BIOGRAPHICAL SKETCH

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NAME: Lieberman, Raquel Limor

eRA COMMONS USER NAME (credential, e.g., agency login): RL1236

POSITION TITLE: Professor of Chemistry & Biochemistry

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
Massachusetts Institute of Technology (MIT) Cambridge, MA	B. Sc.	1998	Chemistry
Northwestern University (NU) Evanston, IL	M.S.	1999	Chemistry
Northwestern University Evanston, IL	Ph.D.	2005	Structural Biology, Membrane protein biophysical chemistry (PhD advisor: Amy C. Rosenzweig)
Center for Neurologic Diseases, Harvard Medical School/Brigham & Women's Hospital (Boston, MA)/Brandeis University (Waltham, MA)	Postdoc	12/2007	Protein misfolding, pharmacological chaperones (Postdoctoral advisors: Gregory A. Petsko, Dagmar Ringe, Michael S. Wolfe)

A. Personal Statement

I am the Specic-Pfeil Professor of Chemistry & Biochemistry; my research program focuses on biophysical and structural characterization of proteins assicuated wutg misfolding. I have a broad background in chemistry, biochemistry, biophysics/structural biology. A major research project in my lab since its inception has been investigations of the glaucoma-associated myocilin protein, which has been funded by R01EY021205 since March 2011 and will continue through at least Feb 2026. The lab has made major strides toward detailed molecular understanding of myocilin structure, function, and disease pathogenesis. We have divulged similarities between myocilin-associated glaucoma and other protein misfolding disorders, particularly amyloid diseases. Cumulatively, our work is leading to the first disease-modifying glaucoma therapeutic. Other projects involve basic investigations of an intramembrane protease of the type linked to Alzheimer disease (NSF funded) and an enzyme linked to both Gaucher disease and Parkinson (MJFF funded). Rooted in basic research, the long-term goal of my research program is to convert mechanistic discoveries into disease-modifying therapies.

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

R01 EY021205
Lieberman, RL (PI)
3/2011-2/2026
Characterization of purified myocilin: glaucoma as a protein misfolding disease.

NSF MCB-1817796
Lieberman, RL (PI)

7/2018-NCE
Decoding intramembrane aspartyl protease substrate preferences and activity

Michael J. Fox Foundation
Lieberman, RL (PI)
3/2020-2/2023
Structures of Parkinson-relevant states of acid-β-glucosidase

Citations:

1. Hill, S. E., Donegan, R. K., **Lieberman, R. L.** The glaucoma-associated olfactomedin domain of myocilin forms polymorphic fibrils that are constrained by partial unfolding and peptide sequence. *J. Mol. Biol.***, 426(4)**, 921-35, 2014. PMCID: PMC3946817
2. Donegan, R. K., Hill, S. E., Freeman, D. M., Orwig, S. D., Turnage, K. C., and **Lieberman, R. L.** Structural basis for misfolding in glaucoma-associated myocilin. *Hum. Mol. Genet.* **24**, 2111-2124, 2015. PMCID: PMC4380063, cover article.
3. Hill, S. E., Nguyen, E., Donegan, R. K., Patterson-Orazem, A. C., Hazel, A., Gumbart, J. C., **Lieberman, R.L.** Structure and misfolding of the flexible tripartite coiled-coil domain of glaucoma-associated myocilin. Structure, **25**, 1697-1701, 2017, PMCID: PMC5685557, cover article.
4. Hill, S. E., Kwon, M. S., Martin, M. D., Suntharalingam, A., Hazel, A., Dickey, C. A., Gumbart, J. C., **Lieberman, R.L.**, Stable calcium-free myocilin olfactomedin domain variants reveal challenges in differentiating between benign and glaucoma-causing mutations. J. Biol. Chem. 294(34):12717-12728, 2019. PMCID: PMC6709634.

