

The Roy J. Carver Department of Biochemistry,  
Biophysics, and Molecular Biology Undergraduate Club Presents:

# STUPKA<sup>2021</sup>

## UNDERGRADUATE RESEARCH SYMPOSIUM



**April 9 & 10, 2021**

IOWA STATE  
UNIVERSITY

## EVENT SCHEDULE

### FRIDAY, APRIL 9

- 11:00 am** Dr. Jeff Karp Student Interaction
- 12:00 pm** Dr. Elizabeth Sattely Student Interaction  
Dr. Gaya Amarasinghe Student Interaction
- 1:00 pm** Poster Session 1
- 2:00 pm** Poster Session 2
- JOIN SESSION NOW**, click to go to <https://us02web.zoom.us/j/89490693341>
- 4:00 pm** **Welcome**  
*Dr. Kent Kerby, Assistant Dean, College of Liberal Arts and Sciences*
- 4:10 pm** **Sarah Grambo – Student Speaker**  
*Accumulation of aphid secretions changes the cuticular surface of the soybean plant*
- 4:25 pm** **Mariah Hoye, Ph.D. – Alum Speaker**  
*The role of an Intellectual Disability gene, DDX3X, in regulating translation required for neural progenitor fate decisions during brain development*
- 4:40 pm** **Jeff Karp, Ph.D. – Keynote Speaker**  
*Towards Accelerated Medical Innovation*
- 5:30 pm** **Break, Poster Awards**
- 5:40 pm** **Richard Weerts – Student Speaker**  
*Xyloglucan xylosyltransferase 1 displays promiscuity toward donor substrates during in vitro reactions*
- 5:55 pm** **Jennifer Gribble – Alum Speaker**  
*RNA recombination in coronaviruses*
- 6:10 pm** **Madeline Farringer – Student Speaker**  
*Developing an auxin-inducible degron system for use in Plasmodium falciparum*
- 6:25 pm** **Elizabeth Sattely, Ph.D. – Keynote Speaker**  
*Total biosynthesis of plant-derived therapeutics*
- 7:15 pm** **Close for the Day**

### SATURDAY, APRIL 10

- 9:30 am** **Opening Remarks**  
*Dr. Kristen Johansen, Professor and Chair, Department of Biochemistry, Biophysics and Molecular Biology*
- 9:45 am** **Luke Helgeson, Ph.D. – Alum Speaker**  
*Mechanisms of Load Bearing within the Kinetochore*
- 10:00 am** **Jacob Schmieder – Student Speaker**  
*Wearable Flexible Enzymatic Sensors using Graphene*
- 10:15 am** **Jacqueline Rivas, Ph.D. – Alum Speaker**  
*Enhancing T-cell antitumor immunity in Chronic Lymphocytic Leukemia*
- 10:30 am** **Gaya Amarasinghe, Ph.D. – Keynote Speaker**  
*Molecular Mechanisms of Viral Immune Evasion*
- 11:20 am** **Break**
- 11:30 am** **Behnia Rezazadeh Shirazi – Student Speaker**  
*Reproducibility and Normalization of Reactive Hyperemia using Laser Speckle Contrast Imager*
- 11:45 am** **Tony Cyr, MD/Ph.D. – Alum Speaker**  
*Novel Circulating Sphingolipid Signatures in the Plasma Metabolome are Associated with Better Outcomes Following Blunt Trauma*
- 12:00 pm** **Tyler Gilbreath – Alum Speaker**  
*The Role of the E3 Ubiquitin Ligase UBR5 in B Cell Development and MCL Pathogenesis*
- 12:15 pm** **Samson Condon, Ph.D. – Alum Speaker**  
*Exploring the Sequence Landscape of a Highly Dimeric Transmembrane Helix*
- 12:30 pm** **Denis Tamiev – Alum Speaker**  
*Deep Learning to Improve Bacterial Cell Counting – Implementation of Classification-Type Convolutional Neural Networks (CNN)*
- 12:45 pm** **Closing Remarks**
- 3:00-5:00 pm** **BBMB and Stupka Alumni Virtual Reunion**



# SAVE THE DATE FOR THE 16<sup>TH</sup> ANNUAL STUPKA SYMPOSIUM

**Thursday, April 7, 2022**

email: [stupkaugrs@iastate.edu](mailto:stupkaugrs@iastate.edu)

website: [stupka.bb.iastate.edu](http://stupka.bb.iastate.edu)

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## 2021 STUPKA COMMITTEE

<b>SYMPOSIUM CO-CHAIRS</b>	Ana DiSpirito and Emily Boettger
<b>SECRETARY</b>	Patricia Gallardo
<b>TREASURER</b>	Pahton McDonald
<b>TREASURER UNDERSTUDY</b>	Kevin Beaghan
<b>Sub-Committee Coordinators</b>	
<b>SPEAKER CO-CHAIR</b>	Maddie Farringer and Tulika Sharan
<b>ALUMNI CO-CHAIR</b>	Grace Trembath and Jenna Courey
<b>PUBLICITY</b>	Jacob Schmieder
<b>MEDIA</b>	Zach Olsem
<b>REGISTRATION</b>	Sarah Grambo
<b>OPERATIONS</b>	Grace Trembath, Pahton McDonald, Kathryn Wittrock, Zach Olsem
<b>SPONSORSHIP</b>	Jared Sorenson and Ally Radermacher
<b>FUNDRAISING</b>	Liam Campin
<b>POSTER</b>	Matt Shafer
<b>T-SHIRT</b>	Richard Weerts
<b>FOOD</b>	Sarah Banta
<b>ADVISERS</b>	Desi Gunning and Gustavo MacIntosh



**First row:** Emily Boettger, Zachary Olsem, Ana DiSpirito, Liam Campin, Tulika Sharan; **Second row:** Kevin Beaghan, Richard Weerts, Sarah Grambo, Jared Sorenson, Patricia Gallardo; **Third row:** Madeline Farringer, Matthew Shafer, Desi Gunning (advisor), Courtney Olson, Pahton McDonald; **Fourth row:** Sarah Banta, Grace Trembath, Gustavo MacIntosh (advisor), Kathryn Wittrock, Jacob Schmieder; **Fifth row:** Jenna Courey, Prescott Jeckel; **Not pictured:** James Yegerlehner, Brandon Magnuson, Olivia Popovich, Charlie Beaver, Meghan Collett, Avani Laharia, Michelle Morford, Alexandra Radermacher, Stone Sinnett, Karen Romero-Bello, Ciaran Kelly, Braden Lewis

## 2020 STUPKA COMMITTEE



**First row:** Kayla Uthe, Jenna Courey, Matthew Palizzi, Pahton McDonald, Immaculate Edwin, Tulika Sharan, Patricia Gallardo; **Second row:** Audri Ruble, Sarah Banta, Sarah Zelle, Makayla Villela, Zach Olsem, Tiffany Farrell, Sarah Grambo, Jacqueline Ehrlich; **Third row:** Spydel Nardy, Rick Weerts, Alex Davis, Emily Boettger, Maddie Farringer, Laura Kurr; **Not pictured:** Tristin Baring, Kevin Beaghan, Brandon Buscher, Kathryn Butterfield, Liam Campin, Sam Catron, Meghan Collett, Justyne Crawford, Ana DiSpirito, Moises Garcia, Olivia Gray, Michelle Morford, Ally Radermacher, Jacob Schmieder, Matt Shafer, Jared Sorenson, Isaac Stine and Grace Trembath

<b>SYMPOSIUM CHAIRS</b>	Kayla Uthe and Jacqueline Ehrlich
<b>SECRETARY</b>	Spydel Nardy
<b>TREASURER</b>	Laura Kurr

