BIOGRAPHICAL SKETCH

NAME: Mugridge, Jeffrey S

POSITION TITLE: Assistant Professor of Chemistry & Biochemistry

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

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 Chicago	B.S.	06/2006	Chemistry
University of California, Berkeley	Ph.D.	12/2010	Chemistry
University of California, San Francisco	Postdoctoral	07/2017	Biochemistry & Structural Biology

A. Personal Statement

I began my independent career as an Assistant Professor at the University of Delaware in August 2019. My laboratory is focused on understanding the chemical, structural, and molecular mechanisms that control RNA-modifying enzymes and their impacts on cellular gene expression and human disease. We combine structural biology with enzymology and biophysical assays to uncover how protein-protein and protein-nucleic acid interactions regulate mRNA and tRNA modification activity and selectivity to help control the epitranscriptome. Current projects in our lab are funded by an NIH R35 MIRA and NSF CAREER award.

Our lab has extensive experience in protein crystallography – as a postdoc at UCSF, I determined multiple structures of the eukaryotic mRNA decapping complex (PDB 5KQ1, 5KQ4, 6AM0) and our lab at UD has determined structures of a number of RNA binding/modifying proteins (PDB 9DW6, 9NCZ, 9N80). However, we now have a number of large protein-RNA complexes that are strong candidates for structural analysis by cryo-EM; we are applying to the NCCAT GUP3 program to jump start cryo-EM studies in our lab, which will significantly expand our structural biology skillset and allow us to analyze large protein-RNA complexes.

B. Positions and Honors

Positions and Employment

Aug 2019 –	Assistant Professor, Department of Chemistry & Biochemistry, University of Delaware, Newark, DE.
2017 – 2019	Associate Specialist, JD Gross Lab, Department of Pharmaceutical Chemistry, University of California, San Francisco, San Francisco CA.
2011 – 2017	Postdoctoral Fellow, JD Gross Lab, Department of Pharmaceutical Chemistry, University of California, San Francisco, San Francisco CA.
2008 – 2011	Graduate Staff Scientist, NMR Facility, Department of Chemistry, University of California, Berkeley, Berkeley CA.
2006 – 2010	Graduate Research Fellow, KN Raymond Lab, Department of Chemistry, University of California, Berkeley, Berkeley CA.
2004 – 2006	Undergraduate Researcher, MD Hopkins Lab, Department of Chemistry, University of Chicago, Chicago IL.

Other Experience and Professional Membership

2005 - Member, American Chemical Society
2014 - Member, Biophysical Society
2018 - Member, The RNA Society

Honors

2003-6 University of Chicago Dean's List
2004-5 Richter Grant Recipient, University of Chicago
2009 Bavaria-California Technology Center Grant Recipient
2007-10 National Science Foundation Graduate Student Fellowship
2013-15 NIH Ruth L. Kirschstein NRSA Postdoctoral Fellowship (F32)

C. Contributions to Science

Complete List of Published Works in MyBibliography:
https://www.ncbi.nlm.nih.gov/myncbi/16w2JLjfasDYxf/bibliography/public/

1. The tRNA methyltransferase TRMT1 is recognized and cleaved by the SARS-CoV-2 main protease (Mpro) to regulate tRNA methylation during viral infection. TRMT1 is a methyltransferase that installs the N2,2-dimethyladenosine (m2,2G) modification on human tRNAs. m2,2G is important for tRNA structure, translation, cellular response to redox stress, and has been linked to human intellectual disability. Our lab has combined structural biology, RNA biochemistry, and enzymology to show how the SARS-CoV-2 main protease (Mpro) recognizes a conserved sequence in TRMT1 and catalyzes its cleavage. We found that cleavage of TRMT1 results in decreased tRNA binding and complete inactivation of its methyltransferase activity. This work reveals the structural basis for the TRMT1-Mpro interaction and suggests that SARS-CoV-2 may target RNA modification enzymes to impact translation or other viral phenotypes.

a. D'Oliviera A, Dai X, Mottaghinia S, Olson S, Geissler EP, Etienne L, Zhang Y, **Mugridge JS*** (2025). Recognition and Cleavage of Human tRNA Methyltransferase TRMT1 by the SARS-CoV-2 Main Protease. *eLife*, https://elifesciences.org/articles/91168, PMID: 39773525.

2. Conformational changes in the multi-protein mRNA decapping complex control 5' mRNA cap recognition and activation of decapping to regulate mRNA stability. The coordinated destruction of mRNA is critical for transcript quality control and the post-transcriptional regulation of gene expression. The conserved decapping enzyme Dcp2 plays an important role during mRNA decay by catalyzing cleavage of the 5' mRNA cap, resulting in translational repression and rapid degradation of the mRNA transcript. However, a structure-level, mechanistic understanding of Dcp2 activity and regulation was entirely absent because the decapping enzyme is very challenging to study *in vitro*; it undergoes large conformational changes, has weak affinity for its 5' cap substrate, and operates as part of a large, multi-protein decapping complex in the cell. My primary postdoctoral work at UCSF used macromolecular crystallography to trap and structurally characterize different conformations of the mRNA decapping enzyme Dcp2 in complex with decapping cofactors and a synthetic, tight-binding, 5' cap analog. These crystal structures, together with detailed kinetic analyses, revealed: (1) a large conformational change from an inactive to active conformation generates the Dcp2 active site to recognize and cleave the 5' mRNA cap, (2) decapping cofactors (Edc1 and Edc1-like proteins) stabilize the active conformation of Dcp2 to accelerate catalysis, and (3) multiple cofactors (Edc1 and Edc3) can engage the decapping complex simultaneously to synergistically promote decapping. These studies provide a comprehensive, high-resolution description of Dcp2 function and activation, laying the necessary foundation for future studies to dissect and understand how decapping activity is coupled to diverse

cellular processes where roles for Dcp2 have been identified, including transcription termination, ribosome biogenesis, mRNA translation, and 5'-3' mRNA decay.