B. Positions, Scientific Appointments, and Honors

Positions and Scientific Appointments

2018-Present	Professor, School of Chemistry & Biochemistry, Georgia Institute of Technology, Atlanta, GA
2017-Present	Academic Editor, PLoS Biology
2014-2020	Biochemistry Division Chair, School of Chemistry & Biochemistry, Georgia Institute of Technology, Atlanta, GA (not 2018)
2013-2018	Associate Professor, School of Chemistry & Biochemistry, Georgia Institute of Technology, Atlanta, GA
2010- Present	Member, Center for Nanobiology of the Macromolecular Assembly Disorders
2010- Present	Affiliate Member, Integrative BioSystems Institute
2008-2013	Assistant Professor, School of Chemistry and Biochemistry, Georgia Institute of Technology, Atlanta, GA
2008- Present	Member, Institute for Bioengineering and Bioscience

Honors

2020 Gretzinger Moving Forward Award (College of Sciences)
2017 Sigma Xi Best Faculty Paper Award (Georgia Tech)
2014 Cullen Peck Faculty Fellow (Georgia Tech)
2013 Junior Faculty Outstanding Undergraduate Research Mentor Award (Georgia Tech)
2012 Sigma Xi Young Faculty Award (Georgia Tech)
Paul A. Duke GIFT Action Plan Achievement Mentor Award For K-12 outreach (with Mr. Casey Bethel, teacher)**,** first place
2010 Blanchard Assistant Professor (Georgia Tech)
Pew Scholar in Biomedical Sciences
NSF CAREER award
2009 American Federation for Aging Research Rosalind and Arthur Gilbert New Investigator Award
2008 Glaucoma Research Foundation (GRF) Shaffer Fund awardee
2006 American Chemical Society Nobel Laureate Signature Award for Graduate Education in Chemistry
2005 Ruth L. Kirschstein NIH Postdoctoral Research Fellowship
1998 Phi Beta Kappa, MIT

Merck Index Award for Excellence in Chemistry, MIT

C. Contributions to Science

1. My graduate work focused on structural and biophysical characterization of metalloenzymes and integral membrane proteins. In particular, I solved the crystal structures of a novel (soluble) red copper protein as well as of particulate methane monooxygenase (pMMO), a 300 kDa heterotrimeric copper-containing integral membrane protein. As part of my thesis, I characterized the pMMO metal centers by a variety of spectroscopic techniques. I continue to be interested in metalloenzymes that perform unusual chemistry.
    a) **Lieberman, R. L.**; Shrestha, D. B.; Doan, P. E.; Hoffman, B. M.; Stemmler, T. L.; Rosenzweig, A. C. Purified particulate methane monooxygenase from *Methylococcus capsulatus* (Bath) is a dimer with both mononuclear copper and a copper-containing cluster. *Proc. Natl. Acad. Sci. USA*, **100**, 3820-3825, 2003. PMCID: PMC153005
    b) **Lieberman, R. L.**; Rosenzweig, A. C. Crystal structure of a membrane-bound metalloenzyme that catalyses the biological oxidation of methane. *Nature*, **434**, 177-182, 2005. PMCID: N/A
    c) Kalyoncu S., Heaner D. P. Jr., Kurt Z., Bethel C. M., Ukachukwu C. U., Chakravarthy S., Spain J. C., **Lieberman, R. L.** Enzymatic hydrolysis by transition-metal-dependent nucleophilic aromatic substitution *Nat. Chem. Biol.*, 12(12), 1031-1036, 2016. PMCID: PMC5110390
    d) Huard, D. J. E., Demissie, A., Kim, D., Lewis, D., Dickson, R. M., Petty, J. T., **Lieberman, R. L.** Atomic structure of a fluorescent Ag_8 cluster templated by a multi-stranded DNA scaffold, J. Am. Chem. Soc., **141**(29), 11465-11470, 2019. PMCID: PMC6606393