### Sub-Committee Coordinators

<b>SPEAKER</b>	Makayla Villela and Maddie Farringer
<b>ALUMNI</b>	Pahton McDonald and Zach Olsem
<b>PUBLICITY</b>	Meghan Collett and Jacob Schmieder
<b>MEDIA</b>	Sarah Zelle
<b>REGISTRATION</b>	Emily Sarvis
<b>OPERATIONS</b>	Immaculate Edwin
<b>SPONSORSHIP</b>	Grace Trembath and Jared Sorenson
<b>FUNDRAISING</b>	Ana DiSpirito
<b>POSTER</b>	Patricia Gallardo
<b>T-SHIRT</b>	Audri Ruble
<b>FOOD</b>	Liam Campin
<b>VOLUNTEER</b>	Richard Weerts
<b>ADVISERS</b>	Desi Gunning and Gustavo MacIntosh



## REMEMBERING ROB



Rob Stupka was an undergraduate student majoring in biochemistry at Iowa State University. His passion for science and research led the effort to establish the BBMB Research Symposium. His unparalleled enthusiasm propelled the planning process forward. As chair, Rob led fellow students Tony Cyr, Claire Kruesel, Adam Krupicka, and Jordan Witmer in planning the first symposium for spring 2006. Their vision for this symposium was for it to be a medium for scientific research interactions between undergraduates and professionals. Rob also wanted this to be a place where undergraduate researchers could be recognized for their achievements in science. On November 30, 2005, Rob's life was tragically cut short after a pedestrian vehicle accident in front of the Molecular Biology Building. In tribute to Rob's drive and passion to create this event, the BBMB Undergraduate Symposium bears his name. BBMB undergraduates, who are inspired by Rob's story, continue to organize this event. We believe that through your attendance and participation, we continue to honor Rob's memory. The 2021 Stupka Undergraduate Research Symposium planning committee would like to thank you for being here. We hope you enjoy the wonderful research presented by our promising young scientists.

### LIPID TRANSFER PROTEIN

Rob and his father visited several universities in early 2002. In June of that year, they visited Iowa State University. The beauty of the campus and the friendliness of the staff convinced them that this was Rob's new home. Rob initially entered ISU in the fall of 2002 as a chemical engineering student. During his first semester, he took BBMB 101: Introduction to Biochemistry as an elective. Rob was so captivated by the biochemical discoveries explained in class that he became a biochemistry student at the end of his first semester. He worked on making cDNA libraries from floral nectaries in the Thornburg lab. This molecule is the nectary Lipid Transfer Protein (LTP2). The cDNA that encodes this protein was made by Rob. It was subsequently cloned and characterized. It is currently being expressed in bacterial cells with the goal of determining its antimicrobial activity.

## STUPKA LEADERSHIP AWARDEES

The Stupka Undergraduate Leadership Award shall recognize students who have demonstrated sustained leadership for the success of the Stupka Undergraduate Research Symposium and the Roy J. Carver Department of Biochemistry, Biophysics and Molecular Biology undergraduate program.

Adam Krupicka	Founding Member
Julia K. Nguyen	2018
Alyssa M. Lantz	2019
Samuel L. Tufts	2019
Laura A. Kurr	2020
Makayla Villela	2020
Emily Boettger	2021
Ana DiSpirito	2021
Grace Trembath	2021



## POSTER SESSION 1 (1:00-2:00)

1	<b>Jenna Courey</b>	Biochemical Characterization of Mycobacterium Tuberculosis MazG Enzyme
2	<b>Kathryn Wittrock</b>	A Structural Landscape of Neutralizing Antibodies Against SARS-CoV-2 Receptor Binding Domain
3	<b>Matthew Shafer</b>	Heterologous Expression of CSLC4 and XLT2 in Yeast for Structural Analyses
4	<b>Johanna Bailey</b>	Vismax™ Peptide: Field testing to improve citrus tree health under endemic disease pressure
5	<b>Dinitha Caldera</b>	Characterization of CRISPR RNA cleavage by a Cas5/6 fusion protein from <i>Methanosarcina barkeri</i>
6	<b>Charlie Beaver</b>	Heterologous expression of three Xylosyl Transferases in yeast to study the xyloglucan synthesizing complex
7	<b>Alexandra Radermacher</b>	Exploring the Function of Novel Proteins Targeted to the Malaria Parasite Vacuole
8	<b>Patricia Gallardo</b>	Role of isoflavones in the resistance of <i>Glycine max</i> against <i>Aphis glycines</i> Matsumura

## POSTER SESSION 2 (2:00-3:00)

9	<b>Paiton McDonald</b>	Effects of <i>Saccharomyces cerevisiae</i> fermentation product supplementation on the acute-phase response during bovine respiratory disease in neonatal calves
10	<b>Kathryn Butterfield</b>	Functional Analysis of Cas12 Proteins
11	<b>Liam Campin</b>	Exploring the Function of an Essential Inner Membrane Complex Protein in the Human Malaria Parasite <i>Plasmodium falciparum</i>
12	<b>Tulika Sharan</b>	Adaptation in Type II-C CRISPR-Cas systems
13	<b>Mark Schmidt-Dannert</b>	Single Residue Switches in Class I Diterpene Synthases
14	<b>Abigail Fowler Alexandra Dunnum</b>	Characterizing the growth of bone marrow-derived mesenchymal stem cells on composite polycaprolactone hydroxyapatite scaffolds
15	<b>Peyton Hamel</b>	Development of a human lung-on-a-chip platform
16	<b>Tung Mei Khu Grant Warren Juyoung Shin</b>	Disrupted in Schizophrenia 1 (DISC1) Associated with Arsenic Compounds is a Potential Response to Oxidative Stress

Undergraduate

Graduate

Alum

Stupka Scholar

Linder Research Fellow

## STUDENT SPEAKER

### Sarah Grambo

LINDER RESEARCH FELLOW



Sarah is a senior in biochemistry with minors in French and genetics. Sarah works in the lab of Dr. Gustavo MacIntosh on soybean-soybean aphid interactions. She is a learning assistant in the biochemistry department, Registration Chair for the Stupka Committee and a member of the BBMB Club. When she's not in the Molecular Biology Building, Sarah enjoys dancing, reading, and gardening. She intends to pursue a Ph.D. and continue academic research in the future.

#### ABSTRACT:

##### **Accumulation of aphid secretions changes the cuticular surface of the soybean plant**

The epidermis of soybean (*Glycine max*) leaves produces a cuticle made of lipids such as fatty acids, hydrocarbons, and various waxes that prevent water loss and solute intake. Previous studies have shown that the presence of soybean aphids (*Aphis glycines* Matsumura) causes changes in the genetic expression of the plant, specifically in the genes suspected to be responsible for wax production. To investigate the role of these cuticle lipids, we profiled the lipids present in the cuticular layer after aphid feeding. The cuticle lipids were extracted and then analyzed using GS-MS. The results showed a significant change in the quantity and composition of soybean cuticle lipids with the introduction of aphids. The most significant change was the presence of triacylglycerides (TAGs) only on aphid-treated plants. After comparing our results to previous studies with different aphid species in the literature, the appearance of TAGs can be attributed to aphid secretions rather than changes in the lipids produced by the soybean plant. Removal of the aphid-derived TAGs from our analysis revealed an increase in cuticle lipids produced by the soybean plant, especially fatty acids. Our results suggest that aphids may use lipid secretions to make the host plant more suitable for habitation or to signal an adequate site for further infestation.

## ALUM SPEAKER

### Mariah Hoyer, Ph.D.

DUKE UNIVERSITY MEDICAL CENTER



Mariah received her B.S. in biochemistry from Iowa State University in 2012. While at Iowa State, Mariah worked in the laboratory of Dr. Ravindra Singh, which studies the alternative splicing mechanism of the *Survival Motor Neuron 2* pre-mRNA and its association with Spinal Muscular Atrophy. Mariah completed her Ph.D. at Washington University in Saint Louis, under the direction of Timothy Miller, MD, Ph.D. Mariah's doctoral work was focused on identifying motor

neuron-enriched microRNAs and their relationship to the motor neuron disease Amyotrophic Lateral Sclerosis (ALS). Mariah's doctoral work resulted in the development of a pharmacodynamic biomarker for ALS and other motor neuron diseases, and the identification of a novel mechanism of microRNA-mediated communication between motor neurons and astrocytes. In June 2018, Mariah began her postdoctoral work in the laboratory of Dr. Debra Silver. The focus of Mariah's work has been elucidating the role of DDX3X, a new intellectual disability gene, in neural progenitors and neurons during cortical development. Mariah hopes to one day have her own lab studying defects in RNA metabolism in neurological disorders and to develop new therapies for these devastating diseases.

