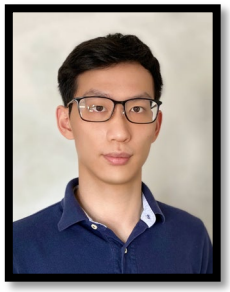
Houston Area Molecular Biophysics Program (HAMBP) Training Program
Grant No. T32 GM008280

Program Director: **Theodore Wensel**, PhD
Professor and Chair, Department of Biochemistry and Molecular Biology
Baylor College of Medicine

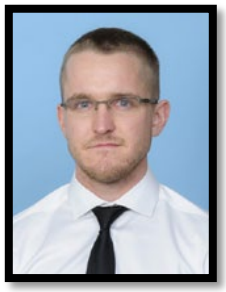
http://www.gulfcoastconsortia.org/home/training/molecular-biophysics-hambp/

Meet the Trainees
2022-2023

Sara Abouelniaj
Appointed August 1, 2022 – July 31, 2023
Department of Materials Science and NanoEngineering, Rice University
Primary Mentor: Dr. Yimo Han, Materials Science and NanoEngineering (RU)
Investigation of the Structural Dynamics of Voltage-Responsive Membrane Proteins
Biological membranes and their embedded proteins are of tremendous significance in modern biology and medicine, as they are encoded in approximately 30% of genes in most genomes and are the target of over 50% of FDA-approved drugs. Among them, transmembrane proteins are essential for the transport of ions and molecules in and out of the cells, making them of significant interest in cellular pathways and drug discovery. Voltage-gated ion channels (VGICs) are transmembrane proteins that activate in response to changes in the voltage (membrane potential), allowing specific ions to travel across these channels. Better study of the structure of these channels will be crucial in understanding the functionality and properties, which can be the target for developing drugs for mechanical pains, seizures, or cardiac arrhythmias. Though studies have been made, usual techniques disrupt the membrane potential by studying the VGIC only under its active state. Here, I propose to develop a voltage-gated nanodevice for cryogenic electron microscopy by using techniques from materials science, which will allow us to investigate the protein in its native electrical potential environment. This would enable us to gain new insights into the molecular mechanisms of these channels, allowing better understanding and utilization of the pore-opening, and ion-inductance mechanisms, which would potentially lead to new drug delivery methods by targeting the voltage sensors on these proteins. Not only that, but this will have a long-lasting impact on the research of all types of membrane proteins by providing protein structure in their native voltage potential.



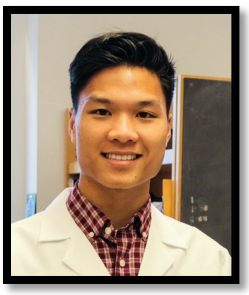
Caleb Chang
Appointed August 1, 2020 – July 31, 2023
Department of BioSciences, Rice University
Primary Mentor: Dr. Yang Gao, BioSciences (Rice Univ)
Adapting polymerase η with X-ray crystallographic techniques for drug design
Many current-day cancer treatments, including nucleoside-analog drugs remain ineffective, and frequently, patients end up developing resistance. The improvement of such therapy is severely limited by the inadequate knowledge of how polymerases differentiate between correct and incorrect DNA bases. The non-replicative translesion polymerase, polymerase η is unregulated in many cancers; thus, this protein remains an essential drug target for combined chemotherapy. I plan to use polymerase η as a model to investigate the molecular mechanisms behind polymerase fidelity and nucleoside-analog drug resistance with time-resolved X-ray crystallography. I also aim to find novel, allosteric inhibitors against polymerase η with fragment-based drug design screenings in collaboration with Xpose Therapeutics.



