UNIVERSITY OF NOTRE DAME

# Colleges of Science & Engineering Joint Annual Meeting

## 4th Annual COSE-JAM for Graduate Students & Postdoctoral Fellows

**Thursday – December 9, 2021**

**Jordan Hall**

# COSE-JAM 2021 Schedule

*Morning Podium Session (9:00-12:00pm):* 22 podium presentations in Jordan auditoriums (101 & 105) (continental breakfast/snacks/drinks in Galleria; 10:15-10:30 mid-morning break)

*Lunch (12-1:00pm):* Jordan Galleria – participants, discussants, moderators, co-authors, etc.

*Afternoon Podium Session (1:00-2:30pm):* 6 podium presentations in Jordan auditorium 101

*Afternoon Poster Session (2:30-4:30pm):* 38 poster presentations in Jordan Galleria (light snacks/soft drinks in Galleria)

*Afternoon Social (3:30-4:30pm):* Jordan Galleria – opportunities for formal and informal peer-to-peer interactions (snacks/soft drinks/beer/wine)

## Morning Podium Session (9:00am-12:00pm, Jordan Hall 101 & 105)

**Jordan 101:**

9:00 – **Eckhart Spalding** : Instrumentation for the Direct Imaging of Exoplanets

9:15 – **Kathleen Nicholson** : EspN is a novel regulator of the ESX-1 secretion system that is required for *Mycobacterium marinum* pathogenesis

9:30 – **Hannah Wesselman** : Esrrγa regulates nephron development and ciliogenesis by controlling prostaglandin synthesis and cooperation with Ppargc1a

9:45 – **Maggie Fink** : Impact of metabolic crosstalk on polymicrobial communities containing *Pseudomonas aeruginosa* and *Enterococcus faecalis*

10:00 – **Chelsea Southworth** : Pregnancy is associated with distinct gut microbiome features in adult female baboons

10:15 – 10:30: Break

10:30 – **Kurtis Breger** : Elucidating the Kinetic Mechanism of Human METTL16

10:45 – **Taylor Tobin** : Pictures of Exoplanets around Accelerating Stars: A Direct Imaging Survey with SCExAO/CHARIS

11:00 – **Khoa Nguyen** : emx2 is required for nephron segment development in the zebrafish pronephros

11:15 – **Shirantha Welikala** : Efficient Robust Path Planning Under Temporal Specifications for Mobile and Aerial Robots

11:30 – **Marycruz Flores Flores** : Characterization of cell recruitment mechanism driven by vestigial in the imaginal wing disc of *Drosophila melanogaster*

11:45 – **Chinedu Madukoma** : Multigenerational type IV pili inhibition of *Pseudomonas aeruginosa*

**Jordan 105:**

9:00 – **Reyer Band** : Upgrades to Off-Detector Electronics of the CMS Electromagnetic Calorimeter

9:15 – **Brooke Stemple** : Evaluation of the microbial community and geochemistry in produced water collected from $CO_2$-EOR in the Niagaran Pinnacle Reef

9:30 – **Derek Shank** : Milky Way Substructure: Discovering Dynamical Groups Using Stellar Surveys

9:45 – **Olivia Ginn** : Effect of Substrate on Removal Rates of Antibiotic Resistance in Controlled Experimental Streams

10:00 – **Angélica García-Martínez** : Validation and efficacy of a prognostic model for control domiciliary of the event of fever and neutropenia in children with cancer

10:15 – 10:30: Break

10:30 – **Jaynise Perez** : On Synergy between Equatorial Convective Disturbances and Monsoon Intraseasonal Oscillations in the Bay of Bengal

10:45 – **Nicole Weaver** : Delineating the role of *gldc* in nephron patterning during kidney development

11:00 – **Mindo Choi** : Performance of multi-resolution Coastal Ocean Modeling for storm surge in Alaska, US

11:15 – **Alexis Waldschmidt** : Visual Impairment in *norpA*-Mutated *Aedes* Mosquitoes

11:30 – **Madison Schmidtmann** : Elucidating the cellular mechanisms involved in breast tumor progression

11:45 – **Alex Boomgarden** : Investigating tumor microvesicle cargo and function

## Afternoon Podium Session (1:00-2:30pm, Jordan Hall 101)

**Jordan 101:**

1:00 – **Ryan Posh** : Hybrid Volitional Control for Lower-Limb Prostheses

1:15 – **Vanessa Rubio** : Functional groups, determinism and the dynamics of a tropical forest

1:30 – **Harrison Hill** : Generation of functionalized azepinone derivatives via a (4+3)-cycloaddition of vinyl ketenes and α-imino carbenes derived from N-sulfonyl-triazoles

1:45 – **Adrian Navarro Hernandez** : Planar Brownian Motion: Kinetic Monte-Carlo and Asymptotic Methods and the Role of Directional Sensing

2:00 – **Nilay Bostan** : Hadron production measurements for neutrino physics

2:15 – **Leah Lund** : Correlating tumor microenvironment stiffness and intracellular pH dynamics

## Afternoon Poster Session (2:30-4:30pm, Jordan Galleria)

### Session 1: Bryant – Pruitt present for questions from 2:30-3:30pm

1. **Annamarie Bryant:** CLIPR-76: a novel CLIPR protein linking membranes and microtubules

2. **Emma Cobian:** Variational Inference Via Normalizing Flows and Adaptive Annealing

3. **Rachel Cronin:** Proteo-genetic analysis of ESX-1 secretion reveals distinct roles for ESX-1 substrates in *Mycobacterium marinum*

4. **Homero Domínguez:** Penicillin-Binding Protein 2x as a Target in Allosteric Inhibition by Antibiotics

5. **Xiaozheng Dou:** Late-stage Modification and Biological Evaluation of Cruentaren A Derivatives

6. **Michael Dugas:** Chemically Resilient Nanofiltration Membranes Fabricated from Copolymers for Organic Solvent Nanofiltration

7. **Madeline Glennon:** Establishing the binding mode of a diphenylfuran to an RNA triple helix

8. **Benjamin Gombash:** Multiple Factor Analysis Reveals Variable Interactions Between the Diet and Mycobiome of Long-tailed Macaques (Macaca fascicularis) on Two Islands in Southeast Asia

9. **Kathleen Hayes:** Recent advances in deployability of paper-based field test for illicit drugs

10. **Hanna Hlushko:** Silicon and Zirconium Ceramic Material Irradiation in the Presence of Water

11. **Tiffany Huwe:** Strong clustering of asymptomatic Plasmodium falciparum and Plasmodium vivax infections among ethnic groups but absence of population structure in Chittagong Hill Tracts, Bangladesh

12. **Bradley Jones:** A Conserved N-Acetyltransferase is Responsible for N-terminal Acetylation of EsxA

13. **Charlotte Kunkler:** A Single Modified Nucleoside Destabilizes an RNA•DNA-DNA Triple Helix

14. **Guoming Ling:** Development and Validation of an Alaska Coastal Ocean Forecast System with Coupled Tides, Storm Surge and Waves under Sea Ice Conditions

15. **Daniele Miranda**: A Survey of Sportfish for Per- and Polyfluoroalkyl Substances (PFAS) – An Emerging Contaminant in the Great Lakes

16. **Jonathan Ouimet:** Automated Diafiltration Experiments Advance Material and Process Development

17. **Dillon Peng:** Upgrading the Gemini Planet Imager: Testing and Installation of New Cameras for GPI 2.0

18. **Rebecca Prest:** Molecular Analysis of a Bifunctional Mycobacterium tuberculosis Virulence Factor

19. **Abagael Pruitt:** Examining the influence of periodical cicadas on stream nutrient uptake and their roles as a resource subsidy

## Afternoon Poster Session (2:30-4:30pm, Jordan Galleria)

### Session 2: Riney – Zhong present for questions from 3:30-4:30pm

**20. Logan Riney:** Fermi level tuning and band alignment in Mn doped InAs/GaSb

**21. Chissa Rivaldi:** Bacterial Community Association with Key Protozoans in the Guts of Long-Tailed Macaques in Southeast Asia

**22. Kevin Sanchez:** Structural and Functional Characterization of a Conserved Mycobacterial Transcription Factor

**23. Anna Santa:** Effect of clinical mutations in OXA-24/40: an analysis of chemical shift temperature dependence

**24. Mika Schievelbein:** Selecting for an Aptamer that Targets RNA Triple Helices

**25. Minwoo Shin:** High order collision-based hybrid method for the BGK equation

**26. Krishna Shivakumar:** Selective Recognition of RNA Triple Helices by Small Molecules

**27. Elise Snyder:** Velocity and substrate conditions influence the particle size distribution of environmental DNA (eDNA) in recirculating mesocosms

**29. Shannon Stoffel:** Investigations of Macrocycle Breathing Dynamics in a Cyclodextrin-based Rotaxane

**30. Emma Thrift:** Agricultural conservation influences nutrient and pathogen dynamics at both the field and subwatershed scale

**31. Emma Troth:** High-throughput screening of a novel structure-activity library against N. fowleri in a newly developed cytopathogenicity assay

**32. Anna Vincent:** The Effects of Conservation on Ammonium Dynamics in Two Agricultural Watersheds

**33. Sabrina Volponi:** The Use of an Inverse Multi-Domain Rate Transfer Model in Predicting Colloid Attachment under Different Environmental Charge Conditions

**34. Jiashu Wang:** Coexistence of superconductivity, fluctuations and spin-orbit splitting in Sn1-xInxTe thin film

**35. Yu Wang:** Variational Inference with NoFAS: Normalizing Flow with Adaptive Surrogate for Computationally Expensive Models

**36. Jialing Xu:** Fabrication of a hierarchical polymer membrane adsorbent by a combined 3-D printing and in-situ phase separation process

**37. Joseph Zepeda:** Metal-Poor Stars Observed with the Southern African Large Telescope II. An Extended Sample

**38. Shukun Zhong:** Stripe-patterned membrane for multi-ions detection

# *Abstracts for Oral Presentations*

***Upgrades to Off-Detector Electronics of the CMS Electromagnetic Calorimeter***

Reyer Band
Department of Physics
Advisor: Colin Jessop

The Compact Muon Solenoid (CMS) detector currently records collisions of protons at the Large Hadron Collider (LHC) at a rate of approximately 40 MHz. This rate will substantially increase with the High Luminosity LHC upgrade, which will operate at a peak luminosity of $5 \times 10^{-34}$ $cm^{-2}$ $s^{-1}$. The electromagnetic calorimeter, which provides hermetic coverage for pseudo rapidity values $|\eta| < 5$, will have its front- and -back-end electronics upgraded to increase the trigger rate and latency to 750 kHz and 12.5 μs. Hardware upgrades to the on-detector electronics will enhance timing resolution and allow for suppression of anomalous signals. This presentation will describe the development of a new off-detector calorimeter processor board which utilizes commercially available FPGAs to provide fast trigger generation, control of on-detector electronics, and conversion of digitized pulses to particle energies.

***Investigating tumor microvesicle cargo and function.***

Alex Boomgarden
Department of Biological Sciences
Advisor: Crislyn D'Souza-Schorey

Several cancers including ovarian and prostate cancers remain dubbed as "silent killers" due to the subtlety and elusiveness of symptoms, delaying diagnosis to a more advanced and often lethal stage. This has caused for heightened pressures towards the advancement of new techniques and tools focused on both prevention and diagnostics. New and emerging advances in liquid biopsies which includes the use of extracellular vesicles (EVs), holds great promise in this regard. One subtype of EVs, tumor microvesicles (TMVs) that are shed from the surface of tumor cells, have emerged as important regulators of cancer progression and through their cargoes can be informative of the molecular profile of a developing tumor. Additional investigations surrounding this exciting and bourgeoning field include defining the functions of individual populations of EVs in the tumor microenvironment. Our knowledge in this regard remains constrained in large part due to limitations in isolating and precisely defining the molecular makeup of individual EV subtypes. Motivated by these considerations, our project aims to identify novel cargos and regulatory proteins within TMVs using a recently developed pupylation-based interaction tagging system. Given our prior findings that TMVs are enriched in the ARF6 protein and that ARF6-regulated membrane recycling directs proteins and microRNAs to the cell surface for incorporation into nascent microvesicles, we have utilized pupylation-based interactions as a proximity ligase screen to interrogate the ARF6 interactome in TMVs. We have successfully engineered our tumor cell model to tag and identify proteins solely within ARF6-positive TMVs, and preliminary results have enabled us to validate previously identified cargos. Generation of a comprehensive TMV proteome across multiple tumor cell lines will provide insight into the molecular cargos dictating pro-cancerous effects on TMVs in the tumor microenvironment, and will also allow the identification of novel tumor biomarkers.

## *Hadron production measurements for neutrino physics*

Nilay Bostan
(On behalf of NA61/SHINE and EMPHATIC experiments)

Department of Physics
Advisor: Prof. Laura J. Fields

The determination of the accurate neutrino flux is very important for the accelerator-based neutrino experiments to understand the neutrino interactions and neutrino oscillation measurements. In these experiments, highly energetic (120 GeV) protons interact with a nuclear target to produce mesons, predominantly pions and kaons that decay to a neutrino. In neutrino beamlines, the largest uncertainty on the produced neutrino flux comes from the hadronic cascade model which are utilized to predict the hadron production (HP) within the target and the beamline elements. Due to the discrepancy between HP models is quite large, neutrino experiments need to use the external hadron production data to constrain the hadronic models in their simulations and estimate the neutrino flux at their detectors with better reliability. The implementation of these data significantly reduces the systematic uncertainties that arise from the hadron interactions mismodeling.

Currently, the NA61/SHINE (CERN) and EMPHATIC (Fermilab) experiments are actively working to provide the hadron production measurements at different energies, different nuclear targets, and particle projectiles for the accelerator-based neutrino experiments. In this talk, I will briefly explain these hadron production experiments and their status.

**Elucidating the Kinetic Mechanism of Human METTL16**

**Kurtis Breger**

**Department of Chemistry & Biochemistry**

**Advisor: Jessica A. Brown**

Over 140 RNA modifications have been discovered, yet only recently have they been studied in depth due to recent technological advancements. $N^6$-methyladenosine ($m^6A$) is an abundant RNA modification in messenger RNA (mRNA) and long non-coding RNA that affects various cellular functions such as mRNA stability, phase-separation of RNAs, and others. Methyltransferase-like protein 16 (METTL16) is one of four catalytically active $m^6A$ RNA methyltransferases in humans. Two well-known methylation targets of METTL16 are U6 spliceosomal RNA and hairpins in the 3' untranslated region of MAT2A mRNA. However, METTL16 binds to many other RNAs, including the 3' triple helix of MALAT1. Using *in vitro* assays, we have started to investigate the kinetic mechanism and other fundamental properties of METTL16. Thus far, we have determined that METTL16 is a monomer in complex with either U6 snRNA or the MALAT1 triple helix. The METTL16•RNA complex has a dissociation constant ($K_D$) of 18 nM with the U6 snRNA and 31 nM with the MALAT1 triple helix. The apparent dissociation constant for S-adenosylmethionine (SAM), the methyl donor, with the METTL16•U6 snRNA binary complex is 112 µM. Under *in vitro* conditions, the MALAT1 triple helix is not a substrate of METTL16 at position A8290 and other adenosine residues. Preincubation assays suggest that there is an ordered mechanism by which U6 snRNA binds to METTL16 before SAM. Steady-state assays established a $k_{cat}$ of 0.074 $min^{-1}$ and single-turnover assays established a $k_{chem}$ of 0.56 $min^{-1}$ which may that product release may be rate limiting. Ongoing work includes the characterization of METTL16 mutants. Mutations in the METTL16 K-loop led to a 7-fold increase of SAM binding to METTL16•U6 snRNA. Future studies will focus on METTL16 mutants, including residues in other structures and those identified in cancer, to ascertain how these alterations affect the kinetic mechanism. This study will enable future research on METTL16 as a therapeutic target.