#### ABSTRACT:

##### **The role of an intellectual disability gene, *DDX3X*, in regulating translation required for neural progenitor fate decisions during brain development**

Mutations in the RNA helicase *DDX3X* are associated with a wide range of developmental deficits and brain malformations and account for 1-3% of intellectual disability (ID) cases in females. We recently found that *DDX3X* is required for cortical development in mice and that loss of *Ddx3x* impairs neural progenitor proliferation and their propensity to form neurons. However, the distinct requirements for *DDX3X* in neural progenitors and neurons have not been characterized. Using a conditional *Ddx3x* (cKO) mouse model, we are disentangling the distinct consequences of *Ddx3x* depletion in neural progenitors and neurons during cortical development. Our preliminary data indicates *Ddx3x* depletion from neural progenitors and their neuronal progeny leads to more progenitors and less neurons. Because *DDX3X* is an RNA helicase with known roles in translation regulation, we hypothesize that *DDX3X* controls neural progenitor fate decisions through regulation of translation. We are currently employing ribosome footprinting to identify *DDX3X* translational targets relevant for progenitor proliferation in *Ddx3x* cKO mice. These studies will enhance our understanding of RNA regulation required for normal brain development and the mechanism by which aberrations in translation can lead to ID.

## KEYNOTE SPEAKER

### Jeff Karp, Ph.D.

HARVARD MEDICAL SCHOOL



Dr. Jeff Karp is a Professor of Medicine at Brigham and Women's Hospital, Harvard Medical School. He is also a principal faculty member at the Harvard Stem Cell Institute, and an affiliate faculty member at the Broad Institute and at the Harvard-MIT Division of Health Sciences and Technology. He works in the fields of drug delivery, medical devices, stem cell therapeutics, and tissue adhesives. He has published over 125 peer-reviewed papers, with >22,000 citations, and has given over 300 invited lectures. He has over 100 issued or pending national and international patents. Several technologies developed in his lab have led to multiple products currently in development or on the market and for the launch of eight companies that have raised over \$450 million in funding. Technologies that include high-tech skincare (Skintifique, products sold in pharmacies throughout EU), tissue adhesives (Tissium, EU Approval in 2017) and 3D printed biomedical devices, immunomodulation with biologically responsive materials (Alivio Tx), small molecule regenerative therapeutics (\$FREQ –NASDAQ), cannabinoid therapeutics (Molecular Infusions acquired by Suterra Wellness in 2019), biomedical devices to improve child safety (Landsdowne Labs), needles that automatically stop at their target (Bullseye Therapeutics), and a bioengineered luminal coating for controlled GI targeting (Altrix Bio). Karp has received >50 awards and honors. Most recently Jeff received the highest award from the Society For Biomaterials for innovation – the Clemson Award for Applied Research. Boston Magazine recognized Karp as one of 11 Boston Doctors Making Medical Breakthroughs. In addition to his research goals, Karp is dedicated to developing the careers of the next-generation bioengineers at the forefront of regenerative medicine. He was selected as the Outstanding Faculty Undergraduate Mentor among all faculty at MIT and he received the HST McMahon Mentoring award for being the top mentor of Harvard-MIT students. To date, 22 trainees from his laboratory have secured faculty positions.

#### ABSTRACT: Towards Accelerated Medical Innovation

When developing technologies to solve medical problems, often one encounters significant hurdles, that at times seem insurmountable. Overcoming these hurdles requires new ways of thinking. One approach is to turn to nature for inspiration. Millions and millions of years of research and development at our fingertips, and all we need to do is look outside to the amazing creatures that inhabit our planet. This talk will explore medical technologies being developed that harness lessons from nature for inspiration, from creatures such as geckos, spider webs, jellyfish, porcupine quills, snails, to spiny headed worms. Another approach is radical simplicity – the art and discipline of reducing a problem to its essence. This tool has been harnessed to develop a new skin care approach that is advancing towards global market adoption, and therapeutic strategies to combat inflammatory bowel disease and arthritis that are advancing towards clinical studies. Some of the technologies that will be described are rapidly advancing to the clinic and some are already on the market helping patients. This talk opens new paths to the continual innovation that is so important in our fast-changing world.

## STUDENT SPEAKER

### Richard Weerts

STUPKA SCHOLAR 2021



Richard is a senior in biochemistry with minors in Spanish, French, and computer science. He is the current president of the BBMB Club, as well as T-shirt Chair for Stupka. He has worked in Dr. Olga Zabolina's lab for the last two years, researching the biosynthesis of xyloglucan in *Arabidopsis*. Outside of his lab, he can be frequently seen rock climbing, tutoring, or DJing for the ISU college radio station. After graduation, Richard is planning on going to medical school and studying neurology.

#### ABSTRACT:

##### Xyloglucan xylosyltransferase 1 displays promiscuity toward donor substrates during *in vitro* reactions

Glycosyltransferases (GTs) are a large family of enzymes that add sugars to a broad range of acceptor substrates, including polysaccharides, proteins, and lipids, by utilizing donor substrates in the form of activated sugars. Individual GTs have been considered to exhibit a high level of substrate specificity, but this has not been thoroughly investigated across the extremely large set of different GTs. Here we investigate Xyloglucan Xylosyltransferase 1 (XXT1), a GT involved in synthesis of plant cell wall xyloglucan. Xyloglucan has a glucan backbone with the first side chains exclusively xylose residues from UDP-Xylose. While these conserved substitution patterns suggest a high substrate specificity for XXT1, our *in vitro* kinetic studies find a more complex set of behavior. Reactions using UDP-Xylose, UDP-Arabinose, UDP-Glucose and UDP-Galactose demonstrate  $k_{cat}$  values for reactions with UDP-Xylose and UDP-Glucose being comparable, while reactions with UDP-Arabinose and UDP-Galactose are over 10-fold slower. If  $k_{cat}/K_M$  is a measure of efficiency, then UDP-Xylose is 8-fold more efficient as a substrate than the next best alternative, UDP-Glucose. In conclusion, we demonstrate for the first time that plant XXTs are not all highly substrate specific, and some do show significant promiscuity in their *in vitro* reactions. Kinetic parameters alone are not likely to fully explain the high substrate selectivity *in planta*, suggesting there may be additional control mechanisms operative during polysaccharide biosynthesis. The improved understanding of substrate specificity of the GTs will aid in protein engineering, development of diagnostic tools, and understanding of many biological systems.



## ALUM SPEAKER

### Jennifer Gribble

VANDERBILT UNIVERSITY



Jennifer is from Cedar Rapids, Iowa and in 2016, obtained her B.S. in biochemistry from Iowa State University with minors in genetics and microbiology. She joined the Interdisciplinary Graduate Program (IGP) at Vanderbilt University in Nashville, Tennessee in 2016, and in 2017, joined the lab of Dr. Mark Denison in the Ph.D. program of Microbe-Host Interactions. Her work in the Denison Lab has focused on coronavirus replication, with particular emphasis on RNA synthesis and viral recombination during both normal viral infection and in viral inhibition through either genetic attenuation or antiviral treatments. She has established multiple computational pipelines to study coronavirus RNA mutations and recombination, combining both traditional virological approaches with next-generation RNA sequencing platforms. Jennifer lives with her husband, David, in Smyrna, Tennessee, and in her free time enjoys traveling, painting, and baking for her lab.