2. My postdoctoral work shifted towards biophysical and structural characterization of proteins prone to misfolding and implicated in neurodegenerative disorders, particularly Gaucher, Fabry, and Alzheimer disease. I solved crystal structures of the defective enzyme in Gaucher and Fabry diseases in complex with small molecule pharmacological chaperones (PCs) in clinical trials; the one for Fabry was recently FDA approved. I also made strides in molecular comprehension of the integral membrane enzyme signal peptide peptidase (SPP), a relative of γ-secretase implicated in Alzheimer disease. In my independent career, I have continued to work on aspects of these projects. The SPP project is one major subgroup of my independent laboratory (see below), and I have contributed to studies of next generation Gaucher and Fabry PCs.
    a) **Lieberman, R. L.** Wustman, B. A.; Huertas, P.; Powe, A. C., Jr.; Pine, C. W.; Khana, R.; Schlossmacher, M. G.; Ringe, D.; Petsko, G. A. Structure of acid-β-glucosidase with pharmacological chaperone provides insight into Gaucher disease. *Nat. Chem. Biol.*, **3,** 101-107, 2007 PMCID: N/A
    b) Landon M. R., **Lieberman R. L.**, Hoang Q. Q., Orwig, S. D., Kosakov D., Ju S., Brenke R., Chuang G. Y., Vajda S., Petsko G. A., and Ringe D. Detection of ligand binding hot spots on protein surfaces using fragment-based methods: application to DJ-1 and glucocerebrosidase. *J. Comp. Aid. Des.,* 23,491-500, 2009. PMCID: PMC2889209
    c) Orwig, S. D., Tan, Y. L., Grimster, N. P., Yu, Z., Powers, E., Kelly, J. W., **Lieberman, R. L.** Binding of 3,4,5,6-tetrahydroxyazepanes to the acid β glucosidase active site: Implications for pharmacological chaperone design for Gaucher disease, *Biochemistry*, 50(49), 10647-10657, 2011 PMCID: PMC2699628
    d) Yu Y., Mena-Barragán T., Higaki K., Johnson J. L., Drury J. E, **Lieberman R. L.**, Nakasone N., Ninomiya H., Tsukimura T., Sakuraba H., Suzuki Y., Nanba E., Mellet C. O., García Fernández J. M., Ohno K. Molecular basis of 1-deoxygalactonojirimycin arylthiourea binding to human α-galactosidase A: Pharmacological chaperoning efficacy on Fabry disease mutants. *ACS Chem. Biol.* 9(7), 1460-9, 2014. PMCID: N/A

3. A major interest of mine continues to be the high-risk high-payoff study of membrane enzymes, particularly SPP (see section 2) and methods to improve the likelihood of obtaining diffraction quality crystals for structure determination. The lab has developed new assays to measure SPP proteolysis as well as peptide-specific antibody fragments to promote membrane protein crystallization. We also use neutron scattering to probe structure and lipids in solution. This work is currently funded by NSF (MCB-1817796), previously by NSF (MCB-0845445) and NIH (R01 GM095638 and R21 DK091357).
    a) Johnson, J. L., Entzminger, K., Hyun, J., Kalyoncu, S., Heaner, D., Morales, I., Sheppard, A., Gumbart, J.C., Maynard, J. A., **Lieberman, R. L.** Structural and biophysical characterization of

epitope-specific engineered Fab fragment and complexation with membrane proteins: implications for co-crystallization. *Acta Crystallographica* D71(pt. 4) 896-906, 2015. PMCID: PMC4388267

b) Naing, S.-H., Vukoti, K. M., Drury, J. E., Johnson, J. L., Kalyoncu, S., Hill, S. E., Torres, M. P., **Lieberman, R. L.** Catalytic properties of intramembrane aspartyl protease substrate hydrolysis evaluated using a FRET peptide cleavage assay. *ACS Chem. Biol.* 10(9), 2166-74, 2015. PMCID: N/A

c) Naing, S.H., Kalyoncu, S., Smalley, D. M., Tao, X., George, J. B., Jonke, A., Kim, H., Oliver, R.C., Urban, V.S., Torres, M. P., **Lieberman, R. L.**@ Both positional and chemical variables control in vitro proteolytic cleavage of a presenilin ortholog. J. Biol. Chem., 293(13), 4653–4663, Editor's Choice, cover article. PMCID: PMC5880133.

d) Naing, S.H., Oliver, R.C., Weiss, K.L., Urban, V.S., **Lieberman, R. L.** Solution structure of an intramembrane aspartyl protease via small angle neutron scattering. Biophys. J., 114(3), 602-608, 2018. PMCID: PMC5985038.