***Performance of multi-resolution Coastal Ocean Modeling for storm surge in Alaska, US***

Mindo Choi
Department of Civil and Environmental Engineering and Earth Sciences,
University of Notre Dame, Notre Dame, IN
Advisors: Joannes J. Westerink and Damrongsak Wirasaet
Coauthor: Guoming Ling and Maria Teresa Contreras Vargas

Abstract: Understanding coastal and ocean hydrodynamics in Alaska (US) is crucial for assessing fishery and oil refinery operations and optimizing coastal protection strategies, mainly due to the rough winter weather conditions characterized by low atmospheric pressure storms with very strong winds. The current Alaska Coastal Ocean Forecasting System (Ling et al., 2021) forecasts water levels once a day for the upcoming five days. The model has a minimum resolution of 100-200 m, enough to provide accurate predictions only at basin-to-estuarine scales. However, the simplification of the nearshore region does not allow the model to capture the hydrodynamics of inlets, rivers, and smaller-scale topographic features. We implemented a multi-resolution ocean model for water level estimation in Alaska that seeks to upgrade the current forecasting system to address this problem. By using the most updated mesh generation tools (OceanMesh2D/Mesh2D), along with recently published shoreline and bathymetric databases, we refined the model down to 50-100 m minimum resolution nearshore and optimized the nodal distribution prioritizing topo-bathymetric features relevant for the hydrodynamics of the system. We have controled the quality of the model by exhaustively comparing it with satellite images, maps, and the original databases. We validated the model using ADCIRC for tides and storms, forcing winds, atmospheric pressure, and waves. We compare our results with the current Alaska Coastal Ocean Forecasting System assessing the maximum water levels and water levels time series and comparing our results with a set of observations from NOAA coastal gauges.

***Impact of metabolic crosstalk on polymicrobial communities containing Pseudomonas aeruginosa and Enterococcus faecalis***

Maggie Fink
Department of Biological Sciences
Advisor: Joshua Shrout
Coauthors: Abigail Weaver

The bacterium *Pseudomonas aeruginosa* is an opportunistic pathogen that can cause lung, skin, and eye infections. *P. aeruginosa* has been identified in prosthetic joint infections (PJIs) along with several other pathogenic bacteria including *Staphylococcus aureus, Enterococcus faecalis,* and *Corynebacterium striatum*. In most conditions, *P. aeruginosa* engages in a competitive interaction with other bacterial species, employing its extensive arsenal of virulence factors and metabolic flexibility to become the dominant species in polymicrobial communities; however, when co-cultured with *E. faecalis*, *P. aeruginosa* exhibits and attenuation of competitive behaviors and production of *Pseudomonas* quorum sensing signal (PQS). Additionally, when these two species are co-cultured in conditions with sugars not metabolizable by *P. aeruginosa*, metabolic crosstalk is observed, resulting in more robust growth of *P. aeruginosa.* These results also implicate the role iron and production of *P. aeruginosa* siderophores are key to the mediation of these cooperative interactions. Using a variety of techniques, including fluorescence microscopy, biofilm assays, and mass spectrometry, these results provide a temporal, spatial, and biochemical characterization of dual-species interactions in defined environments. These findings provide a framework for understanding how polymicrobial interactions can initiate colonization in specific infections environments through metabolic cooperation and alter the bacterial composition of polymicrobial infections.

**Validation and efficacy of a prognostic model for control domiciliary of the event of fever and neutropenia in children with cancer**

Angélica García-Martínez
Computer Science and Engineering
Advisor: Nitesh Chawla
Coauthors: Jennifer Schnur, Jasmin Botello, Sugana Chawla, Patrick Soga, Javier Olivar, Elisa Dorantes, Martha Avilés, Horacio Márquez, Paula Cárdenas, Emanuel Orozco, Edson Serván-Morin.

Fever and neutropenia are complications derived from chemotherapy that are associated with the development of septic shock and high mortality rate. It is critical to have an early detection of fever and neutropenia risk in pediatric patients to appropriately inform interventions; however, there is, yet, no singular risk index to govern risk progression among pediatric patients, especially in low- and medium-income countries, largely stemming from the lack of sufficient clinical infrastructure (resources, data, tools). We are collaborating with the Hospital Infantil de México Federico Gómez (HIMFG) to develop a risk prediction model for bacteremia, septic shock, and death, in pediatric oncological patients with fever and neutropenia. It is expected that this risk prediction model that can help identifying high risk and low risk patients so that appropriate interventions can be activated. For e.g., high risk patients might be hospitalized, and low risk patients might be situated at associated shelters or even go back to their communities with a care plan that can be coordinated with the health care providers in their community, as available and applicable. A commonly used risk index is the *Santolaya* criteria that was developed in Chile; this index, however, only uses clinical attributes and does not consider any social and structural information, and as such can be incomplete about risk evaluation. In our work, we are developing a risk model that builds on the clinical attributes, relevant to the Mexican context, and also incorporates the social and structural determinants about the families. In addition, we are also developing an application that will be used by the physicians, families, and the care providers in the respective communities (social workers, physicians). The results expected are minimizing the chances of complications, promoting well-being and quality of life in cancer patients, minimizing hospital costs and improve Hospital bed capacity.

**Effect of Substrate on Removal Rates of Antibiotic Resistance in Controlled Experimental Streams**

Olivia Ginn
Civil and Environmental Engineering and Earth Sciences
Advisors: Kyle Bibby and Jennifer Tank
Coauthors: Andrei Badilla Aguilar and Ellie Snyder

A majority of global antibiotic use is accounted for by animal husbandry. Specifically in the United States, it accounts for 60% of all antibiotic consumption. Antibiotics used in animal husbandry are excreted untransformed in an approximate 1.1 billion tons of animal manure produced per year. Much of this manure is subsequently land applied as soil amendment or fertilizer leading to potential runoff and microbial pollution in surface waters. In order to effectively inform antimicrobial resistance (AMR) monitoring and mitigation efforts, a thorough understanding and description of the persistence and transport of AMR in flowing waters is needed. Utilizing controlled experimental mesocosms as proxies for streams, we assessed the degradation, removal, transport and sorption of AMR originating in cow manure from a dairy farm in the Paw Paw River Watershed in Michigan. We assessed the effects of three substrate variations (pea gravel, no substrate and fine benthic organic matter or FBOM) and collected water samples prior to spiking and at 0, 4, 16, and 24 hours after spiking. We hypothesized that tetracycline and erythromycin resistance associated DNA (tetA and ermB respectively) would be removed faster from the water column with pea gravel as the substrate. The decay rates for ermB in pea gravel, no substrate, and FBOM were -1.85 $day^{-1}$, -1.49 $day^{-1}$, -1.94 $day^{-1}$ respectively. TetA started at a higher concentration and was not removed from the water column after 24 hours, though decay occurred faster. Decay rates for tetA in pea gravel, no substrate and FBOM were -3.84 $day^{-1}$, -2.4 $day^{-1}$, and -3.84 $day^{-1}$ respectively. Our results indicate that antibiotic resistance may remain in the water column for over 24 hours, providing ample opportunity for spread through bacterial horizontal gene transfer. Furthermore, our results reveal that substrate may play a role in AMR removal from streams.

***Generation of functionalized azepinone derivatives via a (4+3)-cycloaddition of vinyl ketenes and α-imino carbenes derived from N-sulfonyl-triazoles***

Harrison Matthew Hill
Department of Chemistry and Biochemistry
Advisor: Brandon L. Ashfeld
Coauthors: Zachary D. Tucker, Kevin X. Rodriguez, Katelyn Wendt, and Brandon L. Ashfeld

An intermolecular $Rh^{II}$-catalyzed, formal (4+3)-cycloaddition between vinyl ketenes and *N*-sulfonyl-1,2,3-triazoles for the construction of biologically interesting azepinone products is described. Utilizing *N*-sulfonyl-1,2,3-triazoles as "masked," α-imino diazo compounds in conjunction with employing vinyl ketenes as 1,3 dipolar surrogates as opposed to the far more commonly employed dienyl moieties allows for both an intermolecular and selective formation of azepinone products derived from a formal (4+3)-cycloaddition over a potential (3+2)-cycloadduct under mild reaction conditions.

## *Correlating tumor microenvironment stiffness and intracellular pH dynamics*

Leah Lund
Department of Chemistry and Biochemistry
Advisor: Dr. Katharine White

As an emerging hallmark of cancer, dysregulated pH dynamics may hold crucial information on the molecular mechanisms driving cancer cell metastasis and proliferation. Cancer cells have an increased intracellular pH (pHi > 7.4) and decreased extracellular pH (pHe ~6.9) relative to normal cells (pHi ~7.2, pHe ~ 7.4). In addition to dysregulated pH, cancer is associated with dysregulated extracellular matrix (ECM). Cancer cells experience a stiffer ECM compared to normal tissue due to increased protein production and crosslinking of ECM proteins. While these disruptions of normal pH and matrix in cancer have been characterized previously, it remains unknown how molecular cues from ECM-cell signaling may contribute to pH dysregulation and whether pH dysregulation reinforces stiffness-associated cancer cell behaviors. Here we show that two metastatic cell lines, one breast and one lung, exhibit a decrease in pHi on increasing ECM stiffnesses. We show this trend is preserved on two types of ECM with tunable stiffness (Matrigel and hyaluronic acid-based gels). Importantly, normal cell lines had no pHi response to varying ECM, suggesting a cancer specific response of pHi to stiffening ECM. Demonstrating a dynamic relationship between pHi and cell-matrix responses in cancer would allow us to further understand how cell metastasize and proliferate, which could provide new methods for prognosis and therapeutic targets.

Multigenerational type IV pili inhibition of *Pseudomonas aeruginosa*

**Chiendu S. Madukoma**[1] and Joshua D. Shrout[1, 2]

[1]Department of Civil and Environmental Engineering and Earth Sciences, University of Notre Dame, Notre Dame, IN, 46556; USA
[2]Department of Biological Sciences, University of Notre Dame, Notre Dame, IN, 46556; USA

*Pseudomonas aeruginosa* is a Gram-negative, ubiquitous, opportunistic human pathogen and a member of the ESKAPE family of bacterial pathogens. According to the Center for Disease Control and prevention (CDC), *P. aeruginosa* is among the leading cause of hospital acquired infections such as pneumonia, ventilator associated infections, and surgical site infections. Importantly, *P. aeruginosa* infection is most notable for its presence in cystic fibrosis lung infections, where it grows as a biofilms. Prior to biofilm development, *P. aeruginosa* move on surfaces, exhibiting TFP mediated motility phenotypes including twitching and snapping. In addition to TFP-mediated surface motility phenotypes, TFP also play important role in irreversible cell-surface attachment. These are all percussors for biofilm development. Prevention and treatment of *P. aeruginosa* biofilms are important to treatment. We recently identified an inhibitor for TFP. We found that supernatant from a quorum sensing signal (Δ*rhlI*) mutant leads to multigenerational TFP inhibition (MTFPI) of *P. aeruginosa* wild-type as new progeny also fail to exhibit TFP functions. Our current results implicates a factor that is proteinaceous in nature. We are continuing to characterize the MTFPI factor.

**Characterization of cell recruitment mechanism driven by vestigial in the imaginal wing disc of Drosophila melanogaster**

Marycruz Flores Flores[1], Jeremiah Zartman[2] & Marcos Nahmad[1]. [1]Department of Physiology, Biophysics and Neurosciences, CINVESTAV-Mexico.
[2]Department of Chemical and Biomolecular Engineering, University of Notre Dame, IN.

Organs mainly attain their size by cell growth and proliferation, but sometimes also grow through the recruitment of undifferentiated cells. Cell recruitment mechanism first described in the developing wing of Drosophila melanogaster, as a non-autonomous way to increase the wing cell population by recruitment of non-wing cells. Cell recruitment is driven by Vestigial (Vg), wing selector gene: a wing cell produces a signal that drives adjacent cells to differentiate into the same type as the inducers, i.e. wing cells. The newly recruited cells are capable to recruit their surrounding neighbors initiating a recruitment signal wave and establishing a feed-forward loop. Recently, we have found that impairing cell recruitment genetically results in adult wings that are approximately 20% smaller and cell recruitment is initiated between early and mid-third-instar larval development. Although the molecular components of cell recruitment have been described, the interplay between them has not been unraveled yet. Here we investigate the participation of each component of cell recruitment mechanism into the establishment of Vg pattern and how they interact together to propagate cell recruitment signal. Using genetics tools, we have found that a source of Vg is necessary for Vg pattern propagation and establishment. Besides, using VgQELacZ as a reporter of cell recruitment signal, we have found out that the signal can be propagated beyond the Vg expressing cells suggesting that the recruitment signal acts as a long-range signal. It has been described that cell recruitment also contributes to the development of different organs as the mammalian thyroid, inner ear, and heart, to fully understand cell recruitment mechanism will let us know how morphogenesis is controlled in animals since cell recruitment could be a conserved mechanism during organ development.

***Planar Brownian Motion: Kinetic Monte-Carlo and Asymptotic Methods and the Role of Directional Sensing***

Adrián Navarro Hernández
Department of Applied and Computational Mathematics and Statistics
Advisor: Dr. Alan Lindsay

Many cells are able to respond to an outside chemical stimulus after the stimulus detection. While the whole cell may not be able to perceive it, different sensing regions on the surface may detect various inputs. The diffusion of such stimuli from its origin towards the cell has been modeled using Planar Brownian motion. However, recreating Brownian motion and waiting for the stimulus' arrival towards the cell is an unbearably inefficient task due to having small targets in an infinite space. We still desire to understand relevant information such as the distribution of arrival times and distribution of arrival location in order to comprehend how sensors behave and where they are situated. To this end, an asymptotic method has been proposed using Laplace transforms of the diffusion equation, followed by a numerical transform inversion to compute the total flux absorbed by each receptor. This result was supported by an efficient Kinetic Monte-Carlo method developed to avoid the simulation of Brownian motion through large solvable steps; with the algorithm consisting of walks on spheres, reinsertion, reflection, and square projection steps. Our analysis indicates that homogenization tools are very successful in obtaining arrival statistics through more efficient receptor configurations.