#### ABSTRACT:

##### RNA recombination in coronaviruses

Recombination is proposed to be critical for coronavirus (CoV) diversity and emergence of SARS-CoV-2 and other zoonotic CoVs. While RNA recombination is required during normal CoV replication, the mechanisms and determinants of CoV recombination are not known. CoVs encode an RNA proofreading exoribonuclease (nsp14-ExoN) that is distinct from the CoV polymerase and is responsible for high-fidelity RNA synthesis, resistance to nucleoside analogues, immune evasion, and virulence. We demonstrate that CoVs, including SARS-CoV-2, MERS-CoV, and the model CoV murine hepatitis virus (MHV), generate extensive and diverse recombination products during replication in culture. We show that the MHV nsp14-ExoN is required for native recombination, and that inactivation of ExoN results in decreased recombination frequency and altered recombination products. These results add yet another critical function to nsp14-ExoN, highlight the uniqueness of the evolved coronavirus replicase, and further emphasize nsp14-ExoN as a central, completely conserved, and vulnerable target for inhibitors and attenuation of SARS-CoV-2 and future emerging zoonotic CoVs.

## STUDENT SPEAKER

### Madeline Farringer

STUPKA SCHOLAR 2021

GOLDWATER SCHOLAR



Madeline is a biochemistry major with a minor in emerging global diseases who will graduate in Spring 2021. She is the Speaker Co-Chair for the Stupka Committee and the Secretary of the BBMB Club. She works in Dr. Josh Beck's lab developing a knockdown system to study the malaria parasite. In recognition of her outstanding academic career and contributions to research, she received the 2020 Goldwater Scholarship. After graduation, she plans to continue in parasitology research while pursuing a Ph.D. in biomedical sciences.

#### ABSTRACT:

##### Developing an auxin-inducible degron system for use in *Plasmodium falciparum*

Malaria is a devastating disease that causes nearly half a million deaths annually. While a deeper understanding of unique parasite biology is urgently needed to provide novel therapeutic approaches, functional genetic studies are challenging given the limited tools available. To bridge this gap, we are adapting the plant auxin-inducible degron (AID) to study the function of essential genes in *Plasmodium falciparum*, the deadliest human malaria parasite. In its endogenous context, the plant hormone auxin binds the AID sequence present in certain target proteins, enabling recognition by an F-box protein within the Skp, Cullin, F-box-containing protein complex (SCF). SCF then recruits an E2 ubiquitin ligase, leading to ubiquitination and rapid proteasomal degradation of the target protein. Due to the high conservation of SCF among eukaryotes, repurposing this system for knockdown in an exogenous context can be achieved by expression of an auxin-sensitive F-box protein and fusion of the AID sequence to a target protein. While the AID system has been successfully employed in related protozoan parasites, its capabilities in *P. falciparum* remain untested. We have developed AID systems in *P. falciparum* based on TIR1 and AFB2, two different auxin-sensitive F-box proteins, and are evaluating their capabilities with a panel of essential protein targets with diverse biochemical properties as well as a reporter. The successful establishment of the AID system will accelerate functional genetic studies in this devastating pathogen.

## KEYNOTE SPEAKER

### Elizabeth Sattely, Ph.D.

STANFORD UNIVERSITY



Elizabeth Sattely is an Associate Professor and HHMI Investigator in the Department of Chemical Engineering at Stanford and a Stanford ChEM-H Faculty Fellow. She also serves as an Honorary Adjunct Staff Scientist at the Carnegie Institution of Science. Dr. Sattely completed her graduate training at Boston College in organic chemistry and her postdoctoral studies in biochemistry at Harvard Medical School where she worked on natural product biosynthesis in bacteria. Inspired by

human reliance on plants and plant-derived molecules for food and medicine, the Sattely laboratory is focused on the discovery and engineering of plant metabolic pathways to make molecules that can enhance human and plant health. Work in the Sattely lab has been recognized by an NIH New Innovator Award, a DOE Early Career Award, an HHMI-Simons Faculty Scholar Award, a DARPA Young Investigator Award, and an AAAS Mason Award for Women in the Chemical Sciences.

#### ABSTRACT:

##### Total biosynthesis of plant-derived therapeutics

Molecules from plants play a critical role in human health as clinically-used therapeutics and dietary nutrients. Notably, 10% of the WHO essential medicines are plant natural products or derivatives (e.g., etoposide, taxol, digoxin, and vinblastine), and the food we eat contains abundant drug-like molecules from plants. Despite the major impact of plant chemistry on human health, numerous drugs are still isolated from difficult-to-cultivate native plants or plant cell culture and the chemistry of dietary crops is poorly understood and unoptimized. Engineering plant biosynthetic pathways is an exciting strategy for transforming how we use plant chemistry. The central challenge is identifying the genes that make up plant metabolic pathways without genetic tools or genome sequence in the producing plant. Our lab is focused on accelerating the discovery of both biosynthetic pathways in plants and plant natural product mechanism of action, and along the way we are developing new tools for engineering plant chemistry. In this talk, I will discuss recent advances in my lab that have allowed us to rapidly identify sets of enzymes that can be used to engineer the production of plant molecules of importance to human health.

## ALUM SPEAKER

### Luke Helgeson, Ph.D.

UNIVERSITY OF WASHINGTON, SEATTLE  
STUPKA SCHOLAR 2008



Luke Helgeson graduated from Iowa State University in 2009 with a B.S. in biochemistry. During his time at ISU, he performed undergraduate research in the laboratory of Dr. Gaya Amarasinghe, where he helped to determine the structure of an Ebola protein domain using X-ray crystallography. Luke was active with the BBMB Club during his four years at ISU, including serving as President, running and organizing multiple Stupka Undergraduate Research

Symposia, and flipping countless pancakes for the monthly Breakfast Club. He was the recipient of the 2009 Robert Stupka Memorial Scholarship. After graduating from ISU, Luke continued his training in biochemistry, completing his doctoral studies at the University of Oregon under the mentorship of Dr. Brad Nolen. While in the Nolen Lab, he published multiple articles on the regulation of branched actin nucleation. Luke was the 2014 recipient of the Pete von Hippel Award as an outstanding Senior Ph.D. student. Upon receiving his doctoral degree in 2014, Luke remained in the Pacific Northwest, joining the laboratory of Dr. Trisha Davis at the University of Washington as a postdoctoral fellow. At the University of Washington, Luke researches the mechanisms of how kinetochore microtubule attachments are strengthened and maintained during chromosome segregation.

#### ABSTRACT:

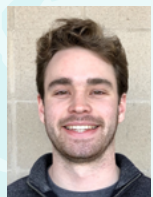
##### Mechanisms of Load Bearing within the Kinetochore

During mitosis, chromosomes are segregated via attachment to dynamic microtubule ends that assemble and disassemble to move the chromosomes around the cell. The kinetochore is an assembly of proteins and protein complexes that forms this attachment from chromosomes, through the centromere, to microtubule ends. Kinetochore to microtubule-end interactions must be strong enough to sustain the generated forces from movement, be weak enough to release when not needed, and be fluid enough to track with both growing and shrinking microtubule ends. Weakening or overly strengthening kinetochore microtubule attachments can lead to mitotic delay and chromosome mis-segregation and instability. These erroneous mitotic outcomes have been implicated in cell death, developmental defects, and tumor formation. Significant progress has been made in finding the proteins that compose the kinetochore and its interface with microtubules. However, the mechanisms of how multiple kinetochore components coordinately interact with each other and dynamic microtubules ends in order to strengthen and maintain attachments remains unknown. To understand these vital kinetochore microtubule interactions I use single-molecule TIRF microscopy to test binding stoichiometry and dynamics, crosslinking coupled with mass spectrometry to find protein-protein interactions, and optical tweezers microscopy to directly test microtubule end attachment strength. Together, these studies have allowed me to better understand how assemblies of protein-protein interactions between kinetochore components and microtubules form the load-bearing attachments that are necessary for correct chromosome segregation.