Nolan Dvorak
Appointed September 1, 2020 – August 31, 2023
Department of Pharmacology and Toxicology, University of Texas Medical Branch at Galveston
Primary Mentor: Dr. Fernanda Laezza, Pharamacology and Toxicology (UTMB)
Elucidating the Structural Constituents of Fibroblast Growth Factor 14 that Confer Its Modulatory Effects on the Voltage-Gated Sodium Channel 1.6
Disruption of protein:protein interactions (PPI) between voltage-gated Na^+ (Na_v) channels and their regulatory accessory proteins causes channelopathies. Despite restoration of these perturbed PPIs serving as a novel therapetuic approach, efforts to develop small molecule modulators of these surfaces is hindered by an incomplete understanding of the structural motifs of auxilary proteins that confer functional modulation of the pore-forming α subunit of Na_v channels. Focusing on the PPI between $Na_v1.6$ and its auxilay protein fibroblast growth factor 14 (FGF14), we employ whole-cell patch-clamp electrophysiology in conjuction with the application of pharmacological mimics of the β12 sheet and β8-β9 loop of FGF14 to interrogate the specific roles of these structural motifs in modulating the kinetics of $Na_v1.6$ channels. These functional studies are additionally coupled with advanced imaging techniques to validate the primacy of these structural motifs in regulating FGF14:$Na_v1.6$ complex assembly and channel trafficking. This approach that employs whole-cell patch-clamp electrophysiology, pharmacological probes, and advanced imaging techniques will enable the elucidation of crucial domains of auxilary proteins that confer functional modulation of Na_v channels through PPIs, strucutral insights that are crucial in developing efficacious small molecule modulators of perturbed PPIs within the Na_v channel macromolecular complex.

Mandi Feinberg
Appointed July 1, 2021 – June 30, 2023
Department of Biochemistry, Cellular, and Molecular Biology - Molecular Biophysics Educational Track, University of Texas Medical Branch at Galveston
Primary Mentor: Dr. Kyung Choi, Biochemistry and Molecular Biology (UTMB)
Structure of West Nile virus RNA promoter, stem-loop A, and its interaction with viral polymerase NS5
West Nile virus (WNV) is the causative agent of West Nile fever in humans, an emerging infectious disease, and the most common mosquito-borne disease in the United States. WNV replication is dependent upon the presence of the stem loop A (SLA) structure in the 5' untranslated region (UTR) of the viral genomic RNA. The viral polymerase, non-structural protein 5 (NS5), interacts with the 5' SLA and initiates synthesis of the negative strand. My project aims to identify and understand how WNV NS5 interacts with SLA to initiate RNA synthesis by determining the structure of the SLA in its native state and bound to the viral polymerase.

Clark Hamor
Appointed August 1, 2022 – July 31, 2023
Department of Biosciences, Rice University
Primary Mentor: Dr. Yizhi Jane Tao, Biosciences (RU)
Structural and functional characterization of +ssRNA viruses that infect a key coral reef symbiont
Coral reefs provide habitats for a diversity of ocean life. However, environmental stress has degraded many reefs, threatening the security and survival of species that rely on them. Coral colonies are formed from corals as well as diverse microorganisms, also called symbionts, that live inside them. In return for protection, symbionts give coral animals sugars they produce. Under stressful conditions, such as high sea temperatures, symbionts are lost from corals in a process called 'bleaching', which often causes coral death. Bleaching events are increasing in frequency and intensity due to climate change, and it is predicted that 90% of reef ecosystems will be threatened by 2050. Therefore, a comprehensive understanding of the factors contributing to bleaching is necessary to inform conservation efforts seeking to protect coral reefs. A group of symbiont-infecting viruses (dinoRNAVs) are thought to contribute to some coral bleaching. It is postulated that dinoRNAV infection may sensitize symbionts to heat stress, as dinoRNAVs appear to replicate in conditions similar to those that cause bleaching. Despite this, dinoRNAVs remain understudied. My research is focused on determining what these viruses look like to facilitate their identification in coral colonies. I will also study the structure and function of nonstructural gene products of dinoRNAVs. Lastly, I will perform infection assays using symbiont cultures to understand how dinoRNAVs replicate and respond to environmental stress, like elevated temperatures. Such information will provide insight into the role dinoRNAVs play in coral bleaching, and aid in developing approaches to increase coral resilience to climate change.