## *emx2 is required for nephron segment development in the zebrafish pronephros*

Khoa T. Nguyen
Department of Biological Sciences
Advisor: Rebecca A. Wingert

The *empty spiracles homeobox gene 2 (emx2)* encodes a transcription factor that is expressed during neuronal, auditory, olfactory, and renal development, but its role in the latter has not yet been fully elucidated. Here, our objective is to utilize the zebrafish animal model to investigate the functions of *emx2* during the development of nephrons, which are the structural and functional units of the kidney. Using whole mount in situ hybridization (WISH), we detected *emx2* transcript beginning at the 6 somite-stage in renal progenitors and this expression continued throughout the process of forming discrete proximal convoluted tubule (PCT) and proximal straight tubule (PST) segment regions at 24 hours post fertilization (hpf). We generated *emx2*-deficient embryos and compared them to wild-type (WT) controls at various time points from 24-96 hpf. Knockdown of *emx2* resulted in head necrosis and diminished yolk sac extension at 24 hpf, as well as development of smaller head, eye and body, pericardial edema, abnormal fin growth, yolk ball inflammation and vascular hemorrhage in the central nervous system from 2-5 days post fertilization. WISH analysis revealed a significantly reduced podocyte area, diminished PCT and reduced multiciliated cell numbers in *emx2*-deficient embryos compared to WT embryos. WISH results also revealed secondary phenotypes, such as reduced fin bud area and somite in *emx2*-deficient embryos. Additionally, Alcian Blue staining at 96 hpf revealed jaw abnormalities and pharyngeal arch malformation in *emx2*-deficient embryos. O-Dianisidine staining showcased vascular hemorrhage in the eyes and head of *emx2* deficient embryos. We concluded that *emx2* is essential for kidney segmentation in zebrafish, especially in podocyte and PCT formation. Future studies will delineate the cellular defects in nephron development in *emx2* deficient embryos and determine the relationship between Emx2 and pathways which are essential for proximal segment ontogeny, such as retinoic acid signaling.

***EspN is a novel regulator of the ESX-1 secretion system that is required for Mycobacterium marinum pathogenesis***

Kathleen R. Nicholson
Department of Biological Sciences
Advisor: Dr. Patricia Champion

*Mycobacterium tuberculosis*, a human pathogen, and *Mycobacterium marinum*, a non-tuberculous mycobacteria (NTM), require the ESX-1 (ESAT-6 system 1) secretion system for virulence. The mycobacterial ESX-1 secretion system transports proteins important for infection of host macrophages. Previously, we showed protein secretion by the ESX-1 system is coupled to regulation of gene expression. We demonstrated that the WhiB6 transcription factor upregulates genes encoding ESX-1 secreted proteins in response to the presence of the ESX-1 secretory machinery. We also showed that the EspM transcription factor, whose gene is divergently encoded from the *whiB6* gene upstream of the *esx-1* genetic locus, represses *whiB6* gene expression in the absence of the ESX-1 secretory machinery. In an effort to identify additional transcription factors at the *whiB6-espM* promoter, we found a probable transcription factor, EspN. We found that EspN bound the *whiB6-espM* promoter only in the absence of EspM. Thus, we hypothesized that EspN regulates *esx-1* genes in the absence of EspM. To test this hypothesis, we used a combination of genetic and biochemical approaches to define the function of EspN in the ESX-1 transcriptional regulatory network. We demonstrate that EspN is an activator of *whiB6* transcription. We also show that EspN specifically regulates transcription of a subset of genes encoding ESX-1 secreted proteins. Additionally, we demonstrate that EspN is a probable regulator of ESX-1 secretory machinery gene transcription. Moreover, disruption of EspN leads to bacterial attenuation in the macrophage. Here, we establish the role of EspN as a critical transcriptional regulator of the ESX-1 system that is required for virulence and regulates gene expression of the *whiB6* transcriptional regulator, a subset of ESX-1 secreted proteins, and ESX-1 secretory machinery.

***On Synergy between Equatorial Convective Disturbances and Monsoon Intraseasonal Oscillations in the Bay of Bengal***

Jaynise M. Pérez Valentín
Department of Engineering
Advisor: Harindra Joe Fernando

The relationship between equatorial convective signals (ECS) and northward propagating Monsoon Intraseasonal Oscillations (MISOs) in the Bay of Bengal (BOB) is studied using observational datasets taken during the MISO-BOB field experiment. Convective envelopes of MISOs arriving from just south of BOB are observed to be originating from strong and weak eastward propagating convective signals along the Equatorial Indian Ocean (EIO) with averaged speeds of ~ 6.4 m/s. Strong equatorial signals contributed to ~20% of the precipitation budget in BOB seemingly spurring convection with northward propagation matching MISOs speeds of 1-2 m/s. In contrast to weak equatorial convective signals which contributed to 14% of the precipitation budget, further dissipating with no apparent northward propagation. Eastward propagating Intraseasonal Oscillations ISOs (with period 30-60 days) and Convectively Coupled Kelvin Waves CCKWs (period 4-15 days) accounted for most of the precipitation variability across the EIO during the 2019 boreal summer as compared to that of 2018, with low variability from the eastward ISO. Agreement can be noted between high moisture content in the mid troposphere and the active phases of CCKWs and ISOs for two locations in the BOB. Basin scale thermodynamic conditions prior to the arrival of strong/weak ECS revealed warmer/cooler SSTs. R/V measurements suggest that evolution of MISOs associated with strong ECS is controlled by the local moisture supply from air-sea interactions in the BOB, enhancing deep convection and thus further moistening the upper troposphere.

## *Hybrid Volitional Control for Lower-Limb Prostheses*

Ryan Posh
Department of Aerospace and Mechanical Engineering
Coadvisors: Dr. James Schmiedeler and Dr. Patrick Wensing

Abstract: The recent advancement of robotic lower-limb prostheses has been a key development in increasing the mobility, efficiency, and quality of life for individuals with amputation. Following these hardware developments, it is critical that researchers continue to improve control strategies that ensure that these prostheses are safe, reliable, stable, and intuitively responsive to the user. One approach to ensure consistently safe and reliable performance for the user is to make robotic prostheses increasingly intelligent and robust. However, this control approach, known as Non-Volitional Control, typically does not allow the user to achieve non-standard movements, such as tip-toe walking or tapping one's foot to music. An alternative approach is to make the prosthesis simply respond to the already highly intelligent agent in the system - the human. Such Volitional Control is intuitive and allows users to engage in a wider range of activities by directly controlling the prosthesis via signals from their own anatomical motor pathways. These controllers, though, are often less robust due to limitations in available sensors and the potential for user fatigue. This work proposes a new Hybrid Volitional Control (HVC) that combines the previous two such that the user may alter or augment the actions of a native non-volitional base controller if desired. HVC is currently being implemented in various treadmill experiments using a robotic ankle prosthesis. Early results suggest that users can achieve natural gait dynamics with low effort and perform energetically challenging activities such as ramp ascent. Users have also demonstrated the ability to stand on tip-toes, navigate upcoming terrain, perform a marching motion, and generally move the ankle as desired. These experiments suggest that HVC could allow users to both reliably achieve a variety of basic tasks and alter device motion to participate in activities that are not currently feasible for individuals with lower-limb amputation.

***Functional groups, determinism and the dynamics of a tropical forest***

Vanessa Rubio
Department of Biological Sciences
Advisor: Nathan Swenson

Unravelling the drivers of forests dynamics is one of the main challenges in tree community ecology. These drivers include niche differentiation, dispersal limitation and stochasticity. Previous work has demonstrated that these mechanisms likely interact such that no one process is responsible for forest dynamics. One possibility is that the functional composition in a forest changes in a deterministic fashion, but the abundances of individual species that are functionally similar and have similar life-history strategies drift in a neutral fashion. This framework aligns with the *functional group*-based version of the neutral theory proposed more than 30 years ago, but it has remained poorly understood and is not well-integrated in tree community ecology. To investigate the possibility that determinism and neutrality may operate on the functional and species levels, respectively, we studied the long-term dynamics of trees on Barro Colorado Island, Panama. Specifically, we sorted tree species into functional groups. We defined them as groups of species that cluster together based upon continuous functional trait measurements that are believed to reflect key life-history trade-offs. This information was then used to quantify the observed species and functional group dynamics in the forest and to compare them to that expected from neutral simulations. We found that forest dynamics are likely governed by deterministic processes at the between-functional group level where species relative abundances change or drift through time within group. Species rank distributions for each functional group remained relatively stable suggesting that these groups may act as broad adaptive zones for both common and rare species that may promote species coexistence. Moreover, we found that these functional groups associate with different habitats in the forest.

***Elucidating the cellular mechanisms involved in breast tumor progression.***

Madison Schmidtmann
Department of Biological Sciences
Advisor: Crislyn D'Souza-Schorey
Coauthors: James Clancy, Shireen Jayman

Mammary ductal carcinoma in situ (DCIS) is a pre-invasive breast cancer that has the potential to develop into invasive ductal carcinoma (IDC). DCIS accounts for about 25% of all breast cancer diagnoses but not all cases will transition to IDC. We do not fully understand how DCIS acquires invasive phenotypes to transition to IDC. Generally, tumor cell invasion involves a large network of signaling pathways resulting in a complex process. Current studies indicated that the activation and high expression of ARF6 protein significantly correlates with the invasion and metastasis of several tumors, including breast cancer. Recent studies from our lab using a DCIS mouse model have demonstrated that affecting ARF6 function greatly impacts DCIS to IDC transition. We now show that dominant negative inhibition as well as knockdown of ARF6 expression markedly impacts metastatic progression with effects on tumor invasion as well as cell survival. Compromised cell survival is linked to cytokinetic defects. Deciphering the molecular basis of these alterations in tumor progression will provide new insights into the cellular events involved in to DCIS to IDC transition in breast cancer and better inform therapeutic strategies.

***Milky Way Substructure: Discovering Dynamical Groups Using Stellar Surveys***

Derek Shank
Department of Physics
Advisor: Timothy C. Beers

The Milky Way provides us with numerous opportunities to examine various astrophysical questions such as Galactic formation scenarios, elemental production methods, and stellar creation pathways, among others. Recently the era of large-data astronomy was ushered in thanks to both ground-based programs such as the Sloan Digital Sky Survey and space-based telescopes such as Gaia. Surveys such as these allow a glimpse into the physical, chemical, and kinematic properties of stars which give a more complete picture of stellar history. Efforts to explore the Milky Way's properties with these parameters have allowed astronomers to classify various structures and elemental patterns located in the Milky Way. While large-scale substructure and elemental enhancements have been well-studied, their origins remain shrouded. The research presented here merges these two questions by studying the smallest building blocks of the Milky Way using the kinematic and chemical properties of stars. The discovery of small kinematic groups has revealed a statistically significant correlation between each star's chemical properties, inconsistent with random chance. This suggests these groups of stars formed in the same birth environment and then subsequently merged into the Milky Way, now scattered about the Galaxy. These groups can be associated with the already known large-scale substructure present in the Milky Way, along with associations to various globular clusters that may have gotten stripped and shed these groups. With a large number of stellar surveys available to get kinematic and chemical information, these newly discovered groups provide valuable insights into the birth environments of stars and the formation of the Milky Way.

***Pregnancy is associated with distinct gut microbiome features in adult female baboons***

Chelsea A. Southworth
Department of Biological Sciences
Advisor: Dr. Elizabeth A. Archie
Coauthors: Mauna Dasari, Jeanne Altmann, Susan C. Alberts, Luis Barriero, Ran Blekhman, Jenny Tung, Elizabeth A. Archie

The mammalian gut microbiome both shapes and is shaped by host behavior, endocrine hormones, immunity, and metabolism. Because all of these traits change substantially with female reproductive phases (e.g., ovarian cycling, pregnancy, and lactation), mammalian gut microbiomes should be an important bellwether of reproductive changes and may mediate reproductive success. Particularly strong changes are hypothesized during pregnancy because of its considerable effects on female immunity, behavior, and energetics. . Gut microbiomes are also highly personalized communities; hence some reproductive changes to the microbiome may be host-specific. Despite these potentially important patterns, research linking the gut microbiome to female reproduction is extremely limited. We propose to address these gaps by tracing longitudinal changes in female gut microbiomes, using 16S rRNA gene sequencing-based microbiome profiles generated from 7,116 fecal samples collected from 184 adult female baboons (*Papio cynocephalus*) in the Amboseli ecosystem of Kenya from 2000-2013. Baboons are ideal for this work because, unlike many mammals, female baboons exhibit easily observable external signs of ovarian cycling, including ovulation, and pregnancy. Using field observations, each sample was classified as either cycling, pregnant, or lactating, and each of these three states was further divided into biologically relevant phases. Here, we present preliminary results from this unprecedentedly large and rich dataset that reveal features of the female baboon gut microbiome that are affected by reproductive state. We report differences in alpha diversity, beta diversity, relative abundances of taxa, and identities of indicator taxa that are connected to reproductive state. We find support for our hypothesis that pregnancy is associated with a distinct gut microbiome compared to cycling and lactation.

***Instrumentation for the Direct Imaging of Exoplanets***

Eckhart Spalding
Department of Physics
Advisor: Jeff Chilcote
Coauthors: Dillon Peng

The direct imaging of exoplanets---planets which orbit other stars---is a promising technique for detecting and characterizing alien worlds which are comparable to, or above, the mass of Jupiter. Over the past few years an instrument called the Gemini Planet Imager has successfully carried out an exoplanet direct imaging survey from the Gemini-South telescope in Chile. The instrument will now be shipped to the University of Notre Dame to be upgraded in a clean room environment in the Nieuwland Hall of Science for a second, and more sensitive, exoplanet survey that will be carried out from the Gemini-North telescope in Hawaii. I will describe the instrument upgrades and their motivations, and tests which we have conducted remotely on the instrument before shipment to Notre Dame. I will also discuss observations taken from the Large Binocular Telescope in Arizona, which includes the University of Notre Dame as a participating institution, to make an imaging search for exoplanets around a nearby star in a unique observing mode which involves the combination of light from two separate telescopes. This observation placed constraints on the existence of companions closer in to the star than any previously-published imaging observation.