## STUDENT SPEAKER

### Jacob Schmieder

STUPKA SCHOLAR 2021



Jacob is a senior majoring in biochemistry with a physics minor. He is a member of the Stupka Committee and has spent the last two years serving as the Publicity Chair. Jacob has been a part of Dr. Claussen's lab since the fall of 2018, where he has spent most of his time working on enzymatic sensors, specifically for on-body sensing. In 2020, he was chosen as a Stupka Scholar. After graduation, Jacob will spend a year continuing his research in Dr. Claussen's lab, working towards a

Master's degree in mechanical engineering. He then hopes to pursue an MD/Ph.D. in biomedical engineering.

#### ABSTRACT:

##### **Wearable Flexible Enzymatic Sensors using Graphene**

Laser induced graphene is a new material with many applications, particularly in flexible electronics. Along with other novel 2D materials, LIG has been investigated as a basis for wearable sensing. Sweat represents a promising biological medium to sense chemicals such as glucose and lactate and gain insight for relevant medical and athletic applications. Herein, LIG is functionalized with oxidase enzymes and selective polymer membranes to achieve biosensors tailored to measuring glucose across physiological ranges in sweat. The LIG biosensors were evaluated in buffers, artificial sweat, and integrated into wearable high sampling resolution tape microfluidics.

## ALUM SPEAKER

### Jacqueline Rivas, Ph.D.

UNIVERSITY OF KENTUCKY

STUPKA SCHOLAR 2011



Jackie is a Post-Doctoral Fellow in the Markey Cancer Center at the University of Kentucky under Dr. Subbarao Bondada. As an undergraduate, she worked with Dr. Amy Andreotti (2009-2012) researching interactions between proteins downstream of the T cell receptor. During this time Jackie participated in multiple SURS, and was the chair in 2011-2012. She obtained her Ph.D. in Immunology from UT Southwestern Medical Center with Dr. Nancy Monson

(2012-2017), studying B cell responses in early Multiple Sclerosis. Here she discovered that patients experiencing their first clinical symptoms in adulthood display humoral autoimmunity to neurons, while those with pediatric onset show preference for astrocytes. Now as a post-doctoral researcher, Jackie is sponsored by a fellowship from the Leukemia and Lymphoma Society to study methods of enhancing antitumor immunity in B cell chronic lymphocytic leukemia. She discovered that immunosuppressive cytokines can be targeted to improve T cell anti-tumor immunity and responses to immunotherapy, and plans to continue pursuing research on the anti-tumor effects of T cells in an academic career.

#### ABSTRACT:

##### **Enhancing T-cell antitumor immunity in Chronic Lymphocytic Leukemia**

T-cell dysfunction is a hallmark of B-cell Chronic Lymphocytic Leukemia (CLL), where CLL cells downregulate T-cell responses through regulatory molecules including programmed death ligand-1 (PD-L1) and Interleukin-10 (IL-10). Immune checkpoint blockade (ICB) aims to restore T-cell function by preventing the ligation of inhibitory receptors like PD-1. However, most CLL patients do not respond well to this therapy. Thus, we investigated whether IL-10 suppression could enhance antitumor T-cell activity and responses to ICB. Since CLL IL-10 expression depends on Sp1, we utilized a novel, better tolerated analogue of the Sp1 inhibitor mithramycin (MTM<sub>ox</sub>32E) to suppress CLL IL-10. MTM<sub>ox</sub>32E treatment inhibited mouse and human CLL IL-10 production and maintained T-cell effector function *in vitro*. In the Eμ-Tcl1 mouse model, treatment reduced plasma IL-10 and CLL burden and increased CD8<sup>+</sup> T-cell proliferation, effector and memory cell prevalence, and interferon-γ production. When combined with ICB, suppression of IL-10 improved responses to anti-PD-L1 as shown by a 4.5-fold decrease in CLL cell burden compared to anti-PD-L1 alone. Combination therapy also produced more interferon-γ<sup>+</sup>, cytotoxic effector KLRG1<sup>+</sup>, and memory CD8<sup>+</sup> T-cells, and fewer exhausted T-cells. Since current therapies for CLL do not target IL-10, this provides a novel strategy to improve the efficacy of T-cell-based immunotherapies.



## KEYNOTE SPEAKER

### Gaya K. Amarasinghe, Ph.D.

WASHINGTON UNIVERSITY, ST. LOUIS



Dr. Amarasinghe is a professor at Washington University School of Medicine in St. Louis. He received his Bachelor of Science degree in chemistry from the City College of New York in 1997. From the summer of 1994 through fall 1996, Dr. Amarasinghe also worked as a Research Assistant in the laboratory of Professor David Cowburn at the Rockefeller University in New York, where he gained a keen interest in mechanisms that control signal transduction. Dr. Amarasinghe conducted his thesis studies with HHMI Investigator Dr. Michael Summers at the University of Maryland, Baltimore County, where he characterized the structural basis for HIV-1 genome packaging by nuclear magnetic resonance-based methods. From 2001-2007, Dr. Amarasinghe conducted his postdoctoral research at the University of Texas, Southwestern Medical Center at Dallas, to study signaling and structural dynamics in the multidomain Vav proto-oncoprotein under a fellowship from the Cancer Research Institute. Dr. Amarasinghe began his independent research program in 2007 in the Biochemistry, Biophysics and Molecular Biology Department at Iowa State University. At Iowa State University, Dr. Amarasinghe also served as the faculty advisor to the BBMB Club and the Stupka Symposium. In the fall of 2011, Dr. Amarasinghe moved to the Washington University School of Medicine in St. Louis, where he is currently a Professor of Pathology and Immunology, of Biochemistry and Molecular Biophysics, and of Molecular Microbiology. The Amarasinghe group uses hybrid structural methods along with biochemistry, cell biology, and virology to determine how microbial infections impact host signaling.

#### ABSTRACT:

##### **Molecular Mechanisms of Viral Immune Evasion**

Viruses use a variety of immune evasion mechanisms, including restriction of cell-intrinsic and cell-extrinsic signaling, to counter host responses. These evasion mechanisms contribute to unrestricted viral replication, which contributes to pathogenesis and disease. While cell-intrinsic control of host responses often involves virally encoded components blocking recognition of pathogen associated molecular patterns (PAMPs), the pathways that antagonize host responses are also not yet completely understood for many human and animal pathogens. Work from many groups, including our own studies, highlight the complexities of the arms race at this host-pathogen interface. Germline encoded immune receptors serve as a first line of defense and are responsible for identifying PAMPs. In countering viral pathogens, RNA PAMP recognition plays a critical role in activating host responses. Structural mechanisms of host receptors, such as RIG-I, MDA-5, and IFIT proteins, suggests that PAMP recognition is a complex process that functions to differentiate self from non-self RNA. I will discuss molecular mechanisms of immune evasion by pathogenic viruses such as Venezuelan equine encephalitis virus (VEEV), a new world alpha virus, and hemorrhagic fever viruses, such as Ebola virus and Marburg virus. Using a combined biochemical and structural approach coupled with focused cellular and virological studies, we continue to dissect how the activity of viral nucleic acids and proteins counter immune signaling and disarm host responses. I will also discuss implications for viral infections in vitro and in vivo, their impact of infection and disease, as well as lessons from these studies that support target identification for antiviral development.

## STUDENT SPEAKER

### Behnia Rezazadeh Shirazi

GOLDWATER SCHOLAR



Behnia Rezazadeh Shirazi is a senior triple majoring in biology, biophysics, and biochemistry. He has held several leadership positions throughout his time at Iowa State University, including serving as president of the ISU UNICEF chapter, working as a Community Advisor, and serving on the Health and Wellness and the Diversity and Inclusion Committees for student government. His research at the Hemodynamics Lab in the Department of Kinesiology is under the supervision of Dr. James Lang where he studies microvascular function. In light of these accomplishments, Behnia was recognized with the 2020 Goldwater Scholarship. After he graduates in Spring 2021, Behnia will spend a summer at Duke School of Medicine researching drug discovery and networking with scientists working in biotech/pharma companies in Durham, North Carolina. Afterwards, Behnia will serve as a research fellow at the U.S. National Institute of Health's Vaccine Research Center where he will work on HIV vaccine development. Later, he plans to earn a Ph.D. in biological engineering and an MBA. His ultimate goal is to start a biotechnology company that focuses on environmental issues or medical therapeutics after graduate school.