4. A second major current interest continues to be in protein conformational disorders. Branching out from Gaucher and Fabry, in my independent career I have focused on studying myocilin-associated glaucoma through the vantage point of protein misfolding. In particular we have focused on the mutation-prone olfactomedin domain, trying to understand its structure, function, and dysfunction. The latter goal has been particularly productive as we have been able to characterize residual stability, propensity to form amyloid, and identify several new categories of small molecules to ameliorate the misfolding phenotype. Our molecular perspective is unique among glaucoma researchers. We have a new grant from the Fox Foundation to continue this line of research as it relates to Parkinson disease.

   a) Orwig, S. D., Perry, C. W., Kim, L. Y, Turnage, K. C., Zhang, R., Vollrath, D., Schmidt-Krey, I., **Lieberman, R. L.**, Amyloid fibril formation by the glaucoma-associated olfactomedin domain of myocilin. *J. Mol. Biol.*, 421, 242-255, 2012. PMCID: PMC3323732.

   b) Hill SE, Kwon MS, Martin MD, Suntharalingam A, Hazel A, Dickey CA, Gumbart JC, Lieberman RL. Stable calcium-free myocilin olfactomedin domain variants reveal challenges in differentiating between benign and glaucoma-causing mutations. J. Biol. Chem. 294(34):12717-12728, 2019. PMCID: PMC6709634.

   c) Huard, D.J.E., Jonke, A.P., Torres, M.P., Different Grp94 components interact transiently with the myocilin olfactomedin domain in vitro to enhance or retard its aggregation. Sci. Rep. 9(1):12769, 2019. PMCID: PMC6726633.

   d) Wang, Y., Gao, Y., Hill, S. E., Huard, D. J. E., Tomlin, M. O., **Lieberman, R. L.**, Paravastu, A. K., Hall, C.K. Simulations and experiments delineate amyloid fibrilization by peptides derived from glaucoma-associated myocilin. J. Phys. Chem. B., 122(2), 5845-5850, 2018. PMCID: PMC6186006.

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

BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors.
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NAME: Emily Saccuzzo

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

POSITION TITLE: Graduate Research Fellow, School of Chemistry and Biochemistry

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, Storrs, CT	B.S.	08/2014	05/2018	Chemistry
Georgia Institute of Technology, Atlanta, GA	PhD	08/2018	12/2023 (Expected)	Biophysical Chemistry

A. Personal Statement

My formal scientific research training began at The University of Connecticut, when I joined a research lab in my second year. Through my experience in Dr. Rouge's lab, I gained both biochemistry and organic chemistry knowledge as well as the opportunity to pioneer my own project and become an independent researcher. This experience motivated me to apply for a Summer Undergraduate Research Fellowship (SURF) for the summer of 2017, which I was awarded. Through my undergraduate research at UConn I gained both a solid biochemistry and chemistry foundation as well as experience working independently on my own project, where I discovered my passion for bench research.

I chose to pursue a PhD in the School of Chemistry and Biochemistry at Georgia Institute of Technology due to the outstanding reputation of Georgia Tech as well as the breadth of opportunities a strong program like this would offer, including the state-of-the art facilities I would have access to. My PhD thesis project centers around characterization of glaucoma-associated myocilin, specifically the misfolding of the C-terminal olfactomedin (mOLF) domain. By training, Dr. Lieberman is an X-ray crystallographer and used this technique to solve the structure of wild type (WT) mOLF. However, glaucoma-causing mOLF mutations are destabilizing and therefore preclude these constructs from being compatible with crystallization. My project involves solution NMR spectroscopy to study the effect of destabilizing mutants on tertiary structure as well as solid state NMR and electron microscopy (EM) to characterize the structures of aggregates formed by these destabilized constructs. We will gain insight into non-native structures adopted by destabilized WT mOLF and variants as well as characterize early aggregation events.

My project(s) will be supervised by Dr. Raquel L. Lieberman who has served on the faculty at Georgia Tech for over a dozen years. Dr. Lieberman is an experienced investigator, whose lab has a subgroup dedicated to studying myocilin. Dr. Lieberman has been the primary PI on numerous NIH funded grants, including a second renewal R01 award. Her work has helped establish a strong infrastructure for biophysical chemistry as Georgia Tech as well as pioneer the way for strong women in STEM. Her vast knowledge in several fields including biochemistry as well as protein science will be a key resource for the successful completion of my project.