a. **Mugridge JS**, Ziemniak M, Jemielity J, Gross JD (2016). Structural basis of mRNA-cap recognition by Dcp1–Dcp2. *Nat. Struct. Mol. Biol.*, 23, 987 – 994. PMCID: PMC5113729
b. **Mugridge JS**, Ziemniak M, Jemielity J, Gross JD (2018). Structure of the activated Edc1-Dcp1-Dcp2-Edc3 mRNA decapping complex with substrate analog poised for catalysis. *Nat. Commun.*, 9, 1152. PMCID: PMC5861098
c. **Mugridge JS**, Gross JD (2018). PDB ID: 6AM0. Crystal structure of K. lactis Edc1-Dcp1-Dcp2-Edc3 decapping complex with synthetic cap substrate analog. DOI: 10.2210/pdb6AM0/pdb
d. **Mugridge JS**, Coller J, Gross JD (2018). Structural and molecular mechanisms for the control of eukaryotic 5'-3' mRNA decay. *Nat. Struct. Mol. Biol.*, 25, 1077 – 1085. PMID: 30518847

3. New molecular tools for mRNA 5' decapping. All eukaryotic mRNAs carry a 5' cap structure that is critical for proper transcript splicing, export, translation, and stability. The 5' cap consists of an N7-methylguanosine nucleotide connected to the 5' end of mRNA by a triphosphate bridge. The cap is installed co-transcriptionally, recognized during a key step in translation initiation, and is removed by the decapping enzyme Dcp2 to commit the transcript to rapid degradation. However, owing to the chemical complexity of the cap structure and its weak affinity to Dcp2, synthetic 5' cap analogs that inhibit or tightly bind the decapping enzyme were not available. Furthermore, a recent commercial shortage of tobacco acid pyrophosphatase (TAP), the molecular reagent of choice for removing the 5' cap during RNA sequencing library preparation and other RNA biochemistry applications, created a need for new reagents that rapidly cleave the 5' cap. With these motivations, I (1) participated in a project which developed, characterized, and validated novel 5' cap analogs that potently inhibit mRNA decapping by Dcp2, which are useful as new synthetic tools to study 5' mRNA decapping; and (2) designed and functionally tested complexes of Dcp2 with its partner Dcp1 that were covalently fused to decapping cofactors to generate stable protein constructs with exceptionally high decapping activity. We showed that these 'super decapping enzymes' are robust molecular biology reagents able to rapidly and efficiently remove the 5' mRNA cap during RNA biochemical applications like RNA-seq. These molecular tools have been shared widely to help facilitate the study of 5' cap-binding proteins and for molecular biology experiments requiring removal of the 5' cap.

a. Ziemniak M, **Mugridge JS**, Kowalska J, Rhoads RE, Gross JD, Jemielity J (2016). Two-headed tetraphosphate cap analogs are inhibitors of the Dcp1/2 RNA decapping complex. *RNA*, 22, 518 – 529. PMCID: PMC4793208
b. **Mugridge JS**, Ziemniak M, Jemielity J, Gross JD (2016). Cap recognition and catalytic activation of the Dcp1/Dcp2 mRNA decapping complex. Talk presented at: FASEB Conference on Post-transcriptional Control of Gene Expression, Lisbon, Portugal.
c. Paquette DR, **Mugridge JS**, Weinberg DE, Gross JD (2018). Application of a *Schizosaccharomyces pombe* Edc1-fused Dcp1-Dcp2 decapping enzyme for transcription start site mapping. *RNA*, 24, 251–257. PMCID: PMC5769751

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: Youmna Moawad

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

POSITION TITLE: Graduate Research Assistant/Doctoral candidate

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
Bridgewater College, Bridgewater VA	B.S.	12/2021	Biochemistry
University of Delaware, Newark DE	Ph.D.	Ongoing	Biochemistry

A. Personal Statement

I am pursuing a doctoral degree in Biochemistry at the University of Delaware in the Mugridge Lab conducting structural biochemistry research on RNA-modifying enzymes. I have a strong background in molecular biology and biochemistry, along with proficiency in protein expression and purification and structural biology techniques such as X-ray crystallography and small-angle X-ray scattering. In addition, I am well-versed in using structural prediction and modelling tools, such as AlphaFold and PyMol. In preparation for this project, I have been working with EM staff scientists at UD to develop preliminary technical skills in negative staining and cryo-EM sample preparation and data collection. Therefore, I have the necessary knowledge and skills to successfully execute the goals of this project.

B. Positions, Scientific Appointments, and Honors

Honors
2020 The Martin Summer Science Research Institute Scholarship.
2019 Dr. Stuart R. Suter Endowed Scholarship

C. Contributions to Science

1. Structural mechanisms controlling tRNA modification. I have contributed to a project in the Mugridge lab aimed at understanding how the tRNA modifying enzyme Elp3 catalyzes carboxymethylation of tRNAs across all domains of life. By combining structural analysis and enzymology, our lab's work on this enzyme has revealed a new mechanism for Elp3-mediated tRNA modification in which a molecular tunnel transports a key cofactor between distant active sites in Elp3.

a. Geissler EP, Moawad Y, Roehling PN, Martin K, Asare-Okai P, Mugridge JS. Elp3 uses a conserved molecular tunnel to transport acetate between distant active sites and catalyze tRNA carboxymethylation. *Submitted*.