#### ABSTRACT:

##### **Reproducibility and Normalization of Reactive Hyperemia using Laser Speckle Contrast Imager**

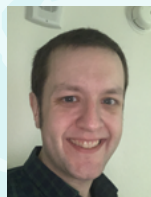
Impaired perfusion indices signal potential microvascular dysfunction preceding atherosclerosis and other cardiometabolic pathologies. Post-occlusive reactive hyperemia (PORH), a vasodilatory response following a mechanically induced ischemia, is characterized by increased perfusion index and can be used to assess microvascular dysfunction. Knowledge of the reproducibility and standardization of such perfusion indices is limited. Our aim was to study the inter- and intra-day reproducibility and standardization of reactive hyperemia using the full-field Laser Speckle Contrast Imaging (LSCI) technique. Seventeen healthy adults (age =  $24 \pm 3$  years) completed three PORH bouts over two lab visits. LSCI region of interest was a standardized 10 cm region on the dominant ventral forearm. A 5-min brachial artery occlusion period induced by inflating an arm cuff to 200 mmHg, preceded a 4-min hyperemic period. Inter- and intra-day reliability and reproducibility of cutaneous vascular conductance (LSCI flux / mean arterial pressure) were determined using intraclass correlation (ICC) and coefficient of variation (CV%). Maximal flow and area under the curve standardized to zero perfusion showed intra- and inter-day reliability (ICC > 0.70). Biological zero is a reliable reference point for standardizing the amplitude, the area under the hyperemic curve, and the overshoot rate-of-change of the PORH response. Time to maximal flow is not reproducible (inter-day CV = 18%). An alternative index for calculating hyperemic velocity (i.e. overshoot rate-of-change) represented as a piecewise function (at 5s, 10s, 15s, and 20s into hyperemia) was reproducible (CV < 11%). PORH measured with LSCI is a reliable assessment of microvascular function. However, not all indices and normalization methods are reliable or recommended for longitudinal assessment.



## ALUM SPEAKER

### Tony Cyr, MD/Ph.D.

UNIVERSITY OF PITTSBURGH



Tony Cyr is a 2006 graduate of Iowa State University from the Department of Biochemistry, Biophysics and Molecular Biology. His initial research experience involved utilizing metabolic engineering approaches to produce complex diterpenoid molecules in a bacterial host. He subsequently pursued a combined MD/Ph.D. degree at the University of Iowa, in the laboratories of Dr. Rick Domann and Dr. Ron Weigel.

There, he studied the contributions of oxidative metabolism to epigenetic signaling processes, as well as the role of AP-2 transcription factors in the pathogenesis of breast cancer. In 2014, he successfully matched into the general surgery residency program at the University of Pittsburgh Medical Center, where he continues to pursue his surgical training. In addition, he has completed a three year research fellowship in the laboratory of Dr. Brian Zuckerbraun studying the contributions of aging, mitochondrial metabolism, and metabolomic flux to the pathogenesis of immunosuppression in the post-trauma period. Notably, he worked with former Stupka chair Luran Chambers extensively during this time, as she was recruited to work in the lab prior to going to medical school herself in 2020. He now has resumed clinical work for the final two years of surgical training. Upon finishing his residency, he plans on pursuing career that allows the best combination of basic science pursuit with clinical practice.

#### ABSTRACT:

##### **Novel Circulating Sphingolipid Signatures in the Plasma Metabolome are Associated with Better Outcomes Following Blunt Trauma**

Trauma is the leading cause of death and disability for individuals under age 55. Many severely injured trauma patients experience a complicated clinical course despite appropriate initial therapy. Here, we sought to identify novel circulating metabolomic signatures associated with clinical outcomes following blunt trauma. Untargeted metabolomics and circulating plasma immune mediator analysis was performed on plasma collected during three post-injury time periods (<6h, 6h-24h, D2-D5) in 86 critically ill trauma patients, enrolled between April 2004 and May 2013 at UPMC Presbyterian Hospital in Pittsburgh, PA. Inclusion criteria were age 18 years, blunt mechanism, intensive care unit admission, and expected survival 24 h. Exclusion criteria were isolated head injury, spinal cord injury, and pregnancy. *Post hoc* endpoints included length of stay (overall and intensive care unit), ventilator requirements, nosocomial infection, and Marshall organ dysfunction (MOD) score. The top 50 metabolites were isolated using repeated measures ANOVA and multivariate empiric Bayesian analysis for further study. Sphingolipids were enriched significantly ( $\chi^2$ ,  $p < 10^{-6}$ ) among this group. Clustering of sphingolipid patterns discerned 3 patient subclasses: (1) non-responders (defined as no time-dependent change in sphingolipids), (2) sphingosine/sphinganine-enhanced, and (3) glycosphingolipid-enhanced. Compared to the sphingolipid enhanced subclasses, non-responders demonstrated higher lengths of stay, more ventilator days, higher MOD scores, and higher circulating levels of proinflammatory immune mediators IL-6, IL-8, IL-10, MCP1/CCL2, IP10/CXCL10, and MIG/CXCL9 (all  $p < 0.05$ ), despite similar injury severity scores ( $p = 0.117$ ). Untargeted metabolomic analysis identified significant alterations in circulating plasma sphingolipids following severe injury. Specific circulating sphingolipid signatures and their correlation with both clinical outcomes and circulating inflammatory mediators raise the possibility of a link between sphingolipid metabolism and the immune response to trauma.

STUPKA 2021 UNDERGRADUATE RESEARCH SYMPOSIUM

## ALUM SPEAKER

### Tyler Gilbreath

UNIVERSITY OF NEBRASKA MEDICAL CENTER



Tyler is from Des Moines, Iowa and graduated in the spring of 2017 with a B.S. in agricultural biochemistry and microbiology from Iowa State University. He joined the Molecular Genetics and Cell Biology doctoral program at the University of Nebraska Medical Center in the fall of 2017. He is currently in Dr. Shannon Buckley's lab at UNMC working on how truncation mutations within the ubiquitin E3 ligase *UBR5* are related to Mantle Cell Lymphoma. Notably, he has helped discover novel *UBR5* interaction partners that are components of the spliceosome. Tyler lives with his wife Jori and enjoys spending time with her and their two dogs.