Future Career Aspirations: Throughout my thesis work I have been fortunate enough to have access to some of the most cutting-edge biophysical characterization methods, such as cryogenic electron microscopy (cryoEM) and nuclear magnetic resonance (NMR) experiments designed for large proteins. Exposure to these techniques fueled my passion for structural biology and inspired me to pursue a career centered around utilizing

these technologies, ideally working in a core centered around structural biology and protein characterization. When I joined the Lieberman lab mine was the first set of projects centered around NMR spectroscopy. It was our relationship with the Van Horn Lab, experts in solution NMR, that have helped to ensure the success of these projects. The personnel of the Lieberman lab is extremely supportive and engaging, but most of my lab mates had expertise centered around characterization with X-ray crystallography and no one had extensive experience with NMR. From the experience of pioneering a technique outside the wheelhouse of my own lab I learned to really value the assistance of experts within this technique, as being a beginner to such a complicated method was extremely intimidating. I would love to take advantage of my own developed skillset, such as with cryoEM grid preparation or NMR resonance assignments, to help other scientists succeed in their own research endeavors centered around protein characterization. A career at a cryoEM or NMR core facility would afford me many opportunities to do so.

B. Positions and Honors

Positions

2021-present	GAANN Fellow, Georgia Institute of Technology
2019-2021	Vision Research Training Fellow, Emory University
2018-present	Graduate Research Assistant, Georgia Institute of Technology, Department of Chemistry and Biochemistry
2018-2019	Graduate Teaching Assistant, Georgia Institute of Technology, Department of Chemistry and Biochemistry
2016-2018	Peer Research Ambassador, University of Connecticut
2015-2018	Undergraduate Researcher, University of Connecticut

Scientific and Academic Honors

2021	Molecular BioMedical Seminar Series Second Place Talk, Georgia Institute of Technology
2018-present	Presidential Fellow, Georgia Institute of Technology
2019-2021	Vision research training grant, Emory University
2019	UAB-SERCAT Structural Biology Symposium Travel Grant
2014, 2018	Deans List, University of Connecticut
2017	Summer Undergraduate Research Fellowship recipient, University of Connecticut
2013-2014	National Honors Society

C. Contributions to Science

My contributions to science began with an undergraduate research career and included both biochemistry and bio-organic work. Most of this work focused on the targeted treating of retinal ganglion cell axon loss, which inspired me to pursue graduate studies focused on ocular diseases, as well.

1. As an undergraduate student, I worked for 3 years in the multidisciplinary laboratory of Dr. Jessica Rouge at the University of Connecticut. Dr. Rouge's lab worked at the interface of materials science, biochemistry and nanotechnology to develop adaptable systems for targeted drug delivery. The project I worked on involved functionalizing a micelle, capable of encapsulating a hydrophobic therapeutic molecule, with both targeting and therapeutic oligonucleotides. My specific contribution involved optimizing the generation of the C3 aptamer, an RNA sequence that binds to a receptor on Retinal Ganglion Cells (RGCs) with high affinity. Functionalization of this aptamer onto the surface of our nanoparticle constructs should result in the specific delivery of this cargo to RGCs. I presented these studies in an oral talk to the Chemistry Department at UConn as well as at 3 different poster presentations, one of which is highlighted below.
    a. **Saccuzzo, E.**, Gudipati, S., Rouge, J. Enzymatic ligation of RNA aptamers to nanoparticle surfaces. ACS Northeast Regional Undergraduate Research Symposium. 2018; Storrs, CT.
2. In the summer of 2018, I joined the Lieberman lab for a 12-week project that involved optimizing the yield of mOLF in minimal media. NMR is a valuable biophysical characterization method, but it requires the incorporation of isotopic labels, most commonly ^{13}C and ^{15}N for biological applications. Due to the notorious insensitivity of NMR, it was estimated that we would need upwards of ~6 mg of mOLF protein to achieve a detectable signal. The previous minimal media growth protocol adapted in the Lieberman lab for mOLF involved growing the cells in nutrient rich media, pelleting the cells and resuspending in minimal media for protein induction, but was never optimized, and would have required ~$10,000 in labeled reagents to generate a uniformly labeled ^{13}C, ^{15}N, ^{1}H-mOLF sample for NMR. I searched the literature and tested different

methods, including increased induction times, adding vitamin and trace element solutions, increasing aeration, growing to higher density in nutrient rich media before pelleting and resuspending in minimal media, etc. This project resulted in an optimized minimal media growth protocol that brought the cost of producing a uniformly labeled mOLF sample down to ~$2,500, a much more economically feasible value. I presented this project in the form of a poster, listed below.

   a. **Saccuzzo, E.,** Hill, S., Gao, Y., Paravastu, A., Lieberman, R. L. Expression of Glaucoma-Associated Myocilin Olfactomedin (OLF) Domain with ^{13}C- and ^{15}N- Labels for NMR Structural Experiments. Georgia Institute of Technology Department of Chemistry and Biochemistry Annual Retreat. 2018 October; Atlanta, GA.