***Evaluation of the microbial community and geochemistry in produced water collected from $CO_2$-EOR in the Niagaran Pinnacle Reef***

Brooke Stemple
Department of Civil and Environmental Engineering and Earth Sciences
Advisor: Kyle Bibby
Coauthors: Kara Tinker, Preom Sarkar, Josh Miller, Djuna Gulliver, Kyle Bibby

Geological carbon sequestration (GCS) is the process of capturing $CO_2$ from industrial sources, compressing it into a supercritical fluid ($scCO_2$) and injecting it into geologic repositories such as enhanced oil recovery (EOR) or deep saline aquifers sites for long term storage. Microorganisms are geochemical catalysts for processes that can alter the efficacy of carbon storage, such as biofouling, biomineralization, and biocorrosion. Comprehensive characterization of the biogeochemistry of these complex systems is essential to enhance carbon storage procedures and industrial usage. Our current study investigated the microbial ecology and biogeochemistry of targeted GCS sites in Northern Michigan including produced water collected from 9 $CO_2$-EOR well separators in the Niagaran Pinnacle Reef. We utilized 16S rRNA metagenomic sequencing to characterize the microbial community. We observed a relatively low overall species diversity with the Gram-negative *Proteobacteria* families *Pseudomonadaceae, Oxalobacteraceae,* and *Caulobacteraceae* and the Gram-positive family *Corynecbacteriaceae* being the most abundant taxa across all evaluated samples, representing 55% of the total microbial population. Our geochemical analysis revealed that $CO_2$ accounted for approximately 86% of the gas composition of each well separator followed by $CH_4$ at 13%. The wells had a slightly acidic average pH of 5.1. Major cations and anions were also detected and used to calculate the average overall total dissolved solids which was 260,317.14 mg/L. Our results show dominant taxa within relevant GCS systems and highlight important geochemical factors in subsurface environments exposed to high concentrations of $CO_2$. Our current conclusions suggest that the presence of injected $CO_2$ in EOR reservoirs is likely driving changes in overall subsurface biogeochemistry that include decreasing reservoir pH, that combined with high TDS and salinity, are responsible for the low biomass and microbial abundance observed in this analysis.

***Pictures of Exoplanets around Accelerating Stars:***
***A Direct Imaging Survey with SCExAO/CHARIS***

Taylor Tobin
Department of Physics
Advisor: Jeffrey Chilcote
Coauthors: Thayne Currie[1,2,3], Julien Lozi[1], Timothy Brandt[4], Olivier Guyon[1,5,6,7], Tyler Groff[8], Masuyaki Kuzuhara[5], Taichi Uyama[9,10,11], Motohide Tamura[5,11,12], Jeremy Kasdin[13]

[1]Subaru Telescope, National Astronomical Observatory of Japan, Hilo, HI, USA
[2]NASA-Ames Research Center, Moffett Field, CA, USA
[3]Eureka Scientific, Oakland, CA, USA
[4]Department of Physics, University of California Santa Barbara, Santa Barbara, CA, USA
[5]Astrobiology Center of NINS, Mitaka, Tokyo, Japan
[6]Steward Observatory, The University of Arizona, Tucson, AZ, USA
[7]College of Optical Sciences, University of Arizona, Tucson, AZ, USA
[8]NASA-Goddard Space Flight Center, Greenbelt, MD, USA
[9]Infrared Processing and Analysis Center, California Institute of Technology, Pasadena, CA, USA
[10]NASA Exoplanet Science Institute, Pasadena, CA, USA
[11]National Astronomical Observatory of Japan, Mitaka, Tokyo, Japan
[12]Department of Astronomy, Graduate School of Science, The University of Tokyo, Bunkyo-ku, Tokyo, Japan
[13]Department of Astrophysical Science, Princeton University, Princeton, NJ, USA

Despite its conceptual simplicity, imaging exoplanets directly is one of the most technologically difficult methods of exoplanet detection. The facilities and instruments used for direct imaging have improved dramatically in recent years, increasing the number of imageable exoplanets by 200% over the last decade. However, even with technological improvements, stellar systems hosting imageable exoplanets are rare, leading to low yields of new exoplanet discoveries in blind imaging surveys. And while directly imaged planets are uniquely positioned to provide observational insight into planet formation models, only about a dozen extrasolar planets have been imaged to date. To amass a sufficient sample of imageable exoplanets for more robust studies of their atmospheric chemistry, formation, and evolution, targeted surveys are needed.In this talk, I will discuss progress from the ongoing direct imaging survey with SCExAO/CHARIS targeting stellar systems displaying astrometric evidence of possible companions, as well as progress on CHARIS's new automated data processing backend, ADEPTS.

## *Visual Impairment in* norpA-*Mutated* Aedes *Mosquitoes*

Alexis Waldschmidt
Department of Biological Sciences
Advisor: Joseph O'Tousa
Coauthors: Matthew Gregory, Brian Dineen, Alex Dittmar, Jeremy Sutterer, Elaine Teeters, Michelle A. Whaley, and Joseph E. O'Tousa

The *Aedes aegypti* β-class phospholipase C (PLCβ, ID=AAEL009380) is the ortholog of the *Drosophila* NORPA PLCβ essential to the signal transduction process responsible for light detection. To analyze the mechanisms underlying *Aedes* photoreceptor adaptations and their visual behaviors, we used the clustered regularly interspaced short palindromic repeats (CRISPR)-Cas9 system to mutate the *Aedes aegypti* PLCβ gene. Mutant animals were initially identified using PCR and capillary electrophoresis, and then breeding schemes were used to create homozygous mutant animals. Here we describe the *Aedes* mutant allele designated as $norpA^{CAT}$. This $norpA^{CAT}$ allele shows three alterations in the targeted region of the gene: (1) a 2 bp deletion in the promoter region, (2) a 6 bp deletion within the coding region, and (3) a nonsense point mutation immediately upstream of the 6 bp deletion. Histological and western blot results revealed that PLCβ protein in the $norpA^{CAT}$ mutant was below the detection limit. Electroretinography (ERG) analysis showed that the light-evoked electrical response of the $norpA^{CAT}$ retina was slow and degraded relative to the wild type control retina. In a behavioral assay designed to detect both a light-triggered startle response and light avoidance behavior, the $norpA^{CAT}$ mutant showed marked deficits relative to the wild type control. These results demonstrate that the $norpA^{CAT}$ mutant has vastly reduced levels of PLCβ protein and has pronounced deficiencies in light-driven behavioral responses.

**Delineating the role of *gldc* in nephron patterning during kidney development**

Nicole Weaver
Department of Biological Sciences
Advisor: Dr. Rebecca Wingert

Glycine is an amino acid that is vital to the proper functioning of the body, but its roles during ontogeny are not well understood. Glycine levels are regulated through the glycine cleavage system (GCS), a molecular machine that produces one-carbon units for later metabolism and folate production. Elevated glycine levels due to congenital mutations in GCS components, such as *glycine dehydrogenase* (*gldc*), cause human birth defects and the rare disease nonketotic hyperglycinemia (NKH). NKH patients suffer from pleiotropic symptoms including seizures, mental retardation, and early death; therefore, it is imperative to elucidate the downstream pathological mechanisms of glycine accumulation. Our lab, and others, have reported *gldc* deficiency and glycine addition cause developmental changes in various tissues. Here, we interrogate the role of *gldc* in renal ontogeny. Whole mount *in situ* hybridization (WISH) revealed that *gldc* transcripts were highly expressed in multiple tissues, including the central nervous system and embryonic kidney, recapitulating mouse and human expression studies. We found formation of nephron cell populations in the kidney was disrupted in *gldc* deficient and glycine treated embryos, where the distal early (DE) segment was increased at the expense of the distal late (DL) segment. These alterations led us to hypothesize that *gldc* is essential for distal segment patterning, a process that is regulated by interplay between the *Iroquois* (*irx*) transcription factors and *transcription factor AP-2a* (*tfap2a*). To test this hypothesis, we have begun to analyze spatiotemporal expression domains of *irx3b* and *irx1a*, which are conserved regulators of intermediate and distal segment development. Using WISH, we found that *gldc* deficient and glycine treated animals exhibited an increase in the expression domain of *irx1a*, coinciding with the changes in DE specific solute transporter gene expression patterns. Taken together, these studies indicate that *gldc* has essential roles in regulating segment pattern during distal nephron development.

***Efficient Robust Path Planning Under Temporal Specifications for Mobile and Aerial Robots***

Shirantha Welikala
Department of Electrical Engineering
Advisors: Prof. Hai Lin and Prof. Panos J. Antsaklis

**Abstract:** In robotics applications, *path planning* is a primitive, yet crucial problem faced by control engineers. In particular, the objective of a path planning problem is to determine a feasible trajectory (a feasible sequence of control inputs) for a robotic system of interest (e.g., a mobile or an aerial robot) under a given set of *specifications*. Often these specifications are *temporal* as they indicate requirements such as reaching/avoiding specific regions (in the state/control space) over specific time intervals. In the recent literature, to symbolize such temporal specifications, Signal Temporal Logic (STL) is increasingly used as it offers a simple set of *semantic rules* to evaluate the degree of satisfaction of the temporal specifications as a scalar *robustness measure*. In essence, STL enables transforming a path planning under temporal specifications problem into an *optimization problem* that optimizes the corresponding robustness measure over all possible sequences of control inputs applicable to the considered robotic system. However, this robustness measure is non-smooth and non-convex, and thus, its optimization calls for computationally inefficient mixed-integer optimization techniques that do not scale well. Consequently, the recent literature has focused on using *smooth approximations* of the robustness measure (known as smooth robustness measures (SRMs)) to enable the application of computationally efficient and scalable gradient-based optimization techniques. In this research, we first generalize two recently proposed SRMs and two novel SRMs and review their strengths/weaknesses. Next, we propose an approach to characterize the *approximation error* associated with each SRM in terms of its inherent tunable parameters. This provides a systematic approach to select an SRM (to optimize) and its parameters. We then propose a technique to evaluate *explicit gradients* of SRMs leading to improve computational efficiency and accuracy compared to using numerically estimated gradients. Finally, to highlight our contributions, we provide an open-source software toolbox with extensive simulation results.

***Esrrγa regulates nephron development and ciliogenesis by controlling prostaglandin synthesis and cooperation with Ppargc1a***

Hannah M. Wesselman
Department of Biological Sciences
Advisor: Dr. Rebecca A. Wingert
Co-authors: Anna L. Flores-Mireles

Cilia are essential for the ontogeny and function of many tissues, including the kidney. In mammals, Esrrγ has been previously established as a significant determinant of renal health, with decreased expression linked to age related dysfunction, cyst formation, and kidney disease. Here, we report that the Esrrγ vertebrate ortholog *estrogen related receptor gamma a* (*esrrγa*) is essential for proper cell fate choice within kidney functional units (nephrons) as well as ciliogenesis. Deficiency of *esrrγa* resulted in nephrons with alterations in proximodistal segmentation and a decreased multiciliated epithelial cell populace. Surprisingly, *esrrγa* deficiency disrupted renal ciliogenesis and caused a similar abrogation within the developing node and otic vesicle—all defects that occurred independently of changes in cell polarity or basal body organization. These phenotypes were consistent with interruptions in prostaglandin signaling, and we found that ciliogenesis was rescued in *esrrγa* deficient embryos with exogenous $PGE_2$ or through overexpression of the cyclooxygenase enzyme Ptgs1. Through genetic interaction studies, we found that peroxisome *proliferator-activated receptor gamma, coactivator 1 alpha* (*ppargc1a*), which acts upstream of Ptgs1-mediated prostaglandin synthesis, has a synergistic relationship with *esrrγa* in the ciliogenic pathway. These data position *esrrγa* as a novel link between ciliogenesis and nephrogenesis through regulation of prostaglandin signaling and cooperation with *ppargc1a*, and highlight *esrrγa* as a potential new target for future ciliopathic treatments.

# *Abstracts for Poster Presentations*

***CLIPR-76, a novel CLIPR protein linking membranes and microtubules***

Annamarie T. Bryant
Department of Chemistry and Biochemistry, Integrated Biomedical Sciences Program
Advisor: Holly V. Goodson
Coauthors: Jill Voreis, Gergana Ugrinova, Claire Whalen

In eukaryotes, the microtubule (MT) cytoskeleton contributes to a number of essential cellular functions and processes including cellular organization, division, motility, and intracellular trafficking. A number of proteins help regulate MT dynamics and the interactions of MTs with other cellular components, such as the Golgi complex. A subset of these regulatory proteins are defined by the presence of a conserved CAP-Gly domain and are referred to as "CLIPRs" due to their relation to the cytoplasmic linker protein CLIP-170. We report here the characterization of CLIPR-76, a novel CAP-Gly containing protein related to CLIP-170. The CLIPR-76 gene (CLIP4) undergoes alternative splicing to yield at least three different splice isoforms in humans. RT PCR shows CLIP4 expression is highest in striated muscle and Western blot analysis with murine myoblast cells (C2C12) show CLIPR-76 expression is induced upon differentiation. All isoforms contain N-terminal ankyrin repeats followed by one, two, or three CAP-Gly domains. Two CLIPR-76 isoforms (CLIPR-76_1 and CLIPR-76_3) localize to MTs and alter MT organization upon overexpression. The third isoform, CLIPR-76_4, localizes to the endoplasmic reticulum (ER) via its hydrophobic C-terminus and shares some sequence similarity to its relative CLIPR-59, which localizes to the Golgi apparatus. Overexpression of CLIPR-76_4 strongly alters ER and ERGIC morphologies and inhibits ER to Golgi transport, indicating that CLIPR-76_4 may play a role in early vesicular transport. We are currently investigating how the expression of specific CLIPR-76 isoforms varies by tissue type and developmental stage and the changes in localization of CLIPR-76 throughout development.

***Variational Inference Via Normalizing Flows and Adaptive Annealing***

Emma R. Cobian
Department of Applied and Computational Mathematics and Statistics
Advisor: Jonathan D. Hauenstein
Coauthors: Fang Liu, Daniele E. Schiavazzi

Normalizing flows are invertible mappings used to transform simpler probability densities into ones that are more complex. Through optimizing parameters associated with these mappings, normalizing flows are used in statistics and machine learning for density estimation and variational inference. In the framework of variational inference, to improve the efficiency and accuracy of the approximation via normalizing flows when a target distribution is multimodal, annealing of the target distribution with a constant annealing schedule in the optimization is often employed. On the other hand, a constant annealing schedule often applies slowly-changing temperatures to the target distribution and can be inefficient computationally. In this presentation, we will introduce a more efficient adaptive annealing schedule that automatically adjusts the incremental step in the annealing schedule to the KL-divergence between two adjacent tempered distributions. We will demonstrate the computational efficiency of normalizing flows, combined with our proposed adaptive annealing scheme, in approximating multimodal distribution and obtaining Bayesian inferences of the parameter for dynamical systems.