#### ABSTRACT:

##### **The Role of the E3 Ubiquitin Ligase *UBR5* in B Cell Development and MCL Pathogenesis**

Mantle Cell Lymphoma (MCL) is a non-Hodgkin's lymphoma (NHL) that typically affects older adults. In MCL, there is uncontrolled growth of mature naïve B cells within the mantle zone of lymph node and spleen germinal centers. Mutations starting and accumulating in early B cell progenitors are thought to be ultimately responsible for MCL development. Although MCL only represents ~5% of NHL patients, it has the poorest survival rate among NHL sub-types due to a lack of successful therapeutic treatments. As such, it is important to identify potential targets for treatment. Recently published data shows that in 18% of all MCL patients have a mutation in the gene encoding the E3 ubiquitin ligase, *UBR5*. *UBR5* is part of the ubiquitin proteasome system, a degradation/recycling pathway in which proteins are tagged with a ubiquitin protein and degraded by the proteasome. Of the identified *UBR5* mutations, over 60% of the mutations are truncations at the carboxy terminus that cut off the cysteine residue linked to ubiquitin transfer in exon 59 suggesting a catalytic dead mutant protein is produced. Interestingly, *UBR5* mutations are specific to MCL and are not found in other sub-types of NHL. By studying *UBR5*, we can determine the role of *UBR5* mutations in MCL, elucidate molecular mechanism(s) of *UBR5* in B cell development, and identify potential therapeutic targets. In order to study the role of *UBR5* in B cell development and MCL, we generated a conditional mouse model targeting exon 58 of *Ubr5* similar to mutations in MCL patients and crossed the mice with *Mb1<sup>CRE</sup>* mice to delete exon 58 specifically in B cells at the pro-pre B cell stage of B cell development (*Ubr5<sup>ΔE58</sup>*). *Ubr5<sup>ΔE58</sup>* mice shows that mice lacking the carboxy terminus of *UBR5* have a block in B cell differentiation at the transitional B cell *IgM<sup>+</sup>IgD<sup>low</sup>* to mature B cell *IgM<sup>+</sup>IgD<sup>+</sup>* transition. *Ubr5<sup>ΔE58</sup>* mice showed a marked decrease of mature *IgM<sup>+</sup>IgD<sup>+</sup>* B cells in the bone marrow and spleen. Mass spectrometry comparing mouse B220+ splenocytes in *Ubr5<sup>ΔE58</sup>* compared to *Ubr5<sup>WT</sup>* mice showed that *UBR5* in B cells in *Ubr5<sup>ΔE58</sup>* mice has over two-fold more expression. Comparing this mass spectrometry with previous endogenous *UBR5* immunoprecipitation coupled with mass spectrometry has identified five novel interacting proteins that are involved in alternative splicing of RNA. Additionally, *IgD*, a protein made from the alternative splicing of the heavy chain gene locus, expression is significantly decreased. Coupled together, this suggests that the loss of the carboxy terminus of *UBR5* impedes B cell maturation by impacting alternative splicing and increased *UBR5* substrate concentrations. These studies aim to identify the role of *UBR5* in the context of normal B cell development and lymphomagenesis with the goal of identifying therapeutic targets for drug discovery.

## Samson Condon, Ph.D.

UNIVERSITY OF WISCONSIN, MADISON  
STUPKA SCHOLAR 2012



Samson Condon is a postdoctoral research associate in the Department of Biochemistry at the University of Wisconsin-Madison. He graduated from Cedar Falls High School in 2009 and began attending Iowa State University in the Roy J. Carver Department of Biochemistry, Biophysics and Molecular Biology that fall. Samson joined the Nikolau Group as an undergraduate researcher in 2010 under the mentorship of Dr. Marna Yandea-Nelson. There, he analyzed the cuticular wax composition of maize silks using gas chromatography-mass spectroscopy and developed new protocols to increase sample throughput. He spent the summer of 2010 at SUNY-Albany as an REU intern in the laboratory of Dr. Janice Pata and the spring of 2012 at the Nihon University School of International Relations in Mishima, Japan through the ISEP study-abroad program. He graduated summa cum laude with Honors from Iowa State University in 2013 with a B.S. in biochemistry. For his involvement in academics, research, and the BBMB Club, he was named a Goldwater Scholar and a Stupka Scholar.

Samson joined the laboratory of Professor Alessandro Senes at the University of Wisconsin-Madison in 2013. During his studies, he received the support of the Computation and Informatics in Biology and Medicine (CIBM) traineeship. He defended his thesis, titled "Understanding membrane protein association through molecular modeling and evolution", in the spring of 2020. He has continued his research as a postdoc in the Senes lab, combining bioinformatics, molecular modeling, and biochemistry to better understand how membrane proteins fold, interact, and evolve.

### ABSTRACT:

#### Exploring the Sequence Landscape of a Highly Dimeric Transmembrane Helix

Transmembrane helix oligomerization is a notable and often functionally important feature of many membrane proteins. Better understanding of the sequence requirements for oligomerization can lead to insight into membrane protein folding. The transmembrane helix of the mitophagy receptor BNIP3 dimerizes with particularly high affinity due to tight interhelical packing driven by a GxxxG sequence motif as well as a set of interhelical hydrogen bonds. These sequence features are highly conserved in BNIP3 homologues, but surprisingly, their locations within the transmembrane helix are not. The impact of this shift on dimerization as well as how it occurred through the course of BNIP3's evolutionary history are unclear. We explore the sequence landscape of BNIP3, examining what features of its interface are critical for dimeric stability. Using ancestral sequence reconstruction and *de novo* structure prediction, we demonstrate that the helix-helix crossing point of the BNIP3 dimer shifted, but this shift occurred in a manner that preserved its dimerization. Additionally, we shuffled the interfacial sequences between human and nematode BNIP3 to better understand the relative importance of the GxxxG motif and hydrogen bonding region on BNIP3 oligomerization. Structural and energetic prediction of these shuffled variants indicate that the location of the helix-helix crossing point may have a strong influence on self-association. BNIP3 dimers that cross near the middle of the membrane bury more surface area than dimers with C-terminal crossing points, resulting in more favorable energy scores. These results contribute to our understanding of how individual transmembrane helices can associate so strongly and how such association may be preserved and changed through evolution.

## Denis Tamiev

IOWA STATE UNIVERSITY  
STUPKA SCHOLAR 2014



Denis Tamiev graduated in 2016 with a degree in biochemistry from the Roy J. Carver Department of Biochemistry, Biophysics and Molecular Biology at Iowa State University. During his undergraduate years, he worked in the laboratory of Dr. Mark Hargrove studying anaerobic nitrogen metabolism in *E. coli*. Denis participated in multiple Stupka Undergraduate Research Symposia and was selected as a Stupka Scholar in 2014. Since completing his undergraduate degree, Denis is pursuing a Ph.D. at Iowa State University in the laboratory of Dr. Nigel Reuel. Denis' research focuses on developing spore based protein expression systems that can be used in locations with limited cold storage capacity for therapeutic applications. During his graduate research work, Denis developed an artificial intelligence based image processing algorithm to count bacterial cells on microscope images. This work enabled him to create a startup, Curiosity Labs, that Denis is currently managing while pursuing his Ph.D. degree.

### ABSTRACT:

#### Deep Learning to Improve Bacterial Cell Counting – Implementation of Classification-Type Convolutional Neural Networks (CNN)

Accurate classification and counting of bacterial cells is important in various microbiome assessment experiments. Most common methods rely on flow cytometry or manual microscopy. Application of flow cytometers in bacterial cell typing is limited due to cell size; bacteria are up to 1000 times smaller than eukaryotes. This leads to misclassification of multicellular clumps (biofilms), and cell debris for single cells. Automation of manual microscopy can be achieved with software. Most common software solutions for microscope image processing such as imageJ cannot accurately parse multicellular clumps into single cells or classify them into bacterial cell types (cocci, bacilli, spirilla, spores, etc.). Recent progress in the field of big data and deep learning can be used to develop a classification type Convolutional Neural Network (CNN) tool for bacterial cell counting. This presentation focuses on describing a custom CNN algorithm that identifies bacterial cell clusters on microscope images, and classifies them based on the number of cells found in each cluster. Here, we developed a robust image preprocessing algorithm that improves the neural network's confidence in classifying bacterial clusters by 13.96%. This image preprocessing algorithm increases the size of the training database through image augmentation by 72 times. Such augmentation allowed us to use a very small image dataset, and increased CNN's accuracy by 13.83%. In this work, we also compare our algorithm to existing techniques and human counting performance. We conclude that classification type CNN can serve as a reliable tool for bacterial cell counting and sorting, and can be leveraged by groups that possess a simple camera-enabled microscope setup.

## 2021-2022

### Liam Campin



Liam is a junior majoring in biochemistry with a minor in global health. He is currently the Fundraising Chair for the Stupka Committee, Breakfast Club Chair for BBMB Club, and a peer mentor for the BBMB Learning Community. He is a research assistant in Dr. Joshua Beck's lab, where he studies proteins in the parasitic vacuole of malaria parasites. After graduation, he plans on attending grad school and furthering his career in biochemistry.