3. My current graduate thesis project has involved utilizing my optimized minimal media growth protocol for both solid state and solution NMR applications. The main goal of my solid-state NMR (ssNMR) project is to elucidate the amino acid residues compromising the fibril core of mOLF amyloid fibrils. I produced a ^{13}C, ^{15}N, ^{1}H-uniformly labeled mOLF amyloid fibril sample and acquired preliminary 2D ssNMR spectra, with the help of the Paravastu lab at Georgia Tech, revealing that out of the 277 residues of mOLF, only <30 are responsible for forming the fibril core. 3D HNCACX and HNCOCX spectra were then acquired on this sample to reveal a tripeptide stretch in the fibril core, which is only present once within the mOLF sequence. Additional assignments were not possible due to low spectral resolution caused by sample heterogeneity. This work has also allowed me the opportunity to adventure into the exciting new world of cryoEM, as this technique can be more forgiving with heterogenous samples. I have begun this part of the project by collecting preliminary dark field transmission electron microscopy (TEM) images of seeded fibril samples and made attempts to optimize the fibril-to-grid ratio before proceeding forward with cryoEM. Throughout this optimization process, I developed an aggregation protocol that produces small oligomeric particles with a pore in the center as observed with EM grids. We are interested in characterizing these oligomers further with cryoEM, as they are likely the building block species in amyloid formation. These works have resulted in my first co-author publication and a poster presentation, listed below.

   a. **Saccuzzo, E**., Martin, M., Gao, Y., Kim, M., Paravastu, A. K., Lieberman, R. L. Molecular characterization of glaucoma-causing mocilin variants and corresponding aggregates. Georgia Institute of Technology Department of Chemistry and Biochemistry Annual Retreat. 2019, October; Atlanta, GA.
   b. Gao, Y., **Saccuzzo, E.,** et al. (2021). Structural Arrangement within a Peptide Fibril Derived from the Glaucoma-Associated Myocilin Olfactomedin Domain. *The Journal of Physical Chemistry B*, 125(11), 2886-2897.
   c. **Saccuzzo, E.**, Gao, Y., Mebrat, M., Paravastu, A., Van Horn, W., Lieberman. R. L., Structural Characterization of the Glaucoma Associated Myocilin Olfactomedin Domain. Molecular BioMedical Seminar Series. 2021, November; Atlanta, GA.

4. For my solution NMR based projects, I am focused on developing a better understanding of the early stages of mOLF aggregation as well as what structural deviations differentiate a pathogenic variant from both WT and benign mutations. Preliminary work on this project has involved utilizing my optimized minimal media growth protocol to generate ^{15}N-labeled samples of WT mOLF and glaucoma-causing variants D380A and I499F. These purified protein constructs were sent to the Van Horn lab at ASU where ^{15}N-HSQC spectra were acquired for all samples, revealing good signal dispersion and resonance overlap for WT and D380A that is suggestive of both proteins adopting a similar native state. Fewer resonances present for the spectra of D380A suggest this construct is more dynamic and likely accesses a low-lying excited state more prone to aggregation. The spectra for I499F had poor signal dispersion and few sharp resonances present, consistent with this more severe mutant being poorly folded with likely only the adoption of a slight residual structure. These exciting preliminary results motivated our desire to determine specifically which resonances within mOLF are most relevant to aggregation, specifically by identifying the resonances not present in the D380A and I499F spectra, as these are the residues experiencing increased dynamics. To address this question, I produced a ^{13}C, ^{15}N, ^{1}H-uniformly labeled WT mOLF protein sample that was sent to ASU where the Van Horn lab acquired a wide range of 3D NMR spectra required for resonance assignments. I have thus far been able to assign more than half of the WT mOLF resonances in the HSQC, and plan to wrap up these assignments within the next few months. Completion of assignments will help us determine the most aggregation-relevant regions of mOLF, as well as help us develop a better understanding of what differentiates a glaucoma-causing mutant from non-pathogenic variants and WT, from a structural perspective. This work has resulted in multiple poster and oral presentations, listed below.