**Proteo-genetic analysis of ESX-1 secretion reveals distinct roles for ESX-1 substrates in *Mycobacterium marinum***

Rachel Cronin
Department of Biological Sciences
Advisor: Dr. Patricia Champion
Coauthors: Micah J. Ferrell, Clare W. Cahir, Matthew M. Champion, and Patricia A. Champion

Pathogenic mycobacteria, including *Mycobacterium tuberculosis,* require the ESX-1 secretion system for survival within the host. ESX-1 actively transports protein substrates that are required for lysing the phagosomal membrane. ESX-1-dependent phagosomal lysis is essential for bacterial survival; mycobacteria lacking an ESX-1 system are retained in the phagosome and attenuated. Although ESX-1 substrates are required for pathogenesis, these substrates are largely uncharacterized. Using *Mycobacterium marinum,* an established model of *M. tuberculosis*, we sought to characterize the role of these substrates in ESX-1 secretion and virulence. We generated a collection of *M. marinum* strains with single deletions of all known ESX-1 substrate genes, along with the relevant complementation strains. With this collection, we characterized the contribution of each substrate to hemolysis and virulence. We demonstrated that the loss of each individual substrate has differential impacts on ESX-1 function as measured by virulence in a macrophage model of infection and hemolytic activity. We quantified changes to the secreted proteome in each substrate deletion strain as compared to the WT and corresponding complementation strain. We found that each protein substrate differentially impacts the secreted proteome of *M. marinum.* Moreover, the substrates impact secretion differently from deletion of the ESX-1 secretory machinery. We used statistical analyses to identify distinct substrate groups that had similar secretion profiles, potentially indicating shared or redundant function. Lastly, we identified potential new ESX-1 substrates whose secretion profiles correlate with known substrates. Overall, our study provides a comprehensive understanding of how ESX-1 substrates differentially impact ESX-1 function and secretion in *M. marinum.*

*Penicillin-Binding Protein 2x as a Target in Allosteric Inhibition by Antibiotics*

Homero I. Domínguez
Department of Chemistry and Biochemistry
Advisor: Professor Shahriar Mobashery
Coauthors: Mohini-Mohan Konai, Jeshina Janardhanan, Neha Rana, Kiran Mahasenan, Rhona Feltzer, Jed Fisher, Shahriar Mobashery

Abstract: The penicillin-binding protein 2x (PBP2x) of *Streptococcus pneumoniae* is an essential enzyme that catalyzes the cross-linking of the bacterial cell wall, a structure whose integrity is critical for the survival of bacteria. This enzyme is a target for β-lactam antibiotics. Mutations in the *pbp2x* gene resulting in amino-acid substitutions at or near the active site of PBP2x constitutes the primary means of resistance to β-lactam antibiotics.[1] Virtually all known antibiotics bind the active sites of their respective target enzymes. Activities of some enzymes/proteins are regulated at sites outside of the active site, referred to as the allosteric site. The work disclosed herein represents a rational medicinal chemistry approach aiming to target an allosteric site known as the Penicillin-Binding Protein and Serine/Threonine Kinase Associated (PASTA) domain of PBP2x.[2] Using high-throughput computational screening, one hit molecule was found to bind to the allosteric site at the PASTA domain. We have documented that this molecule exhibits antibacterial activity. The goal of this work is to pursue the lead optimization of the hit compound by improving its antibacterial activity in search of novel PBP2x inhibitors targeting the PASTA domain.

[1]Gordon *et al*., *J. Mol. Bio*. **2000**, *299*, 477-485.
[2]Bernardo-Garcia *et al*., *ACS Chem. Bio*. **2018**, *13*, 694-702.

***Late-stage Modification and Biological Evaluation of Cruentaren A Derivatives***

Xiaozheng Dou
Department of Chemistry & Biochemistry
Advisor: Brian Blagg
Coauthors: Chitra Subramanian, Hui Guo, Bhargav Patel, Terin D'Amico, John Rubinstein, Mark Cohen

The 90-kDa heat shock protein (Hsp90) is a molecular chaperone that plays essential roles for the folding, stabilization, activation, and degradation of over 400 client proteins, many of which are directly associated with cancer progression. Consequently, inhibition of the Hsp90 protein folding machinery has been an attractive anticancer target since its inhibitor results in a combinatorial attack on numerous oncogenic pathways. Unfortunately, 18 Hsp90 *pan*-inhibitors entered clinical trials and failed to reach FDA approval for use as a cancer monotherapy due to off- and on-target toxicities. The discovery and development of isoform-selective Hsp90 inhibitors is regarded as a promising approach to achieve the desired anticancer effect without the harmful side effects observed with Hsp90 *pan*-inhibitors. Cruentaren A, a potent cytotoxic natural product isolated from myxobacterium, selectively inhibits F1Fo ATP synthase, which disrupts its interactions only with Hsp90α isoform and results in Hsp90 client protein degradation without induction of the pro-survival heat shock response. Cruentaren A also exhibits sub-nanomolar activity against multiple human cancer cell lines, while manifesting >500 nM activity against normal cell lines, providing a large therapeutic window for drug development. However, its clinical development has been hampered by limited structure-activity relationship studies. Recently, we obtained the first cryo-EM structure of cruentaren A bound to $F_1F_o$ ATP synthase, which allows for the rational design of improved inhibitors. Herein we report the current progress towards the late-stage modification and biological evaluation of cruentaren A analogs.

***Chemically Resilient Nanofiltration Membranes Fabricated from Copolymers for Organic Solvent Nanofiltration***

Michael Dugas
Department of Chemical and Biomolecular Engineering
Advisor: Dr. William A. Phillip
Coauthors: Shukun Zhong, Bumjun Park, Jennifer Schaefer

As membrane technologies become more critical to meeting the growing demand for energy-efficient organic solvent separations, a need has emerged for membrane platforms that can be tailored to accommodate the highly varied applications, feed compositions, and treatment demands for applications such as catalyst recovery and pharmaceutical product purification. Nanofiltration (NF) membranes based on copolymer materials are a promising separations platform because they can be engineered at the molecular scale to address an array of process needs. In addition, these copolymer materials can be reacted post-casting with functionalities that can crosslink and create an electrostatic charge, thereby creating a durable, more effective membrane. Here, a resilient NF membrane is developed through the molecular design of a poly(trifluoroethyl methacrylate-co-oligo(ethylene glycol) methyl ether methacrylate-co-glycidyl methacrylate) [P(TFEMA-OEGMA-GMA)] copolymer that can be either dip-coated onto hollow fiber supports or blade casted onto flat sheet supports that has a pore diameter of approximately 2nm in water. The epoxide rings on the GMA repeat units were functionalized post-coating with diamines of various chains lengths to incorporate moieties along the pore walls that crosslink the overall polymeric structure. By varying the chain length, the extent of crosslinking is affected. This extent of crosslinking has shown, through permeability and cycling experiments, SAXS analysis, and poly(propylene glycol) rejections, to affect the pore diameter of the membrane in various organic solvent environments, including THF, DMF, and ethanol without damaging the structure of the membrane itself. Dye rejection experiments demonstrate an absorptive characteristic of the membrane. These results demonstrate that by combining the versatility of molecularly designed copolymers with functionalities that can be utilized to increase the integrity of the membrane, copolymeric NF membranes can further the use of membrane technologies into emerging applications.

***Establishing the binding mode of a diphenylfuran to an RNA triple helix***

Madeline M. Glennon[1]
[1]Department of Chemistry and Biochemistry, University of Notre Dame, Notre Dame, IN, 46556
[2]Department of Chemistry, Duke University, Durham, NC, 27708
Advisor: Jessica A. Brown
Coauthors: Krishna M. Shivakumar[1], Charlotte N. Kunkler[1], Amanda E. Hargrove[2]

Metastasis-associated lung adenocarcinoma transcript 1 (MALAT1) is a long non-coding RNA whose 3′-end terminates with a triple helix. This triple-helical segment increases the stability of MALAT1, allowing the long non-coding RNA to accumulate in multiple types of cancer. Moreover, MALAT1 promotes metastasis, making it an attractive therapeutic target. Small molecules have been shown to target RNA and alter their stability and overall function. Diphenylfuran (DPFp8, DPFp9, and DPFp10) small molecules are known to bind to the MALAT1 triple helix, however the binding mode of these molecules has not been elucidated. Cryogenic electron microscopy (cryo-EM) and x-ray crystallography are being employed to solve high-resolution 3-dimensional (3D) structures of the MALAT1 triple helix in complex with DPFp small molecules. Single particles of a modified MALAT1 RNA triple helix were visualized using cryo-EM and the resulting 3D model is at a working resolution of ~7 Å. Additionally, a MALAT1 triple helix variant has been successfully crystallized in complex with DPFp10. Crystallization conditions are being optimized and crystals will be screened for diffraction. Solving 3D structures will reveal the binding mode of the DPF small molecules to the MALAT1 triple helix and will enable the rational design of the DPF scaffold to increase specificity and selectivity.

***Multiple Factor Analysis Reveals Variable Interactions Between the Diet and Mycobiome of Long-tailed Macaques (Macaca fascicularis) on Two Islands in Southeast Asia***

Benjamin J. Gombash
Department of Biological Sciences
Advisor: Dr. Hope Hollocher
Coauthors: Amanda Acevedo, Chissa Rivaldi, Carson Smith

The mycobiome is the group of fungal taxa that are detected in samples from other organisms. Research focused on variation in the mycobiome has become popular recently, with some of that focus being on wildlife mycobiomes. When variation is detected in the mycobiome of wildlife, it is frequently ascribed to variation in the host's diet. While the host's diet is often implicated, the diet itself is rarely assessed alongside the mycobiome. We used the V9 hypervariable region of the 18S ssu rRNA amplicon to assess the diet and mycobiome of 127 fecal samples from long-tailed macaques (*Macaca fascicularis*) on the islands of Singapore and Bali, Indonesia. Linear regressions and partial Mantel tests confirm that there are interactions between the diet and mycobiome occurring on both islands. The fungal and dietary taxa were categorized into several relevant groups (e.g., plant pathogens, animal pathogens, and saprotrophs for fungi, crop plants, non-crop plants, and anti-fungal plants for diet items) to run Multiple Factor Analyses to search for more nuanced interactions between groups of dietary and mycobiome taxa. Although dietary taxa are more numerous on both islands, taxa in mycobiome groups are the most important for explaining variation in the results of the Multiple Factor Analyses. Despite sharing ~70% of their detected taxa, the interactions between dietary and mycobiome groups on the two islands are distinct. These results confirm that the diet and mycobiome do interact in long-tailed macaques. However, these interactions are not necessarily consistent between different populations of the same host organism. Future work should incorporate other factors that are important for host organisms, such as environmental factors, which may interact with the diet, the mycobiome, or both groups of taxa.

***Recent advances in deployability of paper-based field test for illicit drugs***

Kathleen Hayes
Department of Chemistry and Biochemistry
Advisor: Marya Lieberman
Coauthors: Heather Whitehead

Interest in presumptive field tests for illicit drug detection has increased in recent years as deaths from opioid overdoses have risen. Previously, a microfluidic paper analytical device for detecting (idPAD) was described that sensitively and specifically detects cocaine, crack cocaine, heroine, and methamphetamine by using a library of 12 colorimetric lane tests which detect functional groups found in illicit drugs and their cutting agents. Each drug elicits a unique color "barcode" which users match to standard barcodes for sample identification. Since its original publication, we have redesigned the idPAD to increase field-friendliness and deployability. One important modification is the introduction of a sample collection area. This collection area is outside of the colorimetric lane tests and protected from water exposure, so the sample can be stored and saved for downstream applications such as LC-MS/MS analysis. This collection area allows for drug samples to be transported more conveniently and safely than before. Average recovery from the collection area is 34% of the applied drug, and is stable for storage for up to 6 weeks. Analysis of idPADs has historically relied on users to match the color barcode with its corresponding drug. To eliminate the need for human judgement and increase efficiency, we are training a neural network to read barcodes and report drug identity. Another potential application of the idPAD for detecting hormones used for birth control, abortions, and hormone replacement therapy is being explored as a screening tool for social justice groups interested in ensuring quality of dosage forms in settings where access to such drugs is limited.

***Silicon and Zirconium Ceramic Material Irradiation in the Presence of Water***

Hanna Hlushko
Radiation Laboratory
Advisor: Jay A. LaVerne
Coauthors: Patricia L. Huestis

Water molecules decompose under ionizing radiation, producing free radicals that further can combine into reactive species such as hydrogen, oxygen, and corrosive hydrogen peroxide. Water radiolysis occurs in nuclear reactors and nuclear waste storage, where products of radiolysis can cause degradation of the equipment and safety concerns. For this reason, significant research effort is focused on the exploration of the water radiolysis mechanisms. In particular, it was previously shown that the presence of solid surfaces can affect water radiolysis. The yields of the radiolysis products can change due to the transfer of energy or matter through the interface from solid to water. For zirconium oxide, it was shown, that the presence of hydroxyl groups at the surface facilitates adsorption of water molecules and increases hydrogen production under irradiation. In this work, we focus on other zirconium ceramics, including zirconium nitride and carbide, as well as silicon nitrides. Exploration of water radiolysis on the surface of these ceramic materials is important for understanding if they are reliable for use in cladding in nuclear reactors. Additionally, it will help to further understand the role of surface chemistry on water radiolysis. The effect of the surface of materials on water radiolysis is studied through evaluation of hydrogen production by slurries of these ceramics with water, as well as by ceramics containing only adsorbed water. Surface area, surface chemistry, and composition are characterized prior to and after irradiation. The effect of the surface chemistry and the ability to adsorb water on hydrogen production will be discussed.

***Strong clustering of asymptomatic Plasmodium falciparum and Plasmodium vivax infections among ethnic groups but absence of population structure in Chittagong Hill Tracts, Bangladesh***

Tiffany Huwe
Department of Biological Sciences
Advisor: Dr. Cristian Koepfli
Coauthors: Mohammad Golam Kibria, Fatema Tuj Johora, Ching Swe Phru, Nusrat Jahan, Mohammad Sharif Hossain, Wasif Ali Khan , Ric N Price, Benedikt Ley, Mohammad Shafiul Alam, Cristian Koepfli

Malaria remains endemic in eastern Bangladesh, with most cases in the forested, mountainous southeastern region called the Chittagong Hill Tracts (CHT). This area is home to Bengali settlers and diverse groups of indigenous people (Pahari) residing largely in mono-ethnic villages. We purposively selected 10 prominent ethnicities of the CHT and randomly selected afebrile individuals from each group. All 1002 enrolled participants were screened for malaria by RDT and qPCR. The prevalence of *Plasmodium falciparum* and *P. vivax* infection was 0.7% by RDT (*Pf*: 6/1002; *Pv*: 0/1002, mixed: 1/1002) and 4% by qPCR (*Pf*: 21/1002; *Pv*: 16/1002, mixed: 5/1002). Infections were highly clustered, with 63.8% (30/47) occurring in only two groups, the Khumi and Mro. To investigate whether transmission occurs primarily within groups, parasites were genotyped. Twenty *P. falciparum* isolates were typed by *msp2* and deep sequencing of 5 amplicons (*ama1-D3, cpmp, cpp, csp*, and *msp7*), and 21 *P. vivax* infections by microsatellite typing of ten loci and amplicon sequencing of *msp1*. Diversity was high with 5-15 alleles per marker. Expected heterozygosity was 0.93 for *P. falciparum* and 0.81 for *P. vivax*. In total 85.7% (18/21) of *P. vivax* and 25% (5/20) of *P. falciparum* infections were polyclonal. No population structure was evident for either species, suggesting high transmission and gene flow among groups; however, pairwise relatedness of infections measured by identity-by-state (IBS) was higher among the Mro than other groups. IBS for both *Plasmodium spp*. was greater among samples from the Mro (*Pf*: 0.50; *Pv*: 0.31) than among all samples (*Pf*: 0.11; *Pv*: 0.15). This suggests that the Mro may be isolated from transmission from other groups, perhaps due to geographic distance or some Mro-specific behavior. High subclinical infection prevalence and high genetic diversity suggest sustained transmission in CHT. Control activities should be specifically directed to groups at high risk.