### Paiton McDonald



Paiton is a junior majoring in agricultural biochemistry and international agriculture with a minor in pharmacology and toxicology. She is the Treasurer for the Stupka Committee but has served as the Alumni and Newsletter Co-Chair in the past. Paiton has worked in Dr. Jodi McGill's lab since 2019. Currently, she is studying the acute-phase response in the bovine liver during bovine respiratory disease. Her work led to recognition as a 2021 Goldwater Scholar. She plans to continue in livestock disease research while pursuing a Ph.D. in immunology.

### Tulika Sharan



Tulika Sharan is a junior majoring in biochemistry with a minor in genetics. She is a research assistant in Dr. Dipali Sashital's lab, where she studies interference and adaptation mechanisms in Type II-C CRISPR-Cas systems. She is currently serving as Speaker Co-Chair of the Stupka Symposium Planning Committee, is an International Student Ambassador, as well as a B&B Learning Community peer mentor. After graduating from Iowa State, Tulika plans to go to graduate school and further her career in Biochemistry.

### Richard Weerts



Richard is a senior in biochemistry with minors in Spanish, French, and computer science. He is the current president of the BBMB Club, as well as T-Shirt Chair for Stupka. He has worked in Dr. Olga Zabotina's lab for the last two years, researching the biosynthesis of xyloglucan in Arabidopsis. Outside of his lab, he can be frequently seen rock climbing, tutoring, or DJ-ing for the ISU college radio station. After graduation, Richard is planning on going to medical school and studying neurology.

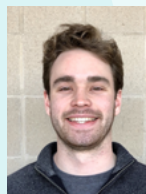
## 2020-2021

### Madeline Farringer



Madeline is a biochemistry major with a minor in emerging global diseases who will graduate in Spring 2021. She is the Speaker Co-Chair for the Stupka Committee and the Secretary of the BBMB Club. She works in Dr. Josh Beck's lab developing a knockdown system to study the malaria parasite. In recognition of her outstanding academic career and contributions to research, she received the 2020 Goldwater Scholarship. After graduation, she plans to continue in parasitology research while pursuing a Ph.D. in biomedical sciences.

### Jacob Schmieder



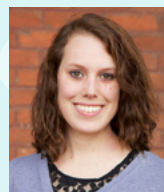
Jacob is a senior majoring in biochemistry with a physics minor. He is a member of the Stupka Committee and has spent the last two years serving as the Publicity Chair. Jacob has been a part of Dr. Claussen's lab since the fall of 2018, where he has spent most of his time working on enzymatic sensors, specifically for on-body sensing. In 2020, he was chosen as a Stupka Scholar. After graduation, Jacob will spend a year continuing his research in Dr. Claussen's lab, working towards a Master's degree in mechanical engineering. He then hopes to pursue an MD/Ph.D. in biomedical engineering.



## PAST STUPKA SCHOLARS

<b>Claire Kruesel</b> B.S. Biochemistry, Genetics M.F.A. Creative Writing Asst. Teaching Professor, ISU	2006	<b>Denis Tamiev</b> B.S. Biochemistry Ph.D. candidate biochemistry, ISU Tech start-up company	2014
<b>Mara Determan Aleexv</b> M.D., University of Iowa M.P.H., UC-Berkley Clinical Informatics Fellowship, Boston Children's Hospital	2007	<b>Zachary Young</b> B.S. Biochemistry, Mechanical Eng. Senior Researcher, OK Medical Research	2014
<b>Luke Helgeson</b> Ph.D., University of Oregon Post-doc, University of Washington	2008	<b>Flora Yen Wheat</b> D.D.S., 2020 University of Iowa General Dentist, Ankeny Smiles	2015
<b>Mina Farahbakhsh</b> Ph.D. M.D. University of Kansas Pediatric Residency Children's Mercy Hospital, MO	2009	<b>Adrienne Smith</b> B.S. 2016, in memoriam	15,16
<b>Dayna Peterson Forbrook</b> Ph.D., Arizona State University	2010	<b>Morgan Barrett</b> B.S. Biochemistry, Genetics M.S. Forensics Science University of New Haven DNA criminalist, MO State Highway Patrol	2016
<b>Jacqueline Souleyrette Rivas</b> Ph.D., University of Texas S.W. Post-Doc, Markey Cancer Center University of Kentucky	2011	<b>Natalie Whitis</b> B.S. Biophysics Ph.D. candidate, UCSF	2016
<b>Craig Brown</b> M.D., University of Chicago 5th year General Surgery Resident University of Michigan	2011	<b>Lauran Chambers</b> B.S. Biochemistry 1st year medical school A.T. Still University of Osteopathic Medicine	2017
<b>Johanna Jass Bailey</b> B.S. Biochemistry M.P.H., University of Missouri Associate Research Scientist Site Manager, Elemental Enzymes, FL	2011	<b>Alexander Donelson</b> B.S. Biochemistry Pursuing graduate training	2017
<b>Mollie Tiernan Schubert</b> B.S., M.S Biochemistry, ISU Staff Scientist Integrated DNA Technologies, IA	2011	<b>Drew Tonsager</b> B.S. Biochemistry, ISU Ph.D. candidate, Colorado State University	2017
<b>Samson Condon</b> Ph.D. 2020 UW-Madison Post-doc, UW-Madison	2012	<b>Matthew Cook</b> B.S. Biochemistry Ph.D. candidate, Yale University	2018
<b>Alana Jackson</b> M.D. University Minnesota-Duluth Resident Physician - Rural Medicine Spokane Teaching Health Ctr.	2012	<b>Bailey Mooney</b> B.S. Biochemistry, Genetics 2nd year medical school, UCLA	2019
<b>Kristen McKibben</b> Ph.D. 2020, University Pennsylvania School of Medicine Post-Doc, Switzerland	2013	<b>Emily Knuth</b> B.S. Agricultural Biochemistry 2nd year graduate training, UW-Madison	2019
<b>Kinsey Cornick</b> D.O., Des Moines University 3rd yr Family Medicine Resident Lincoln Family Medical Group, NE	2013	<b>Sarah Zelle</b> B.S. Biochemistry, Genetics 1st year graduate training, Vanderbilt	2020
<b>Jennifer Kaczynski Meyer</b> B.S. Biochemistry, Genetics M.S. S21 Genetic Counseling, UW-Madison Genetic Counselor, University of Minnesota Maternal Fetal Medicine	2013	<b>Jacqueline Ehrlich</b> B.S. Agricultural Biochemistry 1st year graduate training Cornell University	2020
		<b>Spydel Nardy</b> B.S. Biochemistry 1st year graduate training University of Iowa	2020

## REMEMBERING ADRIENNE



On February 27, 2017, we lost a very special member of our BBMB family, Adrienne Leah Smith. In 2011, she was diagnosed with Hodgkin's lymphoma. She never let cancer limit or define her; most never knew of her battle. In the fall of 2013, Adrienne joined BBMB as a freshman and immediately got involved with the undergraduate club, Stupka Planning Committee, and of course, research! She worked with Dr. Olga Zabolotina and was preparing for graduate training. Adrienne loved research, and in 2015 and 2016 she was recognized as a Stupka Scholar, BBMB's most prestigious undergraduate award. In 2016, she was a featured student speaker for the Stupka Undergraduate Research Symposium. In the fall of 2016, Adrienne earned her B.S. in biochemistry and graduated summa cum laude. She was a remarkable and brilliant young woman and is greatly missed. In her honor and with her blessing, the 2017 Stupka Planning Committee dedicated a new feature for the symposium, the Adrienne Smith Alumni Interaction Event.

The Adrienne Smith Alumni Interaction Event is yet another unique and valuable opportunity for biochemistry students to prepare for their post graduate plans. Conversations with alumni not only expand their professional network, but also create lasting friendships. The event is a beautiful tribute to Adrienne, as she loved serving on the Stupka planning committee and studying biochemistry. Adrienne will be with us forever and continue to inspire us all.



# THANK YOU!

Your generous support makes the 2021  
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