goals include how the permutation of the G-domain, leading to the repositioning of the switch I region, influences the enzymatic properties of the protein. Our experience with intrinsically disordered proteins has led to a collaboration with Dr. Brian Aneskievich (UConn School of Pharmacy) to understand how the disordered regions in tNIP, a protein involved in inflammatory pathways, provide a scaffold for its interactions with A20 and ubiquitin.

a. Rosby, R., Cui, Z., Rogers, E., deLivron, M.A., Robinson, V.L. and DiMario, P.J. (2009) Knockdown of the Drosophila GTPase nucleostemin 1 impairs large ribosomal subunit biogenesis, cell growth, and midgut precursor cell maintenance. *Mol. Biol. Cell*. **20**:4424-34.
b. Shamilov, R, Robinson, V.L. and Aneskievich, B.J. (2021) Seeing Kerotinocyte Proteins Through the Looking Glass of Intrinsic Disorder. *Int. J. Mol. Sci.*, **22**:7912.

4. **Two-Component Signaling Field:** My work contributed to the understanding that response regulator proteins utilized distinct signaling mechanisms. During my post-doctoral work, I crystallized the second full length

response regulator (RR) from the OmpR/PhoB family of proteins. Mutational study was then designed to examine how intermolecular contacts contributed to communication between the N- and C-terminal domains of these RRs. We also carried out a complete structural analysis addressing the dephosphorylation of CheY by its cognate phosphatase CheZ. Through multiple structures and complimentary biochemistry, we proved that CheZ associates with a “meta-active” state of CheY that is distinct from either the active or inactive form of the protein indicating a continuous flow of information between these two partners. At UConn, I collaborated with Drs. Andrei Alexandrescu and Dan Gage to solve the NMR structure of an unusual RR Sma1. This CheY like protein is missing the fourth α-helix of the consensus 455 face but instead has a dynamic loop suggesting that it has an altered recognition interface for binding to downstream effectors.

a. Stock, A.M., Robinson, V.L. and Goudreau, P.N. (2000) Two-component signal transduction. *Ann. Rev. Biochem*. **69**:183-215.

b. Robinson, V.L., Wu, T. and Stock, A.M. (2003) Structural Analysis of the interdomain interface of DrrB, An OmpR/PhoB family member from *Thermotoga maritima*. *J. Bacteriol*. **185**:4186-94.

c. Guhaniyogi, J., Robinson, V.L. and Stock, A.M. (2006) Crystal structures of beryllium fluoride-free and beryllium fluoride-bound CheY in complex with the conserved C-terminal peptide of CheZ reveal dual binding modes specific to CheY conformation. *J. Mol. Biol.* **359**:624-45.

d. Sheftic, S.R., Garcia, P.P., Robinson, V.L., Gage, D.J. and Alexandrescu, A.T. (2012) Nuclear magnetic resonance structure and dynamics of the response regulator Sma0114 from *Sinorhizobium meliloti. Biochem.* **51**:6932-41.

Complete List of Published Work in MyBibliography:

https://pubmed.ncbi.nlm.nih.gov/?term=robinson+vl+and+connecticut&sort=date

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: Tristan Evans

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

POSITION TITLE: Graduate Research Assistant/Teaching 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
University of Connecticut	BSc	9/1/2017	5/1/2021	Molecular and Cell Bio
University of Connecticut	PhD	9/1/2021	Ongoing	Structural Biochem

A. Personal Statement

I have always maintained a desire to gain and spread knowledge. As a child it began with lighthearted fun facts and as I matured it developed into a hunger to push the boundaries of science. This spark did not fully ignite until my freshman year at the University of Connecticut when I was introduced to epigenetics offhandedly by an Anthropology teaching assistant. My fascination with this topic motivated me to switch my major to Molecular and Cell Biology (MCB). The summer before my sophomore year, I joined Dr. Michael O'Neill's lab in the MCB department where my passion for research was fully ignited. In 2021, I continued at UConn as a doctoral student in the MCB Ph.D. program. My scientific interests pivoted again as I became a member of Dr. Simon White's group. Dr. White's utilizes biophysical methods and structural biology, in particular, cryoEM, to understand how RNA viruses assemble. He is also interested in the evolution, stability, and assembly of actinobacteriophage. I became involved in this bacteriophage research and solved over a dozen structures learning the nuances of cryoEM including grid preparation, data collection and processing, de novo model building and refinement. Earlier this year, Dr. White transitioned out of academia and I joined Dr. Victoria Robinson's group. I am still doing what fascinates me. Instead of working with bacteriophages I am now studying their prey, specifically *Escherichia coli*, and how they respond to environmental stress such as nutrient deprivation. Our lab does not solely focus on structural biology but delves deeply into the concentrations of microbiology and biochemistry. My project is to determine a structural description of how a translational GTPase BipA forms a complex with the 30S ribosome using cryoEM.

B. Positions, Scientific Appointments and Honors

2023 – Present Graduate Research Assistant, Robinson Lab, MCB Department, University of Connecticut
2021 – 2023 Graduate Research Assistant, White Lab, MCB Department, University of Connecticut
2020 – 2021 Institute for Brain and Cognitive Sciences Grant Recipient
2018 – 2021 Undergraduate Research Assistant, M. O'Neill Lab, MCB Department, University of Connecticut

C. Contributions to Science

Undergraduate Research: From the summer of 2018 to the summer of 2021, I was part of Dr. Michael O'Neill's lab at the University of Connecticut. Dr. O'Neill's work focused on the epigenetics of *Mus musculus*. Specifically, a novel imprinted gene on the mouse X chromosome with links to autism. During my time in the lab, I identified potential protein interactions between the X chromosome gene and other proteins involved in the process known as Meiotic Sex Chromosome Inactivation (MSCI). Using gene knockdown mice, I also observed a sublethal event leading to a bias in sex of offspring, relating to the phenomenon of transgenerational epigenetic inheritance.

1. Natali Sobel Naveh, Robert J. Foley, Katelyn R. DeNegre, **Tristan C. Evans**, Anne Czechanski, Laura G. Reinholdt, Michael J. O'Neill. Deficiency of SYCP3-related XLR3 disrupts the initiation of meiotic sex chromosome inactivation in mouse spermatogenesis. **doi:** https://doi.org/10.1101/2021.03.30.437712

Graduate Research: Starting in 2021 I joined the UConn MCB graduate school and decided to switch focus to structural protein work. I was intrigued by the work of Simon White, combining cryoEM and virology to better understand bacteriophages. Before he left the university, I modeled dozens of phage proteins. We are currently finishing a paper that combines alphafold predictions and validated EM structures of the bacteriophage major tail protein. Through this project I have 10 PDB validated structures ready to be deposited in tandem.

After Simon's departure I transferred to the lab of Victoria Robinson, someone who I had worked closely with during my time in the White lab. My current project involves examining complex formation between the ribosome and several proteins required for the bacterial stress response, in particular those involved in protein production. The goal is to merge my acquired cryoEM knowledge with ongoing biochemistry and microbiology in the group to produce new insights into the bacterial stress response.

D. Scholastic Performance

YEAR	COURSE TITLE	GRADE
2021	Research in Progress	S
2021	Lab Rotations	S
2021	Intro to Research	A
2021	Faculty Research	S
2022	Virology	A
2022	Scientific Writing	A
2022	Responsible Conduct Research	S
2022	Foundations of Structural Biochem	A
2022	Structure and Dynamics of Macromolecular Machines	A
2022	Research in Progress	S
2022	Invited Seminar	S
2022	Research	A
2023	Techniques of Biophysical Chemistry	B
2023	Structure/Function of Biological Macromolecules	B