## *A Conserved N-Acetyltransferase is Responsible for N-terminal Acetylation of EsxA*

Owen Collars, Bradley Jones
Department of Biological Sciences
Advisor: Dr. Patricia Champion
Coauthors: Micah J. Ferrell

N-terminal protein acetylation is a ubiquitous post-translational modification. In eukaryotes, this modification is mediated by 7 N-acetyl-transferases (NATs) and regulates protein stability to gene expression. N-terminal acetylation and the NATs that mediate it remain poorly understood in prokaryotes. We set out to characterize this modification in the ESX-1 secretion system, a conserved virulence pathway among mycobacteria. One key substrate of the ESX-1 secretion system is EsxA, which has been previously shown to be N-terminally acetylated. However, the NAT mediating its acetylation and its contribution to virulence have not yet been uncovered. We have identified 23 N-terminal acetyltransferases (NATs) conserved between the human pathogen Mycobacterium *tuberculosis* and it's ortholog, Mycobacterium *marinum*. Because of the high number of NATs and the relatively low number of N-terminally acetylated proteins in these bacteria, we hypothesized that one of these NATs could be responsible for the N-terminal acetylation of EsxA. A mutant library of the most highly conserved NATs has been created and screened for changes in EsxA acetylation and subsequent alterations to virulence. We verified the specificity of an antibody against acetylated EsxA N-termini via Dot Blot and NUT-PAGE analyses. We performed western blots on strains containing deletions of the five most conserved NATs, looking for changes in EsxA acetylation. A deletion of *MMAR_1839* revealed a loss of acetylated EsxA, while retaining the protein. Other NAT deletion mutants did not share this phenotype, indicating that *MMAR_1839* is specifically responsible for EsxA acetylation.

## *A Single Modified Nucleoside Destabilizes an RNA•DNA-DNA Triple Helix*

Charlotte N. Kunkler
Department of Chemistry and Biochemistry
Advisor: Jessica A. Brown
Coauthors: Grace E. Schiefelbein, Nathan J. O'Leary

Recent studies suggest noncoding RNAs interact with genomic DNA, forming an RNA•DNA–DNA triple helix that regulates gene expression. In fact, at least one RNA•DNA–DNA triple helix is predicted in the promoter regions of most human genes, suggesting RNA•DNA–DNA triple helices are a common mechanism for regulating transcription. One mechanism that cells could employ to regulate the formation of these triple helices is through the use of RNA modifications. With over 143 known RNA modifications, it is likely that some modifications stabilize RNA•DNA–DNA triple helices, while other modifications destabilize them. Here, we focus on a pyrimidine-motif triple helix, which is stabilized by the canonical U•A–T and C•G–C base triples. We hypothesize that RNA modifications affect the stability of RNA•DNA–DNA triple helices. Therefore, we employed microscale thermophoresis (MST) to examine how eleven different RNA modifications at a single position in an RNA•DNA–DNA triple helix affects its stability: 5-methylcytidine ($m^5C$), 5-methyluridine ($m^5U$), 3-methyluridine ($m^3U$), pseudouridine (Ψ), 4-thiouridine ($s^4U$), $N^6$-methyladenosine ($m^6A$), inosine (I), and each nucleobase with 2'-*O*-methylation (Nm). Compared to the unmodified U•A–T base triple, some modifications (Um•A–T and Ψ•A–T) have no observable change in stability, some modifications have minor decreases in stability ($m^5U$•A–T at ~2-fold weaker), and some modifications completely disrupt triple helix formation ($m^3U$•A–T and $s^4U$•A–T). These results suggest that modifying RNA may be a mechanism to regulate cellular RNA•DNA–DNA triple helices. Future studies will focus on computationally searching for biological examples of RNA•DNA–DNA triple helices whose formation could be controlled by modified RNA. In summary, RNA modifications play an important role in the stability of RNA structures, emphasizing the importance of examining the modification states of cellular RNAs and to experimentally validate predicted RNA•DNA–DNA triple helices, especially when the RNA contains modifications.

***Development and Validation of an Alaska Coastal Ocean Forecast System with Coupled Tides, Storm Surge and Waves under Sea Ice Conditions***

Guoming Ling
Department of Civil and Environmental Engineering and Earth Sciences,
University of Notre Dame, Notre Dame, IN
Advisors: Joannes J. Westerink and Damrongsak Wirasaet
Coauthor: Mindo Choi

**Abstract**: Western Alaska experiences regular storm surge events. These storms can be especially severe during winter with the presence of sea ice in both the Chukchi and Bering Seas. Existing storm surge models rarely consider sea ice effects together with wave effects. Therefore, in this study, we present an ALaska Coastal Ocean Forecast System (ALCOFS) which considers sea ice and wave effects in a tightly coupled SWAN (wind wave model governed by spectrum action balance equation) + ADCIRC (storm surge model based on shallow water equations) model for real time storm surge forecasting. The sea ice effect is included by incorporating a parameterization of air-sea-ice drag in the ADCIRC, and the wave energy dissipation by sea ice is considered in SWAN. The model utilizes an unstructured mesh with variable resolution (ranging from 30km to 70m) to achieve accurate predictions and fast run times. The model was exercised carefully with tidal tests to obtain good quality of tidal results and the final optimized parameter setups. The impact of sea ice and waves was examined with several storm surge events. Furthermore, an efficient real time continuous storm tide, wave and surge forecasting scheme which performs a cycle with a one day nowcast and then a five day forecast is proposed. ALCOFS has been running as a preliminary operational model for more than one year continuously and stably. The real time daily forecast results are posted on the GitHub page https://gm-ling.github.io/ALCOFS-R/. To examine the effectiveness and accuracy of the forecasting system, the recorded forecast results for the past one year period are validated with observations from NOAA stations. This study underscores the importance of incorporating sea ice and wave effects into simulations of storm surges for the area with sea ice conditions, and presents the skill of the forecasting system.

***A Survey of Sportfish for Per- and Polyfluoroalkyl Substances (PFAS) – An Emerging Contaminant in the Great Lakes***

Daniele de Almeida Miranda
Departments of Physics and Biological Sciences | Environmental Change Initiative
Advisors: Graham Peaslee and Gary Lamberti
Coauthors: Alison Zachritz, Whitney Conard, Heather Whitehead, Meghanne Tighe, and Sarah Klepinger

Abstract: Per- and polyfluoroalkyl substances (PFAS) comprise a class of man-made chemicals largely used in a wide variety of industrial processes. PFAS have unique chemical properties such as resistance to degradation and both lipophobicity and hydrophobicity, which contributes to their global occurrence, environmental persistence, and toxicity to humans and wildlife. PFAS can be released into the environment from either direct use or through the transformation of PFAS precursors. Once in the environment, those compounds can bioaccumulate in the biota and biomagnify in the food web. Currently, classical PFAS analysis techniques by liquid chromatography tandem mass spectrometry (LC-MS/MS) can identify less than 100 PFAS compounds of the thousands in use, which limits information regarding their overall occurrence and impacts in the environment. Our goal is to optimize analytical techniques to assess PFAS contamination in sportfish from the Great Lakes through a total fluorine approach using a low-cost technique of Particle Induced Gamma-ray Emission (PIGE), and then identify the specific PFAS compounds with LC-MS/MS. By analyzing 700 Great Lakes fish of different species and trophic levels, we can evaluate dietary routes for PFAS exposure from prey to predator fish using isotopes signature (i.e., $\delta^{15}N$ and $\delta^{13}C$) along with PFAS speciation. Furthermore, we are assessing the relationship between total fluorine and specific PFAS concentrations to determine the full extent of the PFAS problem by comparing PFAS concentrations in fish from different areas in the Great Lakes, where PFAS are known to be a problem to putative reference sites. This study represents the first survey of Great Lakes sportfish assessing total fluorine, thereby providing an ecological and food security perspective.

## Automated Diafiltration Experiments Advance Material and Process Development

Jonathan Aubuchon Ouimet
Department of Chemical and Biomolecular Engineering
Advisor: William A. Phillip
Coauthors: Xinhong Liu, Zachary Muetzel, Elvis Eugene, Alexander Dowling, William A. Phillip

The improved characterization techniques that will advance membrane processes need to reduce the resources necessary to characterize membranes, address the knowledge gaps related to the interfacial processes that govern solute-solute selectivity and capture the performance of membranes in complex multi-component feed streams. Within this study, guided by the tools of data science, a diafiltration apparatus is developed to inform material and process design by rapidly characterizing membrane performance over a broad range of feed solution compositions. By systematically dosing a high concentration diafiltrate into the stirred cell, a predetermined change in the retentate concentration can be achieved. Within a 5 mM to 80 mM KCl phase space, one diafiltration experiment (3.5 hours) can probe membrane parameters 12 times faster than a campaign of filtration experiments (43.5 hours). Additionally, the interplay between data analytics and instrumentation led to the incorporation of an inline conductivity probe that monitored the real time retentate concentration. In turn, this information provides crucial insight into distinguishing the mechanisms of transport.

## *Upgrading the Gemini Planet Imager: Testing and Installation of New Cameras for GPI 2.0*

Dillon Peng
Department of Physics
Advisor: Jeffery Chilcote

The Gemini Planet Imager (GPI) is an extreme adaptive optics, high-contrast instrument designed for the direct imaging of exoplanets in the near-infrared. GPI saw first light at Gemini South in Chile in November 2013, and was tasked with surveying 531 nearby stars for exoplanets as part of the Gemini Planet Imager Exoplanet Survey until 2019. GPI 2.0 is a project currently underway to upgrade GPI, with improvements in multiple areas including a new wavefront sensor and better cameras.

The cameras are being upgraded to First Light CRED-2s, which boast increased speed and noise performance over the current ones. The installation of the pupil camera requires an optics calculation to ensure the pupil is properly imaged onto the camera detector. For the calibration system (CAL), only a small area of the camera's detector will be used; thus, the optimal 96-by-96-pixel area has been determined. Installation of the lenslet array over this area will require precision tuning, in order to align the lenslets properly along the pixel grid for point spread function (PSF) centering. Additionally, new code must be written to ensure that the GPI user interface can interact with and control the camera properly.

***Molecular Analysis of a Bifunctional Mycobacterium tuberculosis Virulence Factor***

Rebecca Prest
Department of Biological Sciences
Advisor: Patricia Champion
Coauthors: NA

The ESX-1 secretion system is required for virulence of pathogenic Mycobacteria, including *Mycobacterium tuberculosis* and nontuberculous Mycobacteria (NTM) such as *Mycobacterium marinum*. This secretion system is essential for Mycobacteria to rupture the phagosomal membrane and gain access to the cytoplasm of host macrophages. In addition, the ESX-1 system is known to broadly regulate gene expression. Phagosomal lysis is a precedent for host cell death and mycobacterial cell-to-cell spread. However, the mechanism of substrate transport to the extracellular environment, as well as the signals that link transcription factors to the assembly of the ESX-1 membrane complex are largely unknown. One of the ESX-1 secreted proteins, EspE, is known to regulate gene expression in the cytoplasm and promote virulence following secretion. EspE may link known transcription factors to the assembly of the ESX-1 membrane complex. This protein's production and subsequent secretion is also required for mycobacterial virulence. This project aims to further study the dual functions of this protein via structure-function analysis and biochemical approaches.

***Examining the influence of periodical cicadas on stream nutrient uptake and their roles as a resource subsidy***

Abagael N. Pruitt
Department of Biological Sciences
Advisor: Dr. Jennifer L. Tank
Coauthors: Ellie D. Snyder, Anna E.S. Vincent, Erik M. Curtis, Emma M. Thrift

During the summer of 2021, the 17-year Brood X cicada (*Cicadidae Magicicada*) emerged in many parts of the United States. These cicadas die shortly after emerging, providing the surrounding environment with resources including nitrogen, carbon, and phosphorus. During mass emergence, as many as 300 cicadas may enter one square meter of a stream, potentially impacting biofilm growth, nutrient uptake, and stream metabolism. We hypothesized that the addition of cicada-derived nutrients and carbon, released via decomposition, would enhance biofilm growth, nutrient uptake, and reach-scale metabolism (as both autotrophic production and heterotrophic respiration). To quantify the impact of variable inputs of cicada subsidies on stream ecosystem function, we conducted a cicada addition experiment using the replicated streams at the Notre Dame Linked Environmental Ecosystem Facility (ND-LEEF), comparing the following treatments: the addition of 100 litter bags containing 300 cicadas per bag to mimic "high" cicada inputs vs. 100 bags of 150 cicadas per bag to represent future conditions where climate change limits cicada populations, using a stream with 100 empty bags (no cicadas) as a control to isolate the effect of litter bag deployment alone. Over a period of nine weeks, we performed short-term nutrient releases of nitrate-N, ammonium-N, and dissolved reactive P to examine changes in nutrient uptake in the context of cicada decomposition, along with weekly biofilm sampling, and monitoring of continuous metabolism using deployed oxygen sensors. Preliminary results suggest that cicada decomposition alters stream nutrient uptake via changes in biofilm. This research is the first to examine the impact of periodical cicadas on stream ecosystem function, advancing our understanding of how this infrequent but novel resource subsidy impacts ecosystems at present and under a changing climate.

## *Fermi level tuning and band alignment in Mn doped InAs/GaSb*

Logan Riney
Department of Physics
Advisor: Badih Assaf
Coauthors: J. Bermejo-Ortiz, G. Krizman, J. Wang, M. Zhukovskyi, T. Orlova, L.A. de Vaulchier, Y. Guldner, R. Winkler, J.K. Furdyna, X. Liu

InAs/GaSb hosts a broken gap band alignment that has been shown to generate helical topological edge states. Upon the introduction of Mn into the structure, it has been predicted to host a quantized anomalous Hall effect, or have a quantized Hall Effect contribution from the magnetization. Here, we show that dilute Mn doping on InAs in InAs/GaSb, allows a tuning of the Fermi level, the introduction of paramagnetism, but also has a non-trivial impact on the band alignment of the system. The measurement of Shubnikov-de-Haas oscillations, cyclotron resonance, and a non-linear Hall effect in Mn-doped samples indicate the coexistence of a high mobility two-dimensional electron gas and a hole gas. Conversely, in undoped InAs/GaSb, pure-n-type transport is observed. We hypothesize that Mn acceptor levels can pin the Fermi energy near the valence band edge of InAs, far from the interface, which introduces a strong band bending to preserve the band offset at the InAs/GaSb interface. The realization of the QAHE in this structure will thus require a careful control of the band alignment to preserve topological insulating character.