Anthony Hoang
Appointed July 1, 2020 – June 30, 2023
Department of Chemical, Physical, and Structural Biology, Baylor College of Medicine
Primary Mentor: Dr. Ming Zhou, Biochemistry and Molecular Biology (BCM)
Mechanism of iodide and bicarbonate transport and inhibition in mammalian pendrins
Pendrin is a crucial anion exchange transporter with iodide and bicarbonate transport capabilities. Mutations in iodide transport are known to lead to Pendred syndrome, a genetic disorder leading to congenital deafness and goiter, while overexpression of its bicarbonate transport may lead to hypertension. Drug discovery and therapeutics would benefit from understanding the mechanisms of transport, including details on ion binding sites, conformational changes during transport, and the role of a highly conserved region. I aim to use cryo-electron microscopy single particle reconstruction (cryo-EM SPR) to solve structures for pendrin during transport, and to verify its function and binding with proteoliposome transport assays and surface plasmon resonance (SPR). I also propose to analyze inhibition by performing virtual screens, and test binding affinity and thermodynamics with isothermal titration calorimetry (ITC) and a liposome flux assay.

Jordan Johnson
Appointed September 1, 2021 – August 31, 2023
Department of Biology and Biochemistry, University of Houston
Primary Mentor: Dr. Yuhong Wang, Biology and Biochemistry (UH)
Force Generation and Mechanism Elucidation of EF-Tu
Elongation factor thermal unstable (EF-Tu) is a bacterial enzyme that delivers aminoacyl-tRNAs to the ribosome during translation, however its mechanism is not well understood. My project will be observing the conformational changes of EF-Tu using FRET and determining if a power stroke is generated, similarly to other elongation factors, using Force Induced Remnant Magnetization Spectroscopy (FIRMS). I will also observe how mutations in the GTP binding pocket will affect EF-Tu's mechanism and function as mutations in EF-Tu have been linked to a variety of health issues and it is unknown how mutations contribute to the occurrence of these health issues.

Savannah Seely
Appointed August 1, 2021 – July 31, 2022
Department of Biochemistry and Molecular Biology, The University of Texas Medical Branch
Primary Mentor: Dr. Matthieu Gagnon, Microbiology and Immunology (UTMB)
Investigating the Molecular Mechanism of Ribosome Recycling
In all organisms, the ribosome decodes mRNA and synthesizes proteins through four essential steps: initiation, elongation, termination, and recycling. Currently, more than 50% of clinically relevant antibiotics target the ribosome. Remarkably, the final step, recycling, is the least characterized and has not been exploited for structure-based drug design. Recycling is the necessary bridge between termination and initiation, and a better understanding of the molecular aspects of this step could lead to development of new therapeutics. I propose to determine the molecular mechanism of recycling in the human pathogen *Pseudomonas aeruginosa*, which employs a specialized Elongation Factor-G that functions exclusively in recycling. I will also investigate features within the ribosome that are involved in its disassembly by determining the structure of amikacin bound to the ribosome; an antibiotic that has been reported to cause recycling deficits. Taken together, I will determine at high resolution the mechanism of ribosome recycling by elucidating what ribosome features are altered as well as what molecular interactions occur between the ribosome and recycling factors. The anticipated results of these studies will improve our knowledge of this step-in translation and may open new doors for the development of antibiotics.

Justin Van Riper
Appointed August 1, 2022 – July 31, 2023
Department of Biochemistry and Cellular Biology, Baylor College of Medicine
Primary Mentor: Dr. Monica Pillon, Biochemistry and Cellular Biology (BCM)
Characterization of the FASTKD4 mitochondrial RNA binding protein
Mitochondrial homeostasis is critical for vital biological processes such as energy production. Mitochondrial plasticity is largely orchestrated by nucleic acid binding proteins tasked with regulating the mitochondrial genome, transcriptome, and proteome. While strict mitochondrial gene regulation is imperative to maintain ATP pools, little is known about the molecular mechanisms that govern mitochondrial gene regulation. FASTKD4 is a poorly characterized post-transcriptional regulator linked to precursor mitochondrial RNA maturation. I am mounting a biophysical and biochemical program aimed at elucidating FASTKD4 function and regulation in mitochondrial RNA processing.