a. **Saccuzzo, E**., Hill, S. E., Lieberman, R. L. Using solution NMR to observe dynamics of the glaucoma-associated olfactomedin domain of myocilin (mOLF), a five-bladed β-propeller. UAB-SERCAT Structural Biology Symposium. 2019, March; Birmingham, AL.
b. **Saccuzzo, E**., Ma, M., Hill, S. E., Martin, M., Lieberman, R. L. Structure and dynamics of the myocilin olfactomedin domain: differentiating benign from glaucoma-causing mutations. ISER/BrightFocus: Breakthroughs in glaucoma research. 2019, October; Atlanta, GA.
c. Scelsi, H., Barlow, B., **Saccuzzo, E**., Lieberman, R. L. (2021). Common and rare myocilin variants: Predicting glaucoma pathogenicity based on genetics, clinical and laboratory misfolding data. *Human Mutation*, accepted 05/31/21
d. **Saccuzzo, E.**, Mebrat, M., Van Horn, W., Lieberman, R. L. Solution characterization of glaucoma-associated myocilin. Georgia Institute of Technology Department of Chemistry and Biochemistry Annual Retreat. 2021, Atlanta, GA.

5. I was also fortunate enough to have the opportunity to work on a project regarding the SARS-CoV-2 outbreak, specifically helping to design a diagnostic tool kit that can be widely produced. My contribution involved helping to optimize the production and purification of TAQ-polymerase, an enzyme needed for polymerase chain reaction (PCR) and a key reagent for our test kits. This work resulted in a publication, listed below.
   a. Mascuch, S. J., Fakhretaha-Aval, S., Bowman, J. C., Ma, M. T. H., Thomas, G., Bommarius, B., ... **Saccuzzo, E**., …, & Lieberman, R. L.. (2020). A blueprint for academic laboratories to produce SARS-CoV-2 quantitative RT-PCR test kits. *Journal of Biological Chemistry*, *295*(46), 15438-15453.

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

Emory University

YEAR	SCIENCE COURSE TITLE	GRADE
2020	Biology of the Eye	B+

Georgia Institute of Technology

YEAR	SCIENCE COURSE TITLE	GRADE
2019	Enzymology and Metabolism	A
2019	Biophysical Chemistry	B
2018	Macromolecular Structure	A
2018	Molecular Biochemistry	B
2018	Signaling Molecules	B

University of Connecticut

YEAR	SCIENCE COURSE TITLE	GRADE
2018	Inorganic Chemistry Lab	A
2018	Physical Chemistry	A-
2018	Physical Chemistry Lab	A
2018	Thesis Undergrad Chem Majors	A
2018	Fundamental of Microbiology	A-
2018	Intro to Biophysical Chemistry	A
2017	Undergraduate Research	A
2017	Instrumental Analysis I	A-
2017	Physical Chemistry	A
2017	Elem Differential Equations	B+
2017	Biomolecular NMR Spectroscopy	B
2017	Intermediate Inorganic Chem	B+
2017	Quantitative Analytical Chem	B

YEAR	SCIENCE COURSE TITLE	GRADE
2017	Multivariable Calculus	A
2017	General Physics Problems	B
2016	Undergraduate Research	A
2016	Descriptive Inorganic Chem	B+
2016	Introduction to Biochemistry	B-
2016	General Physics	B
2016	Earth’s Dynamic Environment	A-
2016	Genetics	A-
2016	Organic Chemistry 2	B+
2016	Organic Chemistry Lab	B+
2016	Calculus 2	A-
2015	Organic Chemistry 1	B-
2015	Calculus 1	C
2015	General Physics 2	B
2015	General Chemistry 2	A
2015	Principles of Biology 1	A
2015	Principles of Biology 2	C+
2015	General Chemistry 1	A-
2015	Precalculus	C

YEAR	OTHER COURSE TITLE	GRADE
2017	Classical Mythology	A
2016	Multicultural Ed, Equi, Soc Ju	A
2016	Sem/Clin:Teaching&Learning	A
2016	Exceptionality	B+
2016	Educational Psychology	B+
2016	Language and Mind	A
2015	United States History	B
2015	Intro to Sociology	A-
2015	Popular Music&Diversity	A
2015	Seminar in Academic Writing	A-
2015	Intro to Anthropology	B+
2015	Problem Solving	A
2015	General Psychology 1	C+
2015	Intermediate Spanish 1	A