***Bacterial Community Association with Key Protozoans in the Guts of Long-Tailed Macaques in Southeast Asia***

Chissa-Louise Rivaldi
Department of Biological Sciences
Advisor: Dr. Hope Hollocher
Coauthors: Justin Wilcox, Benjamin Gombash

Microbial communities in the guts of mammals are shaped by a number of factors governing their structure and fluctuations. Measures of composition, diversity, and ecological distance reflect how changes of bacterial communities are associated with the host environment, genetics, and behavior. Less well-studied are the interactions between these prokaryotic communities and resident protozoans of mammalian hosts, particularly in free-ranging animal systems. To investigate these associations more closely, we use variable regions of the 16S and 18S rRNA genes amplified from fecal samples of long-tailed macaques in Bali, Indonesia and Singapore to analyze prokaryotic and protozoan taxa. Our findings reveal principal sources of variation in protozoan taxa in this environment centering around few protozoan taxa (*Blastocystis, Plasmodium, Entamoeba)*. We then use the presence and abundances of these taxa to reveal patterns of variation in the prokaryotic community and uncover evidence of associations between protozoan and prokaryotic variation. These findings highlight the importance of the inclusion of taxa beyond bacteria when examining the dynamics of microbial communities, particularly in populations frequently exposed to protozoans.

# Structural and Functional Characterization of a Conserved Mycobacterial Transcription Factor

Kevin G Sanchez[1], Rebecca J. Prest[1], and Dr. Patricia A. Champion[1]

[1]Department of Biological Sciences, University of Notre Dame

*Mycobacterium tuberculosis* and other pathogenic mycobacteria use the ESX-1 secretion system to survive within the host macrophage. In spite of its clear role in mycobacterial pathogenesis, the mechanisms regulating the ESX-1 system are unclear. Using *Mycobacterium marinum*, a mycobacterial pathogen and an established model for *M. tuberculosis* ESX-1 secretion, we found that deletion of the ESX-1 secretory machinery caused significant and widespread changes in gene expression, most notably in regards to its own regulation. Loss of the ESX-1 secretory apparatus leads to repression of the conserved transcription factor, WhiB6. WhiB6 positively controls expression of the ESX-1 substrate genes. We identified a second conserved transcription factor, EspM, that regulates *whiB6* gene expression. Using EMSAs, we demonstrated that EspM binds directly and specifically to the *whiB6* promoter through the C-terminal half of the protein. Using bioinformatic, biochemical, and genetic tools, we are further elucidating the role of EspM and the mechanism by which it regulates the ESX-1 pathway. EMSA data indicated that EspM was able to “supershift” the *whiB6* promoter, suggesting multimerization of EspM. Using HPLC, we demonstrated that EspM multimerizes, and that either the C-terminal or N-terminal half of EspM can multimerize. The ability for each half to self-associate suggests multiple forms of EspM multimers. Lastly, we identified relevant residues in a C-terminal helix-turn-helix domain that are necessary for EspM multimerization using a bacterial-2-hybrid system. We seek to define the relevance of specific EspM residues and EspM-multimerization to ESX-1 regulation and function.

***Effect of clinical mutations in OXA-24/40:***
***an analysis of chemical shift temperature dependence***

Anna Santa
Department of Chemistry and Biochemistry
Advisor: Jeffrey W. Peng
Coauthors: Jamie VanPelt, Shannon Stoffel, Vita Zhang, Kayla Dempster

Beta-lactamases are a group of enzymes that cause resistance in gram-negative bacteria to beta-lactam antibiotics known as carbapenems. One such protein, OXA-24/40, found in *Acinetobacter baumannii*, hydrolyzes carbapenems, but has multiple known clinical mutations that exhibit a range of different behaviours and cause growing concerns in hospital environments where they emerge in secondary infections. Epistasis should also be considered when looking at evolution of clinical mutants – exploration of both clinical and lab-designed mutations can help understand evolutionary trajectories of this rapidly mutating protein. An NMR temperature study can provide information on internal dynamics, exchange and bonding patterns. In this study, wild type (OXA-24) and three mutants (G224D, N87I, double mutant G224D-N87I) were examined. Experiments were conducted on $^{15}$N-labelled samples and 2D correlation spectra of amides were recorded at temperatures ranging 283-303 K. The Shift-Track and Curvalyzer algorithms by Trainor et al. were used to obtain temperature coefficients and curvature fitting. Results show that the effect of G224D is more localized than N87I, which is expected from their positions in the structure. The effect of N87I dominates the double mutant, however, for a cluster of residues, the effect was additive, and behaviors unique in each mutant were also observed at functionally relevant sites. Changes in H-bond patterns are indicated by the results. Further comparisons to previously designed mutant R261S are in progress to understand how each mutation perturbs the surroundings of this residue.

## *Selecting for an Aptamer that Targets RNA Triple Helices*

Mika Schievelbein
Department of Chemistry and Biochemistry, University of Notre Dame, Notre Dame, IN 46556
Advisor: Jessica Brown

Canonical U•A–U and $C^+$•G–C base triple formation was first discovered *in vitro* by Felsenfeld, Davies, and Rich in 1957 (where • and – denote the Hoogsteen and Watson-Crick faces, respectively). Since then, validation of naturally-occurring major groove RNA triple helices, with at least three consecutive base triples, has been limited to thirteen due to the low-throughput nature of solving 3-dimensional structures. There is a need for a high-throughput method for RNA triple helix discovery. Aptamers are a promising tool, outperforming other molecular tools due to their high specificity and affinity, stability, small size, and ease of synthesis, to name a few. To generate an RNA triple helix-binding aptamer, Capture-Systematic Evolution of Ligands by EXponential Enrichment (SELEX) will be employed, selecting for a functional aptamer from a large (~$10^{15}$), randomized pool of DNA aptamer sequences through repeated cycles of selections. Capture-SELEX immobilizes the aptamer pool, allowing for a mixture of RNA triple helices and corresponding duplexes for positive and negative selections, respectively, to flow through. Minimal structures of triple helices from human MALAT1 and telomerase RNAs, a synthetic 11-base triple RNA, and corresponding triplex-deficient structures have been generated and confirmed to have proper triple helix and duplex structures based upon their UV-vis thermal denaturation melting profiles. The efficacy of the aptamer as an experimental tool will be validated by pulling down RNA triple helices in cell lysates. Generating an aptamer that binds to RNA triple helices with broad specificity and high affinity will streamline RNA triple helix discovery, leading to the establishment of the triplexome.

***High order collision-based hybrid method for the BGK equation***

Minwoo Shin
Department of Aerospace and Mechanical Engineering
Advisor: Ryan McClarren
Coauthors: Cory Hauck, Ryan McClarren

We apply the collision-based hybrid method for the Boltzmann equation with the BGK operator in a hyperbolic scaling, which leads to Euler or compressible Navier–Stokes equations in the asymptotic limit. Implicit treatment for the source term is necessary to solve hyperbolic systems with stiff sources. Although it helps the numerical scheme become stable with a large time step size, it is still not obvious to achieve the desired order of accuracy due to the relationship between size of the spatial cell and mean free path. Without the asymptotic preserving property, a very restricted size of the spatial cell is required to resolve the mean free path, which is not practical. Our approaches are based on the noncollision-collision decomposition of the BGK equation. We introduce the arbitrary order of nodal discontinuous Galerkin (DG) discretization in space with semi-implicit time stepping method; we employ the backward Euler time integration for the uncollided equation and the 2nd order predictor-corrector scheme for the collided equation, i.e., both source terms in uncollided and collided equations are treated implicitly, and only streaming term in the collided equation is solved explicitly. This improves the computational efficiency without dealing with the complexity of the numerical implementation. Numerical results are presented for various Knudsen numbers to present the effectiveness and accuracy of our hybrid method. Also, we compare the solutions of the hybrid and non-hybrid scheme.

***Selective Recognition of RNA Triple Helices by Small Molecules***

Krishna M. Shivakumar
Department of Chemistry and Biochemistry,
University of Notre Dame, IN 46556 USA
Advisor: Jessica A. Brown
Coauthor: Charlotte N. Kunkler

The human genome project and deep sequencing revealed that 76% of human DNA is transcribed into RNA but only 3% of human DNA is eventually translated into proteins. Thus, RNAs are emerging as important drug targets with advances in understanding the functions, structures, and binding properties of more and more RNAs. Unlike single- and double-stranded RNAs, RNA triple helices are understudied and their interactions with small molecules is a new frontier. In our current study, we aim to better understand the structure, molecular recognition and interactions of triple-helical RNAs and various small molecules. A deeper understanding of structural and biophysical properties would help to develop a fundamental knowledge and facilitate future research in designing lead drug molecules targeting disease-related RNA triple helices. I will present my preliminary data on small molecules binding to metastasis associated lung adenocarcinoma transcript 1 (MALAT1) triple helix. Differential scanning fluorimetry (DSF), UV melting and dye displacement assay experiments are being employed to evaluate the selectivity, affinity and binding mode of small molecules to MALAT1 RNA triple helix. Thus far, DSF results show selective recognition of MALAT1 triple helix, but not the RNA counterpart lacking triple helix, by berberine, berenil, and sanguinarine. In contrast, neomycin binds non-specifically to the MALAT1 triple helix. The binding affinity of MALAT1 triple helix-small molecule complexes follow the order: quercetin (172 ± 35 nM) > neomycin > sanguinarine > berenil > berberine (14 ± 7 μM). The dye displacement assay with standard intercalating SYBR Green II confirms intercalating behavior of sanguinarine and non-intercalating binding mode for berberine and berenil molecules while neomycin binds nonspecifically. Future work is focused on elucidating the three-dimensional structure of the free and small molecule-bound RNA triple helices using cryo-EM.

***Title: Velocity and substrate conditions influence the particle size distribution of environmental DNA (eDNA) in recirculating mesocosms.***

Elise D. Snyder
Department of Biological Sciences
Advisor: Jennifer Tank & Gary Lamberti
Coauthors: E.D. Snyder, J.L. Tank, P.F.P. Brandão Dias, K. Bibby, A.W. Bivins, A.J. Shogren, D. Bolster, S.P Egan, G.A. Lamberti

Abstract: Environmental DNA (eDNA) sampling offers insight into the distribution of invasive and/or rare aquatic species without the use of intensive traditional sampling. However, our understanding of eDNA fate and transport in flowing waters (e.g. stream and rivers) is limited, which constrains predictions about the location and density of target organisms after positive detection. We conducted pulse releases of Common Carp (*Cyprinus carpio*) and Steelhead (*Oncorhynchus mykiss*) eDNA in recirculating mesocosms to quantify transport and removal for a range of eDNA particle size classes and compare transport between the two eDNA sources. The mesocosms (n=24) were distributed between three substrate treatments (no substrate, bare substrate, biofilm-colonized substrate) and two light availability treatments (shaded, unshaded), which allowed us to measure eDNA removal under different environmental conditions. To estimate eDNA removal we collected water samples from each mesocosm at intervals of 40 minutes, 6, 18, and 48 hours and filtered the samples through 10, 1.0, and 0.2μm filters to separate particles by size. Total eDNA removal rates were highest for the biofilm-colonized mesocosms, which is consistent with previous findings linking biofilm colonization levels with increased eDNA degradation. For the different eDNA particle size classes, we found that smaller fragments made up greater proportion of the total eDNA for the bare and biofilm substrate mesocosms, which suggests that, in rivers and streams, eDNA samples taken farther from the target organism would have a greater proportion of small fragments present. We also found variation in eDNA particle size distribution between Carp and Steelhead, suggesting the potential for eDNA transport differences between the two sources. This work contributes to our understanding of eDNA transport and will enhance our ability to predict eDNA transport in streams, in time advancing the use of eDNA as a tool for monitoring and management.

**Investigations of Macrocycle Breathing Dynamics in a Cyclodextrin-based Rotaxane**

Shannon Stoffel

Department of Chemistry and Biochemistry

Advisor: Jeffrey W. Peng

Coauthors: Qi-Wei Zhang, Dong-Hao Li, Bradley D. Smith, and Jeffrey W. Peng

Biological machines have inspired the creation of synthetic molecular machines. One interest is the ability to switch “ON” the molecular machine in response to external stimulus. A molecular switch can be seen in [1]rotaxane and [3]rotaxane where the rapid pirouetting of the threaded α-cyclodextrin could be switched “ON” or “OFF” by the addition of chemical additives.[1] This molecular switch allows for the ability to study the rapid pirouetting by contrasting the behavior of the “ON” and “OFF” rotaxane.

We wanted a better understanding of the motion occurring in [3]rotaxane so we compared the relaxation rates of the two systems. [2] The μs-ms motions of methine groups on the cyclodextrin were compared using NMR $^{13}C$ CPMG relaxation dispersion experiments. The “ON” version showed dispersion at positions C1, C2, and C4, suggesting conformation mobility on the μs-ms time scale at these sites, whereas dispersion was not detected for the “OFF” version.

We combined these results with Molecular Dynamics studies to understand the intercomponent motion of Rotaxanes. Understanding the nature of the mobility of these nanomachines is imperative in understanding the rotary motion.

**References:**

1. Zhang, Q. W.; Zajicek, J.; Smith, B. D., Cyclodextrin Rotaxane with Switchable Pirouetting. *Organic Letters* **2018,** *20* (7), 2096-2099.
2. Stoffel, S.; Zhang, Q.-W.; Li, D.-H.; Smith, B. D.; Peng, J. W., NMR Relaxation Dispersion Reveals Macrocycle Breathing Dynamics in a Cyclodextrin-based Rotaxane. *Journal of the American Chemical Society* **2020,** *142* (16), 7413-7424.

***Agricultural conservation influences nutrient and pathogen dynamics at both the field and subwatershed scale***

Emma Thrift
Department of Biology
Advisor: Dr. Jennifer Tank
Coauthors: Jennifer L. Tank, Anna E.S. Vincent, Ursula H. Mahl, Alan Hamlet

Fertilizer runoff from farm fields provides a significant source of nutrients to streams draining agricultural watersheds, causing eutrophication of downstream ecosystems. Fertilizer inputs via manure application can also impact health and recreation through pathogen release. Agricultural conservation practices, such as winter cover crops and drainage water management (DWM) can reduce nutrient losses to streams, while their influence on pathogen transport remains understudied. We examined nutrient and pathogen loss at both the field-scale and at watershed outlets within the Paw Paw River Basin (MI). At the field scale, we compared grab samples against nutrient measurements from sensors deployed in subsurface tile drains employing DWM (i.e., regulating water release from agricultural fields); data will be used to develop a model that informs the automated operation of the DWM system. Our analyses indicated that over two years, there was a 66% reduction in *Escherichia coli (E. coli)* loads in the tile drains with DWM. At the watershed-scale, we sampled three watershed outlets every two weeks to assess the role of cover crops in reducing the loss of nitrate, soluble reactive phosphorus (SRP), and *E.coli* from agricultural landscapes. Subwatersheds represent traditional row-crop agriculture (AG), agriculture with winter cover crops (CC), and forested reference (REF) conditions. Nitrate export with CCs was 20% lower in the 2019 water year (WY) and 15% lower in WY 2021. Additionally, *E. coli* counts were 73% lower in 2020, during the CC growing season (Oct-May). We did not see SRP reductions with CC, possibly due to the influence of diffuse SRP inputs from a dairy within the CC watershed. We suggest that cover crops are effective at reducing nitrate and may limit *E. coli* losses under some conditions. Additional conservation strategies, specifically targeting SRP sources (i.e., manure), may be necessary to compensate for heterogeneous agricultural impacts and improve water quality.

**High-throughput screening of a novel structure-activity library against *N. fowleri* in a newly developed cytopathogenicity assay.**

Emma V. Troth
Department of Infectious Diseases, University of Georgia
Center for Tropical and Emerging Global Diseases
Advisor: Cristian Koepfli (Notre Dame); Dennis Kyle (University of Georgia)
Coauthors:
Jopaul Mathew, Department of Chemistry and Virginia Tech Center for Drug Discovery, Virginia Tech, Blacksburg, Virginia, United States
Emilio F. Merino, Department of Biochemistry and Molecular Biology, University of Georgia, Athens, Georgia, United States; Center for Tropical and Emerging Global Diseases, Athens, Georgia, United States
Paul R. Carlier, Department of Chemistry and Virginia Tech Center for Drug Discovery, Virginia Tech, Blacksburg, Virginia, United States
Maria B. Cassera, Department of Biochemistry and Molecular Biology, University of Georgia, Athens, Georgia, United States; Center for Tropical and Emerging Global Diseases, Athens, Georgia, United States
Dennis E. Kyle, Department of Infectious Diseases, University of Georgia, Athens, Georgia, United States; Department of Cellular Biology, University of Georgia, Athens, Georgia, United States; Center for Tropical and Emerging Global Diseases, Athens, Georgia, United States

The brain-eating amoeba, *Naegleria fowleri*, poses a substantial public health risk. The freshwater dwelling *N. fowleri* is the causative agent of Primary Amebic Meningoencephalitis (PAM) in humans. PAM carries a 98% fatality rate, with death occurring 5-12 days after symptom onset. A pressing need exists for the development of new therapeutics for this rapidly lethal disease. *N. fowleri* secretes multiple proteases during infection that aid in the breakdown of tissue. *N. fowleri* also relies on its cytoskeleton to form food cups which allow the amoebae to eat via trogocytosis. Current therapeutics do not target either of these destructive amoebic functions. In this study, we first developed a new cytopathogenicity assay and then used it to assess compounds for ability to inhibit *N. fowleri*-mediated destruction of a mammalian cell monolayer (human foreskin fibroblasts (HFFs)). This assay allows for live-cell imaging of cytopathogenicity as well as quantitative endpoints for compound efficacy. Monolayer degradation was quantified using automated microscopy of the CellTrace-stained HFFs. Using this assay, we are able to measure *N. fowleri*-mediated degradation and derive inhibitory values for compounds tested. First, we assessed currently used PAM therapeutics and a series of protease and actin inhibitors for ability to inhibit *N. fowleri*-mediated monolayer destruction. Next, we standardized the assay for single-point high throughput screening. A novel 80-compound structure-activity library was screened in the assay at 20 µM. Hit compounds were selected for dose-response in addition to screening against *N. fowleri* trophozoites alone in the established CellTiter-Glo assay. Compounds were also assessed for cytotoxicity against HFFs. Hit compounds were incubated with live *N. fowleri* trophozoites and imaged for compound localization. From this library, we have identified 4 hit compounds for further optimization as new drugs for treating PAM.

**The Effects of Conservation on Ammonium Dynamics in Two Agricultural Watersheds**

Anna E. S. Vincent
Department of Biological Sciences
Advisor: Jennifer L. Tank
Coauthors: Abagael N. Pruitt, Shannon L. Speir, Matt T. Trentman, Ursula H. Mahl, Todd V. Royer

The application of synthetic Nitrogen (N) fertilizer and use of modified drainage techniques (e.g., stream channelization and tile drains) are common practices across agricultural landscapes to maximize crop yields. When N fertilizer is applied as ammonium ($NH_4^+$-N), it is rapidly bound to soil particles and transformed by microbes into nitrate ($NO_3^-$-N) via the process of nitrification. When found in excess, $NO_3^-$-N stimulates algal blooms, causes eutrophic conditions, and indicates degraded water quality. Though the widespread use of agricultural conservation practices, such as winter cover crops, is known to reduce stream $NO_3^-$-N losses, less is known about how cover crops influence $NH_4^+$-N losses from tile drains and how in-stream nitrification contributes to overall $NO_3^-$-N export. We assessed the impact of winter cover crops on ammonium cycling and transformation dynamics using long-term datasets collected in two agricultural watersheds in Indiana (USA), Shatto Ditch Watershed (SDW) and Kirkpatrick Ditch Watershed (KDW). We collected water samples every two weeks from watershed outlets, multiple stream sites, and tile drains within the watersheds. We compared losses at both the field (i.e., tile drain) and watershed scale, normalizing for differences in the sizes of the watersheds and flow where appropriate. Preliminary analyses at the field-scale demonstrate that the presence of cover crops results in significant reductions in both flow and $NO_3^-$-N loss from tile drains by 42% and 45%, respectively. However, cover crops did not result in significant reductions in $NH_4^+$-N loss. While patterns in watershed-scale $NO_3^-$-N export mirrors stream discharge, the export of $NH_4^+$-N is more intermittent throughout the study period with the highest peaks coinciding with high flow events. Given the importance of $NH_4^+$-N as a driver of nitrification, understanding associated seasonal and environmental controls is critical to manage $NH_4^+$-N losses and maintain water quality.

# The Use of an Inverse Multi-Domain Rate Transfer Model in Predicting Colloid Attachment under Different Environmental Charge Conditions

Sabrina Volponi
Department of Civil & Environmental Engineering and Earth Sciences
Advisor: Diogo Bolster and Jennifer Tank
Coauthors: Diogo Bolster, William Johnson, Eddy Pazmiño

It has recently been shown that the transport of colloids under colloid-collector repulsive conditions can be predicted by a multi-domain rate transfer model. Constructing a distribution of colloid travel times under this model, however, requires assessing the impact of hydrodynamic forces and physiochemical interactions on colloid attachment within a system. Since these forces are often difficult to assess, this project seeks to build an inverse model capable of predicting the colloid residence time distribution from experimentally measured concentration and retention profiles. Breakthrough curve data was found to not provide enough information on its own to successfully describe the distribution of colloid travel times. Although more difficult to collect, some retention profile information is therefore needed to specify the inverse model.

***Coexistence of superconductivity, fluctuations and spin-orbit splitting in $Sn_{1-x}In_xTe$ thin film***

Jiashu Wang
Department of Physics
Advisor: Badih A. Assaf
Coauthors: W. Powers, B. J. McIntosh, S.K. Bac, L. Riney, M. Zhukovskyi, T. Orlova, X.Liu

Topological superconductors are superconductors that have boundary states that consist of a special type of quasiparticles, known as Majorana fermions. Topological modes like Majorana fermions are stable to perturbation from the environment. This unique property leads to a very promising application in quantum computing, which requires noise stability of qubits.

However, although theoretically predicted, the experimental realization of topological superconductors with Majorana fermions is still being pursued. Here, we report the growth and characterization of Sn1-xInxTe, a possible topological superconductor.

It has been found that by In doping, Sn1-xInxTe (SIT) can be superconducting and also maintains topological surface states. Most previous research of SIT was focused on single crystals but the growth and analysis of SIT thin films are necessary to search for boundary Majorana states. Here, we achieve the MBE growth of Sn1-xInxTe(x=0.04-0.3) thin films(100nm) on BaF2 (111), and characterize them by X-ray diffraction, transmission electron microscopy and magneto-transport measurement. A superconducting transition is consistently observed in films, and the critical temperature Tc increases as we increase the In concentration up to 3.5K. The normal state is shown to host coexisting weak localization (WL) and weak anti-localization (WAL). The WL only occurs in superconducting samples, indicating that electrons from trivial spin-orbit split states may play a dominant role in the superconducting state. Close to but above Tc, a signature of superconducting fluctuations is also observed. Overall, our findings motivate further experiments to find the origin of a possible spin-orbit splitting and study its impact on the pairing symmetry of Sn1-xInxTe and its possible topological character.

## *Variational Inference with NoFAS: Normalizing Flow with Adaptive Surrogate for Computationally Expensive Models*

Yu Wang
Department of Applied Computational Mathematics and Statistics
Advisor: Fang Liu
Coauthors: Fang Liu, Daniele E. Schiavazzi

Fast inference of numerical model parameters from data is an important prerequisite to generate predictive models for a wide range of applications. Use of sampling-based approaches such as Markov chain Monte Carlo may become intractable when each likelihood evaluation is computationally expensive. New approaches combining variational inference with normalizing flow are characterized by a computational cost that grows only linearly with the dimensionality of the latent variable space, and rely on gradient-based optimization instead of sampling, providing a more efficient approach for Bayesian inference about the model parameters. Moreover, the cost of frequently evaluating an expensive likelihood can be mitigated by replacing the true model with an offline trained surrogate model, such as neural networks. However, this approach might generate significant bias when the surrogate is insufficiently accurate around the posterior modes. To reduce the computational cost without sacrificing inferential accuracy, we propose Normalizing Flow with Adaptive Surrogate (NoFAS), an optimization strategy that alternatively updates the normalizing flow parameters and the weights of a neural network surrogate model. We also propose an efficient sample weighting scheme for surrogate model training that ensures some global accuracy of the surrogate while capturing the likely regions of the parameters that yield the observed data. We demonstrate the inferential and computational superiority of NoFAS against various benchmarks, including cases where the underlying model lacks identifiability. The source code and numerical experiments used for this study are available at https://github.com/cedricwangyu/NoFAS.

# Fabrication of a hierarchical polymer membrane adsorbent by a combined 3D-printing and in-situ phase separation process

Jialing Xu, Thomas Kasl, Adam Braegelman, Kevin Gabriel Alvarez, Matthew Webber and William Phillip

Keywords: 3d printing, SVIPS, membrane adsorbent, heavy metal removal

The increasing world population makes meeting the demand for fresh water a challenge for years to come. Among different choices of sorbents, polymer membrane sorbents are a popular choice for separation processes due to their tunable porous structures with the possibility for surface functionalization. To achieve the goals of high selectivity, large permeability, and durable mechanical performance simultaneously, additive manufacturing technologies have emerged because they can fabricate membrane sorbents with precisely controlled patterns and desired hierarchical structures. In this work, a simple solution-based direct writing three-dimensional (3D) printing technique combined with surface-segregation and vapor-induced phase separation (SVIPS) methodology was used to fabricate a hierarchical membrane sorbent. Composite inks containing polysulfone (Psf), polystyrene-*block*-poly(acrylic acid) (PS-*b*-PAA), and carbon nanotubes (CNTs) were pumped at a constant flow rate while the nozzle on the programmed 3D printer was moved to form woodpile structures. The woodpile structures consist of large channels (~250 µm diameter) between filaments that result from the printing process while the filaments themselves contain nanoscale pores (~300nm diameter) distributed throughout due to nonsolvent induced phase separation. The relative hydrophilic PAA blocks were brought to the pore walls during the phase separation process and further tailored by carbodiimide reactions with polyethylenimine (PEI) and terpyridine (Terp). The PEI increases the density of the metal ion binding sites and the Terp increases the affinity towards heavy metal ions. The multi-layers hierarchical membrane has high permeability (i.e., $10^4$ L $M^{-2}$ $h^{-1}$ $bar^{-1}$) with a sorbent capacity of 30 mmol $m^{-2}$ [based on $Cu^{2+}$ uptake]. Flowthrough experiments demonstrate the efficient removal of low concentration copper in feed solutions under dynamic flow conditions. Finally, the sorbent exhibits highly selective absorption towards $Co^{2+}$ in cobalt/lithium mixtures, which provides an insight for cobalt recovery.

*Metal-Poor Stars Observed with the Southern African Large Telescope II. An Extended Sample*

Joseph Zepeda
Department of Physics
Advisor: Tim Beers
Coauthors: Kaitlin C. Rasmussen, Timothy C. Beers, Vinicius M. Placco, Yang Huang, and Éric Depagne

We present results from high-resolution (R ~ 40,000) spectroscopic observations of over 200 metal-poor stars, mostly selected from the RAVE survey, using the Southern African Large Telescope. We were able to derive stellar parameters for a total of 108 stars; an additional sample was previously reported on by Rasmussen et al.. Among our newly reported observations, we identify 84 very metal-poor (VMP; [Fe/H] < -2.0, 53 newly identified) stars and 3 extremely metal-poor (EMP; [Fe/H] < -3.0, 1 newly identified) stars. The elemental abundances were measured for carbon, as well as several other α-elements (Mg, Ca, Sc, Ti), iron-peak elements (Mn, Co, Ni, Zn), and neutron-capture elements (Sr, Ba, Eu). Based on these measurements, the stars are classified by their carbon and neutron-capture abundances into carbon-enhanced metal-poor (CEMP; [C/Fe] > +0.70), CEMP sub-classes, and by the level of their *r*-process abundances. A total of 17 are classified as CEMP stars. There are 11 CEMP-*r* stars (8 newly identified), 1 CEMP-*i* star (newly identified), 2 possible CEMP-*i* stars (1 newly identified), and 3 CEMP-no stars (all newly identified) in this work. We found 11 stars (8 newly identified) that are strongly enhanced in *r*-process elements (*r*-II; [Eu/Fe] > +0.70), 38 stars (31 newly identified) that are moderately enhanced in *r*-process elements (*r*-I; +0.30 < [Eu/Fe] ≤ +0.70), and 1 newly identified limited-*r* star. We plan on using a clustering algorithm on the results of this work as well as stars from the literature to gain a better understanding of CEMP progenitors and the star forming environments that they create.

***Stripe-patterned membrane for multi-ions detection***

Shukun Zhong
Department of Chemical and Biomolecular Engineering
Advisor: William A. Phillip
Coauthors: John R. Hoffman, Tao Wang

Heavy metal is one of the primary pollutants in wastewater. Membrane patterned with various ligands provides an efficient method to detect the metals due to fast ion transport, easy operation, and color indication. Phenolphthalein-based cardo poly(arylene ether sulfone) with pendant carboxylic acid groups (PPAES-COOH) copolymer was applied to cast porous membranes. Then, the membranes were reacted with an alkyne moiety on the pore walls. A UV-initiated thiol-yne reaction was used to introduce two ligands (carboxylic acid and terpyridine) on the membrane surface. The copper adsorption capacity of the carboxylic acid functionalized membrane is close to the copper adsorption of the parent PPAES-COOH membrane. Later on, a patterned module was applied to the thiol-yne reaction to attach both ligands on the membrane surface with a designated shape. Finally, the membrane successfully identified copper (II) and iron (II) with the blue and purple stripes appearing due to metal-ligand interaction.